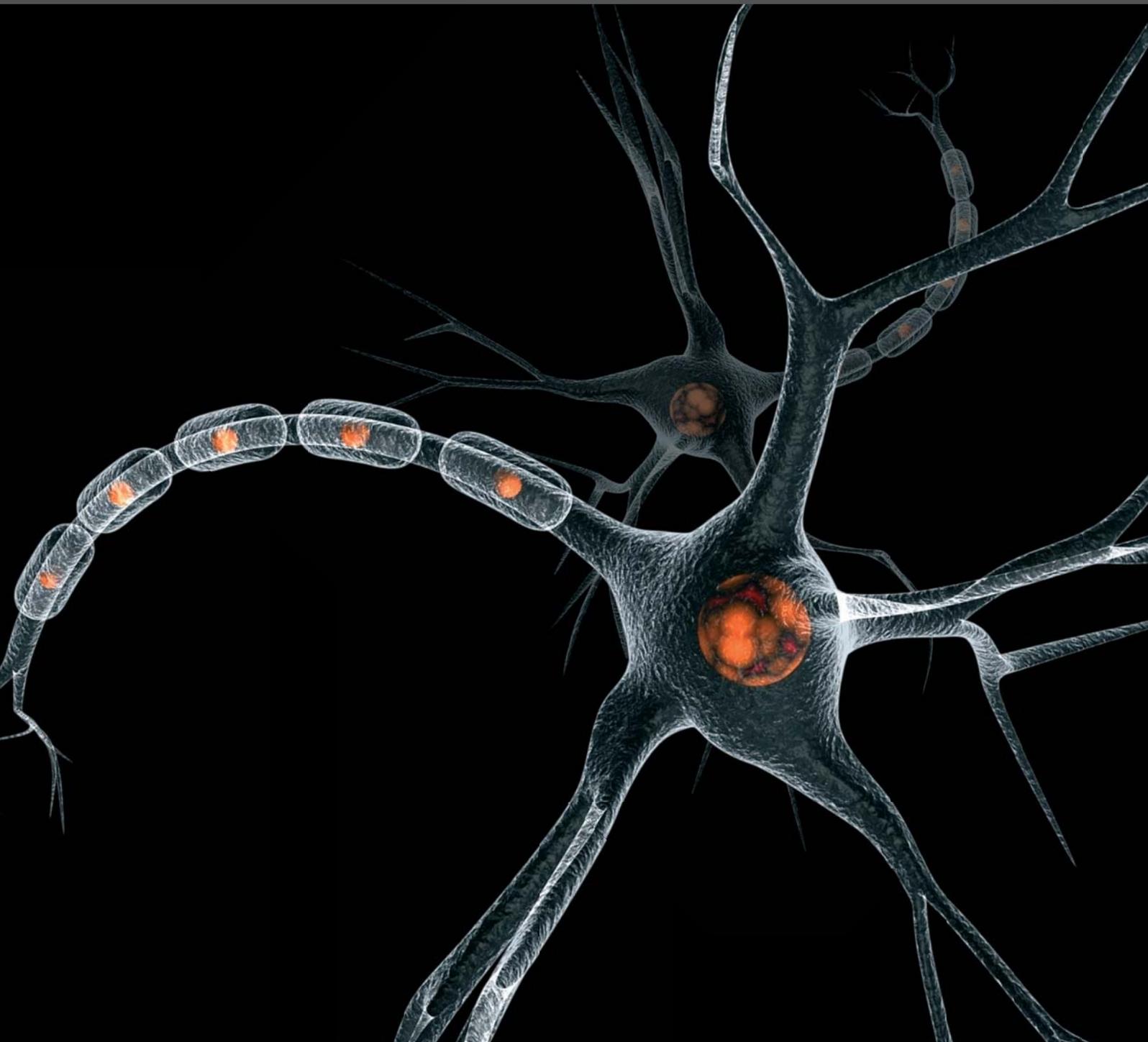


Neural Plasticity

Sensory Deprivation and Brain Plasticity

Guest Editors: Maurice Ptito, Ron Kupers, Steve Lomber,
and Pietro Pietrini





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Editorial

Sensory Deprivation and Brain Plasticity

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Received 9 September 2012; Accepted 9 September 2012

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Early experience plays a key role in the development of the central nervous system as the brain has to adapt continuously to environmental changes that threaten its harmonious development. This adaptation phenomenon is called brain plasticity and refers to the lifelong changes in the structure of the brain that accompany experience (experience-dependent plasticity). Brain plasticity implies that the brain is pliable and can be remodeled in response to challenges that interfere with its normal development such as sensory deprivation, brain injury, or abnormal development. Indeed, several regimens of visual deprivation such as dark rearing, enucleation, or eye-lid suturing lead to alterations of the visual system. Conversely, environmental enrichment also produces regional changes in brain anatomy such as increased dendritic space for synapses, increased cortical thickness, and elevated gene expression.

Sensory deprivation at birth or perinatally has dramatic effects on the organization of the brain circuitry, leading to a takeover of the deprived cortex by other sensory modalities. This phenomenon is known as cross-modal plasticity and is mediated by the development of new inter-modal connections or by strengthening or unmasking of already existing connections. Cross-modal plasticity has been demonstrated following visual and auditory deprivation since birth. In both cases, the deprived cortex becomes activated by one or more of the remaining senses, touch, audition, and olfaction in the case of blindness and visual stimulation in the case of deafness. There is a large body of evidence, based upon anatomical, metabolic, and behavioral findings, that the deprived sensory cortex acquires multiple sensory and

cognitive functions, transforming itself therefore into a multimodal cortex.

In this issue, we have gathered a number of review and original papers on the topic of brain plasticity in humans and in animal models of unimodal sensory deprivation. We have welcomed not only papers dealing with various sensory systems such as vision, audition, touch, and olfaction but also those dealing with basic mechanisms of brain plasticity. Several papers present results on cross-modal plasticity and sensory substitution in humans and in animal models of sensory deprivation. Using elegant methodologies, these studies highlight how other sensory modalities take over the deprived visual or auditory cortices in blind and deaf subjects, respectively. Brain plasticity can also be triggered by changes in sensory experience. Neural reorganization will take place if the environment is modified during the early stages of development as is the case following cortical lesions of the visual cortical areas, visual deprivation through eyelid suturing, or dark-rearing (M. W. Burke et al.; H. Bengoetxea et al.). As shown by J. F. Maya-Vetencourt and N. Origlia, visual cortex plasticity results from a complex interplay between the individual's genetic background and the environment. M. Pietrasanta and coworkers describe how the interhemispheric connectivity between the visual cortices can be altered by visual deprivation. The corpus callosum therefore seems to have a pivotal role in plasticity of the visual cortex. The papers by G. Dormal and colleagues and M. Ptito and colleagues provide evidence that the functional segregation of the efferent projections of the primary visual cortex into a dorsal and a ventral stream is preserved in

congenitally blind humans. A. Leo and colleagues focus on the purported role of the parietooccipital connections in conveying nonvisual information to the deprived visual cortex. Two papers focus on auditory deprivation and cross-modal plasticity. M. A. Meredith and B. L. Allman show how the auditory cortex of deaf ferrets is reorganized by the somatosensory modality, whereas P. Hirst and colleagues use a primate model to highlight cross-modal plasticity. D. M. Coppola describes a model of olfactory system plasticity through unilateral naris occlusion. The author points to the role of the spared ipsilateral olfactory pathway in olfactory plastic responses and also discusses several methodological caveats.

The papers by K. Lehmann and colleagues and S. Desgent and M. Ptito address the role of GABAergic inhibitory interneurons in brain maturation and brain plasticity. These papers describe the important role of GABAergic inhibitory interneurons in maintaining the appropriate dynamic range of cortical excitation, in establishing critical periods of developmental plasticity, in receptive field refinement, and in the cortical processing of sensory information. This interneuron population is very sensitive to sensory experience during development, and visual deprivation studies emphasize the important contribution of these inhibitory interneurons in cross-modal plasticity.

We hope that this special issue will shed light on the various mechanisms involved in cross-modal plasticity and sensory substitution and contribute to the development of new tools that will boost training-induced plasticity.

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Review Article

Plasticity of the Dorsal “Spatial” Stream in Visually Deprived Individuals

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Received 16 December 2011; Accepted 6 June 2012

Academic Editor: Pietro Pietrini

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Studies on visually deprived individuals provide one of the most striking demonstrations that the brain is highly plastic and is able to rewire as a function of the sensory input it receives from the environment. In the current paper, we focus on spatial abilities that are typically related to the dorsal visual pathway (i.e., spatial/motion processing). Bringing together evidence from cataract-reversal individuals, early- and late-blind individuals and sight-recovery cases of long-standing blindness, we suggest that the dorsal “spatial” pathway is mostly plastic early in life and is then more resistant to subsequent experience once it is set, highlighting some limits of neuroplasticity.

1. Introduction

Increasing evidence of experience-based plasticity have challenged the archaic view of the brain as being hard-wired at birth. One of the most striking examples comes from studies on sensory-deprived individuals, documenting that brain regions deprived of their “natural” sensory inputs (i.e., V1 for vision or A1 for audition) in the blind and the deaf brain become responsive to the remaining modalities, a phenomenon that is referred to as *crossmodal plasticity* [1]. Importantly, such plasticity is not restricted to an early sensitive period in life, but rather appears to extent into adulthood. In the case of blindness for instance, individuals who lose vision after the full development of the visual system also present auditory and tactile responses in the deprived occipital regions [2–6].

In the sighted brain, the existence of separate hierarchical pathways for object identification (the ventral “what” stream) and object localization/grasping in space (the dorsal “where” stream) appears as a general principle of organization of the visual cortices [7, 8]. Crucially, recent studies on crossmodal reorganization in the blind suggest that the crossmodal recruitment of the visually deprived cortices in this population might follow organizational principles that

maintain this dual stream segregation ([9], for reviews see [10–12]).

In the present review, we focus on the “dorsal stream” and bring together existing evidence suggesting that this stream is, on the one hand, highly plastic in early life and, on the other hand, very resistant to subsequent experience once it is set. Evidence comes from studies on three different visually deprived populations, that is, cataract-reversal patients, early- and late-blind individuals, and rare cases of sight-recovery individuals after long-standing blindness.

2. Cataract-Reversal Patients

Studies of individuals who were visually deprived early during development, because they were born or developed dense bilateral cataracts which were then surgically removed, represent a unique model to test the role of early visual experience in shaping the functional architecture of the brain. Such studies have documented the existence of different sensitive periods during which visual inputs are necessary for the normal development of different aspects of vision [13]. Specifically, global motion perception, a visual function associated to bilateral occipitoparietal regions in the dorsal visual stream [14–16], and which allows to

integrate local motion information from V1 into a global representation of motion, appears to be permanently altered in cases where vision is absent at birth [17]. However, this function is preserved in cases where the loss of sight occurs after a few months of age [17]. For instance, Ellemberg and colleagues tested global motion perception abilities in a group of bilateral cataract-reversal patients that had been visually deprived at birth and for a period lasting from 3 to 8 months (i.e., congenital group) or in a separate group that had been visually deprived between 8 and 57 months of age, after a period of normal visual experience (i.e., developmental group). The congenital group was found to be strongly impaired compared to the developmental group (Figure 1(b)). In the latter group, all participants performed within normal limits, even in cases where vision had been lost as young as 8 months of age and for a period lasting up to 6 months.

These observations suggest that visual deprivation during the first months of life is sufficient to permanently alter global motion perception abilities, whereas visual experience before the age of approximately 8 months may be necessary and sufficient for the normal development of sensitivity to global motion in humans. Consistent with these findings, kittens raised in the dark up to 4 to 10 weeks of age, showed profound and long-lasting deficits in global motion perception following the period of deprivation, provided it started before 6 weeks of age [18]. Interestingly, the same animals did not show apparent deficits in the perception of simple unidirectional motion, arguing for a larger impact of early visual deprivation on extrastriate than on early visual cortices ([18, 19], but see [20, 21]).

Importantly, studies on the normal development of global motion processing, which is usually considered as an indicator of the dorsal stream maturation [22] suggest that sensitivity to global motion emerges between 6 to 11 weeks of age [23, 24] but reaches adult-like level of performance later in life, although the age of mature performance remains unclear. Some have reported that global motion perception is mature before 3 years of age [25], others between 6 and 11 years of age [17, 26, 27]. A recent study pushes the age of maturity for sensitivity to global motion even later in life, reporting that a group of adults performed equally well than a group of children aged 12 to 14 years old, but significantly better than groups of children aged 6 to 8 years old and 9 to 11 years old [28]. Such discrepancies have been attributed to the different stimuli parameters used in the different studies, such as the dot speed and the dot density [26, 29].

Combining results in cataract-reversal individuals and normally developed individuals seem to indicate that global motion perception can reach adult-like levels of maturity, despite a period of deprivation occurring when the function is not fully developed yet, provided the individual has experienced normal visual input during the early sensitive period starting sometime around birth and lasting within the first year of age [17, 30]. As such, global motion perception is a compelling example of what Maurer and colleagues refer to as a “sleeper effect,” where early visual experience sets up the neural architecture for later normal development [31]. Interestingly, whereas sleeper effects have been documented

in this population for other aspects of low- and high-level vision such as grating acuity, contrast sensitivity, and holistic face processing, some aspects of vision commonly associated to the ventral visual pathway do not necessitate early visual input in order to develop normally. For example, specific aspects of face perception such as face discrimination based on the overall contour of the face, face discrimination based on the shape of internal features, facial expression discrimination, eye gaze, and lip reading perception are all abilities that appear to be preserved even in the absence of early visual input [32–35].

Overall, studies on cataract-reversal individuals constitute a first type of evidence suggesting that sensitivity to global motion, a function related to the dorsal visual pathway, sets up very early in life and is then resistant to subsequent experience.

3. Visually Deprived Individuals

Longstanding blindness is another fascinating model to investigate the role of visual experience on brain development. To date, a wealth of studies have documented that visual deprivation leads the visually deprived occipital regions to massively respond to auditory and tactile inputs (for reviews, see [1, 36]). Comparing the profile of blind individuals who were born as such to those who lost sight later in life after several years of functional vision, further allows questioning the role of *early* visual experience and the role of *the total duration of visual experience* in building the functional architecture of specific brain structures. Beyond the general dual-stream organization of the occipital cortex in sighted individuals, a further segregation concerns its subdivision into several functional areas or “modules,” each of which is specialized for a particular aspect of vision. Within these modules, extrastriate dorsal regions, such as hMT+/V5 and hV3d/V3A, have been extensively described as underlying motion perception in the visual modality [14–16]. Interestingly, in blind individuals who have lost vision at birth or soon after birth, the putative homolog of these regions show responses to motion albeit in the auditory [37–40] and in the tactile [41–43] modalities (Table 1). Moreover, activation in response to auditory motion in putative homolog of area hMT+/V5 bilaterally in blind individuals reflects the direction of perceived moving sounds [40], a property that is known to characterize these regions in the sighted brain for visually moving stimuli [44]. Such results further account for the fact that crossmodal activations in response to auditory dynamic stimulation in these regions subserve a functional role in nonvisual motion processing rather than representing unspecific activation [40]. Therefore, these studies have accounted for the idea that crossmodal plasticity associated to sensory deprivation is functionally specific, in the sense that the mapping of auditory and tactile functions onto visually deprived cortices in the early blind brain appears to follow the natural organization of such regions in the sighted brain [10, 11].

In the same vein, several studies using different paradigms and neuroimaging techniques have consistently demonstrated that spatial hearing in the early blind leads

TABLE 1: Summarizing table of brain coordinates (in MNI space) reported in PET and fMRI studies that investigated nonvisual spatial/motion processing in early blind individuals.

Study	Task	Coordinates in MNI space
Striem-Amit et al., [9]	Location and form identification using a visual-to-auditory sensory substitution device (vOICE).	[Location > Form] × [Blind > Sighted]—whole brain No activated clusters. [Location > Form] in Blind—whole brain No activated clusters in occipital and occipitotemporal cortices.
Bedny et al., [37]	Direction of motion judgment performed on receding and approaching moving sounds.	[High > Low motion] × [Blind > Sighted]—whole brain L inferior temporal gyrus [−44, −72, −6] L middle occipital gyrus [−38, −68, 2]
Bonino et al., [41]	One-back spatial discrimination task performed on 2- and 3-dimensional tactile matrices.	[2D > 3D] in Blind—whole brain (only occipital/occipitotemporal activations are reported) L cuneus [−19, −97, −3] L middle occipital gyrus [−51, −75, 25] L middle occipital gyrus [−35, −90, 11] R middle occipital gyrus [38, −85, 1] R middle occipital gyrus [31, −87, 27] L lateral occipital [−40, −60, −20]
Collignon et al., [38]	Spatial discrimination versus pitch discrimination of pairs of sounds.	[Spatial > Pitch] × [Blind > Sighted]—whole brain R cuneus hV3/V3A [12, −80, 22] R middle occipital gyrus hMT+/V5 [48, −76, 6] R middle occipitotemporal gyrus hMT+/V5 [40, −56, 12] R Lingual gyrus [24, −48, −8]
Gougoux et al., [45]	Binaural and monaural sound localization.	[Monaural localization > Control] × [Blind with superior performance > Sighted]—whole brain R cuneus [13, −81, 15] R lingual gyrus [15, −73, −6] L cuneus [−13, −79, 9]
Matteau et al., [43]	Motion detection task performed on moving versus static tactile stimuli delivered to the tongue using a sensory substitution device, the tongue display unit (TDU).	[Motion > Static] in blind—whole brain analyses (only occipital/occipitotemporal activations are reported) R middle occipital gyrus [20, −88, 22] R middle temporal gyrus hMT+/V5 [42, −54, −4] L middle temporal gyrus hMT+/V5 [−44, −64, 2]
Poirier et al., [39]	Motion detection task performed on horizontally moving sounds versus static sounds presented at different locations.	[Motion > Static] × [Blind > Sighted]—ROIs analyses R cuneus V3/V3A [24, −88, 10] L cuneus V3/V3A [−14, −84, 38] R V1/V2 [2, −82, −8] L V1/V2 [−24, −88, −8] [Motion > Static] in blind (= Sighted)—ROIs analyses R Inferior temporal gyrus hMT+/V5 [44, −72, −2]
Renier et al., [46]	One-back spatial versus frequency discrimination task performed on auditory and tactile stimuli.	[Spatial > Frequency] in Blind—whole brain—(only occipital activations are reported) R middle occipital gyrus [51, −66, −10]
Ricciardi et al., [42]	Passive tactile perception of moving versus static Braille-like dot patterns.	[Motion > Static tactile] in Blind—whole brain—(only occipital/occipitotemporal activations are reported) R hMT+/V5 [38, −69, 7] L hMT+/V5 [−45, −82, 5] R LOC/LOtv [37, −55, −10] L LOC/LOtv [−51, −65, −9] V1/V2 [0, −92, −2] R V1/V2 [37, −88, −4] R cuneus V3A [8, −98, 20]

TABLE 1: Continued.

Study	Task	Coordinates in MNI space
Weeks et al., [47]	Auditory spatial localization task.	[Localization > Rest] \times [Blind > Sighted]—whole brain R superior occipital gyrus [22, -79, 22] R inferior occipital gyrus [40, -70, -9]
		[Moving > Static sounds] in Blind—whole brain (only individual data in occipitotemporal cortices are reported)
Wolbers et al., [40]	Deviant target detection task within blocks of horizontally moving versus static sounds.	Blind 1 [-39, -61, 7]; [-54, -64, -2]; [39, -55, 10]; [42, -64, 10]
		Blind 2 [-39, -67, 4]; [-42, -70, 16]; [45, -64, 10]
		Blind 3 [-33, -76, 19]; [-48, -76, 13]; [39, -55, 10]; [45, -73, 16]
		Blind 4 [-48, -73, 13]; [-42, -61, 13]; [42, -64, 1]

to dorsal occipital recruitment, mainly in the right, spatially dominant, hemisphere (Table 1). In a pioneer PET study, Weeks and colleagues showed that sound localization strongly activated association areas in the right dorsal occipital cortex of early blind individuals but not sighted controls [47]. Another PET study extended these findings, identifying a network of regions in the right dorsal extrastriate cortex that was activated when early blind individuals, but not sighted controls, performed a monaural sound localization task [45]. Further, the functional relevance of such recruitment was ascertained by the fact that several foci of this network correlated with sound localization performance: the blind individuals with the highest performance were the ones who activated these regions the most [45]. Another study suggested that specific recruitment of right dorsal occipital regions in early blind individuals might be present for spatial processing not only of auditory but also of tactile inputs [46].

In a recent fMRI study, we characterized brain activity in early blind and sighted individuals while they were performing discrimination tasks on pairs of sounds differing either in terms of location in space or in pitch [38]. In this study, a staircase paradigm was used in order to equalize difficulty levels across tasks and participants. The spatial localization task relative to the pitch discrimination task was shown to preferentially map onto specialized subregions of the right dorsal occipital stream in the early blind group but not in the sighted group (Figure 3(a)). The two mainly recruited regions were the right cuneus and the right middle occipital gyrus, in the vicinity of regions corresponding to the dorsal hV3d/V3A and hMT+/V5 in the sighted (Figure 3(a)). Interestingly, these two regions have been extensively described as subserving visuo-spatial and motion processing in the sighted brain [14–16, 48]. Although the task involved auditory localization rather than motion processing, we hypothesize that hMT+/V5 was activated because the task generated a vivid perception of apparent motion [38]. Functional connectivity analyses demonstrated that these occipital regions are part of a larger parietofrontal network including multisensory regions (i.e., the inferior parietal lobules, the intraparietal sulcus and the superior frontal gyrus) that are typically involved in spatial attention and

awareness in the sighted brain [49] (Figure 3(a)). Hence, in the absence of visual experience since birth, crossmodal reorganization might be constrained to regions characterized by the same functional specificity, accounting for the fact that these dorsal occipital regions are strongly connected to an extended brain network wired to serve a specific function.

Transcranial magnetic stimulation (TMS) studies further account for the functional relevance of the right dorsal occipital recruitment observed for spatial hearing in the early blind [50, 51]. In an offline TMS paradigm, stimulation applied over these regions led to subsequent alteration of performance in an auditory spatial localization task in early blind but not in sighted individuals, whereas performance in the pitch and intensity discrimination tasks remained unaffected in either group [51] (Figure 3(b)). Most interestingly, the detrimental effect of TMS in the early blind group during the spatial localization task was massively driven by a disruption in the ability of blind individuals to locate sounds presented at the closest position relative to the reference sound in the contralateral (i.e., left) field relative to the right-sided site of stimulation (Figure 3(b), right bottom histogram). This is highly consistent with evidence from the sighted literature documenting a contralateral field preference in several visual areas along the dorsal pathway including hV3d/V3A and hMT+/V5 [15, 48, 52–54]. These results further stress the idea that crossmodal recruitment of the dorsal stream in early blind individuals is functionally relevant and somehow follows the same computational constraints as those observed in sighted individuals when they process visual inputs.

The existence of a critical period in order for sound processing to lead to specific crossmodal recruitment of the dorsal visual pathway was recently suggested. For instance, Bedny and collaborators defined bilateral hMT+/V5 by means of a classical visual localizer in a group of sighted individuals (Figure 2(a), white) and demonstrated that these regions responded to moving sounds in a group of congenitally blind individuals but not in the sighted control group (Figures 2(a) and 2(b), red color), neither in a group of five late blind individuals who lost sight after 9 years of age (range: 9 to 34), (Figure 2(b), blue color) [37]. Interestingly,

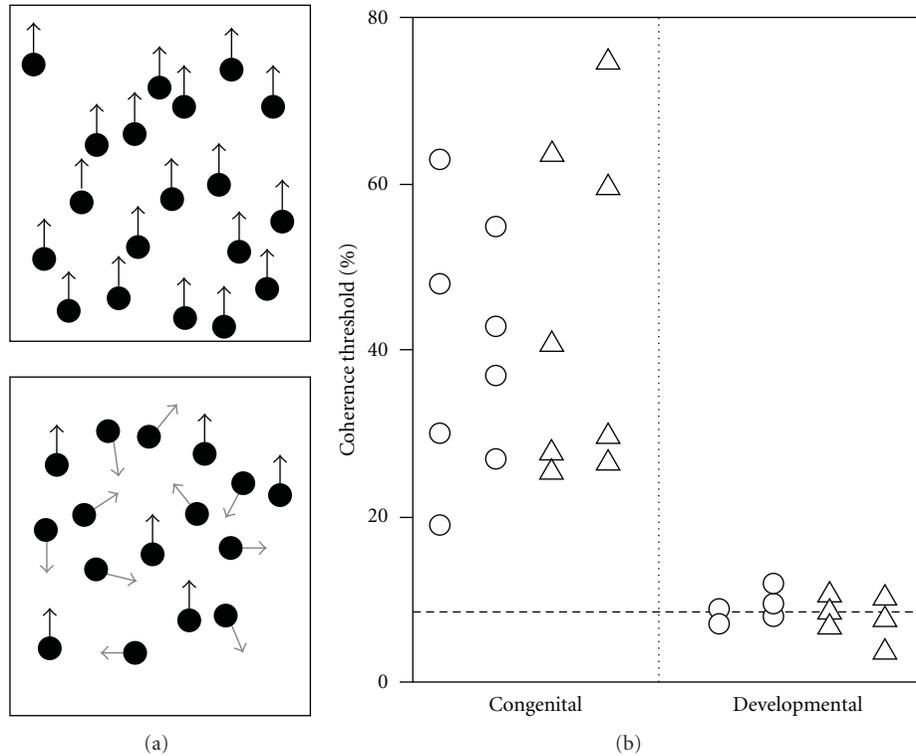


FIGURE 1: (a) Schematic representation of stimuli used to test the coherence threshold, a typical measure of sensitivity to global motion. Among randomly moving dots, the coherence threshold is the minimal percentage of dots moving in the same direction needed for the participant to accurately perceive this predominant direction of motion. Upper-panel represents a trial with 100% coherence as all the dots are moving in the upward direction. Bottom-panel represents a trial with 37% coherence, as 6 out of 16 dots are moving upward whereas the remaining 10 dots are moving in random directions. (b) Global motion coherence thresholds for each subject in the bilateral congenital and bilateral developmental groups tested in the study of Ellemberg et al. [17]. Circles represent the data from the better eyes and triangles represent the data from the worse eyes. The dashed line represents the mean of 24 sighted control subjects. Adapted with permission from [13, 17].

these regions did not present any response to moving sounds in an early-blind individual who had functional vision until he lost it between 2 and 3 years of age (Figure 2(b), black color). In this latter individual, activation evoked by moving sounds in hMT+/V5 was not higher than in any participant in the late blind group and was lower than in each of the participants in the congenitally blind group. Importantly, the total years of blindness in the late blind participants did not predict the amount of response to auditory motion in this region. It is, however, important to note that in this study, auditory motion stimuli, consisting of footsteps (i.e., high motion condition) or tones (i.e., low motion condition) differing in many low-level properties, were either compared to one another, or compared to a rest condition (i.e., scanner noise). Hence, the specificity of hMT+/V5 activity to auditory motion “*per se*” (rather than to the complexity of the sounds) cannot be ascertained. We recently tested late blind individuals who lost sight after 7 to 51 years of functional vision using the staircase paradigm described above [55]. Whereas massive crossmodal recruitment of occipital cortex to auditory processing, irrespective of the task at hand, was found in late blind individuals, the regions in the right dorsal stream that were preferentially activated for the spatial processing of sounds in congenitally blind

individuals did not show any functional preference in blind individuals who lost sight later in life [55].

Taken as a whole, these observations are suggestive of an early sensitive period during which the absence of visual input drives dorsal regions to develop specific cross-modal responses to spatial/motion cues, whereas normal visual input prevents the development of such crossmodal responses, despite years of blindness.

4. Sight-Recovery Individuals

Similar conclusions can be raised from rare cases of sight-recovery individuals after longstanding blindness. Two of such cases, SB and MM, have been quite extensively described in the scientific literature. SB lost effective sight at 10 months of age and received a corneal graft after fifty years as a blind person [56, 57]. MM was blind since the age of 3 years old and received stem-cell transplant in his right eye at the age of 46 [58–60]. SB and MM presented striking similarities in their visual abilities following sight restoration. Despite the fact that their retinas regained some functionality, they both encountered extreme difficulties interpreting what they saw, suggesting these deficiencies were from central rather than from peripheral origins. Although they could recognize colors and simple shapes

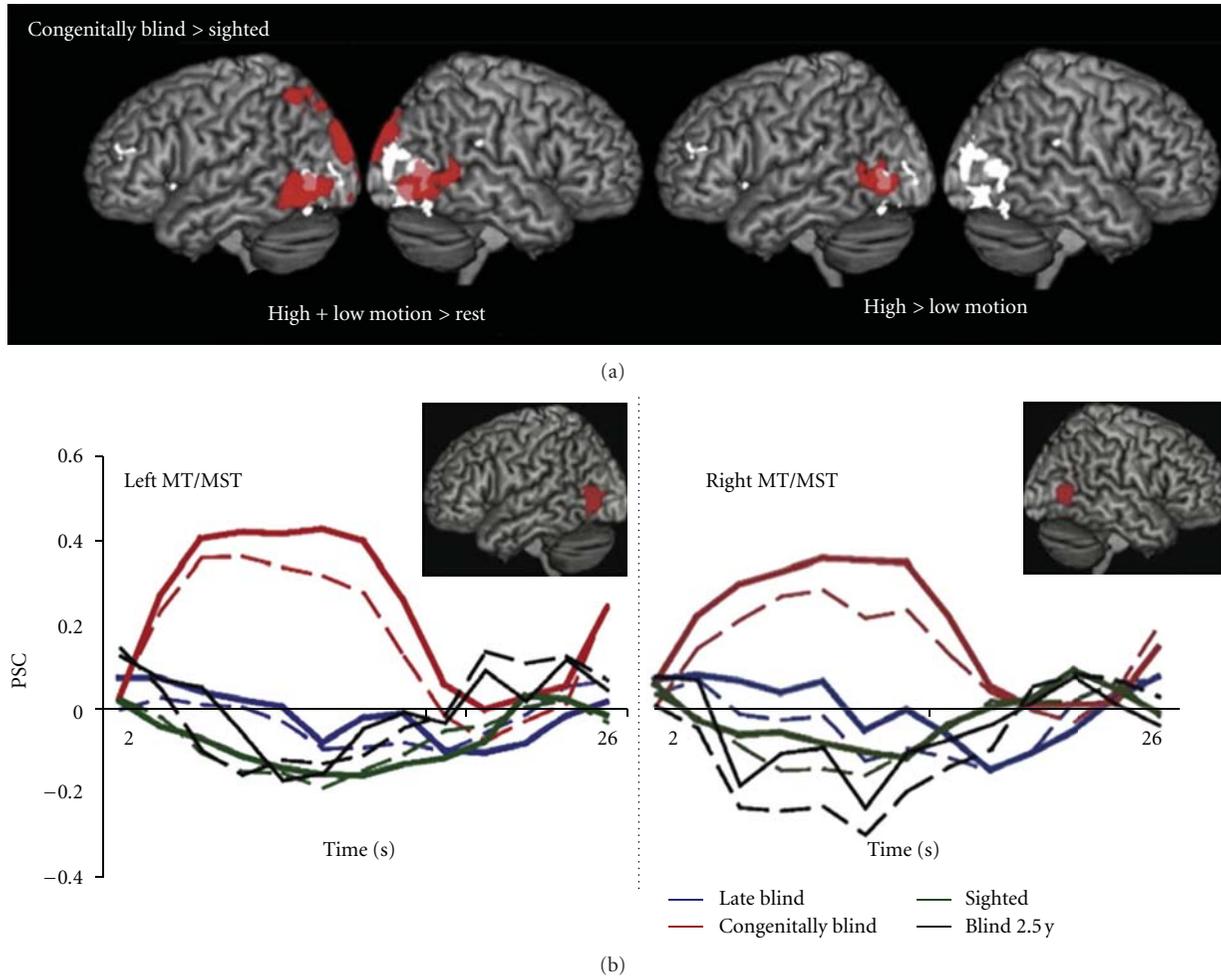


FIGURE 2: (a) Red colored regions denote activations obtained in the study of Bedny et al. [37] from the contrast testing which regions show greater BOLD signal in response to auditory motion in the congenitally blind relative to the sighted, in high and low motion conditions relative to rest (left panel) and in the high motion condition relative to the low motion condition (right panel) ($P < 0.05$, corrected). White colored regions are activated in a sighted group of controls when viewing moving relative to stationary dots. The overlap between regions activated during auditory motion perception in the congenitally blind relative to the sighted, and the ones activated during visual motion perception in the sighted are colored in pink. (b) Percent signal change relative to baseline as a function of time (seconds) in response to auditory motion is displayed in left and right MT/MST ROIs (overlaid in red on a normalized template (left is left) and identified by means of a visual motion localizer in sighted participants) for sighted, congenitally blind, and late blind participants. Solid lines represent percent signal change in response to the high motion condition (footsteps) and dashed lines represent percent signal change in response to the low motion condition (tones). Adapted with permission from [37].

quite accurately, recognition of complex shapes, including faces and everyday life objects, perception of depth cues as well as detection of illusory contours were all abilities that were highly altered. In MM, these visual deficits were further accounted by neuroimaging evidence showing a massive reduction of activation to faces and objects in the fusiform and lingual gyri bilaterally (i.e., the brain areas usually devoted to object and face perception) [58]. Seven years after the intervention, he still had poor spatial resolution and limited visual abilities that prevented him from efficiently relying on his vision in every day life [59, 60].

In contrast to these marked difficulties encountered by SB and MM, motion perception abilities appeared to be quite well preserved in both cases despite years of blindness. MM for instance, performed within normal limits in several

motion tasks, whether he had to detect the direction of a moving pattern, or perceive the orientation or the shape of a moving object. Similarly, Gregory and Wallace reported that SB was only able to recognize certain objects in the environment provided they were moving [56, 57]. As such, motion cues constituted information on which these patients could rely more confidently in order to use their newly acquired vision in their day-to-day activities. Consistently with these preserved motion perception abilities, fMRI measures in MM documented normal size of area hMT+/V5 and normal activation in response to moving versus stationary visual stimuli when tested within months following sight restoration [58] as well as 7 years later [60].

Hence, in marked contrast to deficiencies observed in several aspects of vision, the preservation of motion

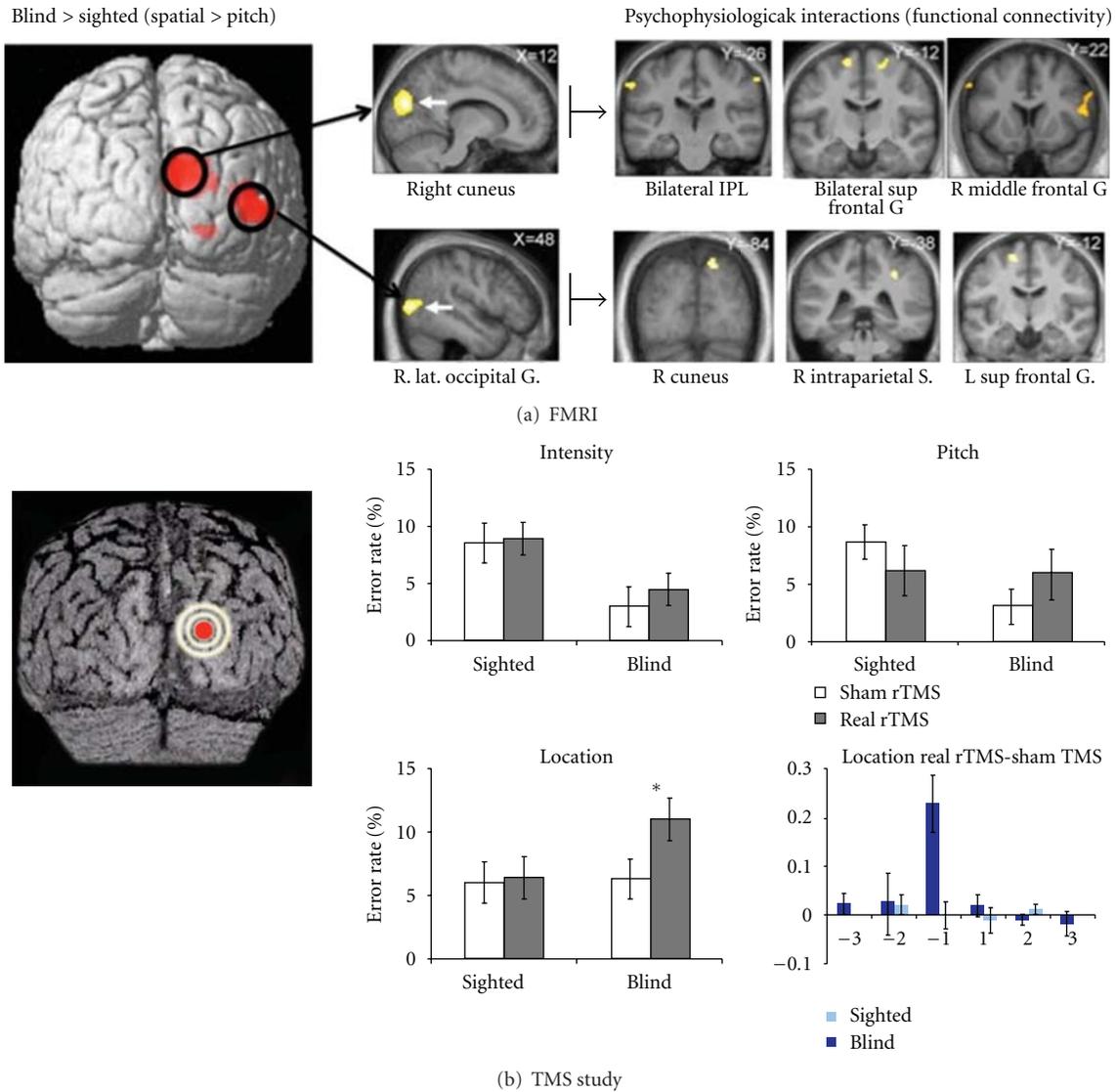


FIGURE 3: (a) The left part of the figure illustrates the activations obtained in the study of Collignon et al. [38] from the contrast testing which regions are specifically dedicated to the spatial processing of sounds in early blind subjects relative to sighted controls: [Blind > Sighted] × [Spatial > Pitch]. Functional data are overlaid (uncorrected $P < 0.001$) over a 3D render of the brain (left is left). The right part of the figure displays psychophysiological interaction results using the two main activity peaks as seed areas. (b) The 3D brain representation (left is left) displays the projection of the site of TMS application in the study of Collignon et al. [51]. This area corresponds to the right dorsal extrastriate occipital cortex (BA 18). The histograms denote the average error rate in early blind and sighted subjects after sham (control) and real rTMS targeting the dorsal occipital stream during auditory tasks involving the discrimination of intensity, pitch and spatial location of sounds. The data show a significant increase of the error rate after real rTMS only in the early blind group and selectively for the sound location task. The histogram on the right bottom of the figure represents the percentage of errors in the spatial location task in early blind and sighted subjects for the real rTMS condition minus the sham TMS condition (isolating the effect of the TMS), as a function of sound position. Negative values on the x -axis are referring to the left external space, positive values on the x -axis are referring to the right external space. Adapted with permission from [38, 51].

perception abilities in both patients accounts for the idea that such abilities might have developed with early visual experience. Moreover, it appears that such abilities do not require prolonged visual experience in order to crystallize, as opposed to more ventral-related visual functions such as face and object perception [13]. However, an alternative account is that such cases possessed residual visual capacities for motion perception during the extended period of visual deprivation. Indeed, the vast majority of operable blind

individuals are cases of “near-blindness” in the sense that, for blindness to be operable, the retina and eye tissues must be at least partially functional [56, 57]. In cases where blindness is strictly total, that is, where both eyes are insensitive to light, the successful outcome of surgery is minimal. Hence, rare cases of sight-recovery individuals are cases that present at least light perception and maybe also crude motion perception in at least one eye. For instance, one of the last medical records of SB before he received corneal grafts

indicated that the left eye was reduced to light perception and showed a normal pattern of retinal vessels, whereas the right eye was capable of perceiving hand movements [56, 57]. Similarly, Ackroyd and colleagues reported that HD, an early-onset blind woman who partially recovered sight at the age of 27, was still capable of perceiving moving shadows during the period she was blind [61]. No such records of medical history previous to surgery are provided in the published reports regarding MM, neither for other more recent cases of sight-recovery individuals following years of congenital blindness [62, 63]. The preserved motion perception abilities observed following years of blindness in such cases might thus be, at least partially, explained by the fact that some dynamic information was still available to them after the onset of visual deprivation [62].

In other words, even a brief period of vision after birth or the maintenance of crude visual abilities in these visually deprived individuals may be sufficient to appropriately tune the motion system. Therefore, in sight-recovery individuals, motion cues are likely to play an important role in rehabilitation, guiding the individual to improve learning of other, more disrupted, visual abilities such as object and face recognition. This is well illustrated by a study of Ostrovsky and colleagues, who reported the cases of three supposed congenital blind individuals whose vision was partially restored after years of blindness [63]. Following cataract removal, similarly to SB and MM, these individuals presented marked difficulties in form and depth perception when looking at static images. However, the introduction of motion cues immediately improved these perception abilities. Further and most importantly, when these subjects were tested several months following partial vision restoration, they could recognize static images that they previously could not recognize unless motion cues were provided. These observations suggest that motion perception abilities, because they were spared as compared to other visual abilities such as form perception, might have guided visual learning for these latter, altered abilities [63].

5. Involvement of the Dorsal Occipital Stream for Nonvisual Spatial Processing in the Sighted

A growing body of evidence suggests that occipital cortices participate in processing information from other nonvisual modalities not only in visually deprived individuals but also, to some extent, in the normal sighted brain. For instance, modulatory effects of auditory or tactile motion on visual hMT+/V5 responses have been previously documented in typically developing individuals as a result of multisensory integration [64, 65]. Beyond multisensory processes, it appears that visual cortices in the sighted brain can also be modulated by nonvisual stimuli presented alone. On the one hand, several studies have documented crossmodal *activation* of extrastriate dorsal visual regions by auditory and tactile motion/spatial information in sighted individuals ([41, 42, 66, 67], for a review see [68]). When similar crossmodal activation of “visual areas” has been observed in

both blind and sighted subjects, some have interpreted it as evidence for the metamodal/supramodal nature of the brain [69, 70]. It should be stressed that similar activations might actually subservise completely different mechanisms in these two populations. For example, recruitment of visual areas during nonvisual processing could be mediated by visual imagery through top-down mechanisms in the sighted brain ([68, 71, 72], for a review, see [73]) whereas it might subservise nonvisual processing *per se* in the blind brain.

On the contrary, other studies have shown that nonvisual processing in the sighted brain is associated to *deactivations* in extrastriate visual regions [74, 75]. In fact, within the studies mentioned in the previous sections of this review, some have reported deactivations in the brain of their sighted control participants in area hMT+/V5 during auditory motion processing (Figure 2(b), green color) [37, 60] and in dorsal extrastriate visual areas during auditory and tactile spatial localization [38, 45, 46]. It is worth noting that such crossmodal deactivations observed in the sighted brain might in fact be task dependent [75]. In our staircase paradigm described earlier on, we found significant positive differences in the vicinity of hMT+/V5 when contrasting the spatial and the pitch discrimination condition (i.e., Spatial > Pitch) in sighted participants [38]. However, when plotting the activity estimated in this region, it appeared that both pitch and spatial processing of sounds deactivated (as compared to baseline) this region in sighted subjects (Figure 4(a)). Because the deactivation was found to be greater in the pitch condition relative to the spatial condition, it led to a positive value when contrasting the two conditions (Figure 4(a)). A recent study reported similar observation in a sighted group of participants when plotting activation estimates in their tactile localization and identification conditions in the right middle occipital gyrus [46]. Interestingly, in our study [38], the region showing spatial specificity in terms of deactivation in sighted participants overlapped with the region showing spatial specificity in terms of activation (i.e., functionally specific crossmodal responses) in our early blind group (Figure 4(a)). The voxels with the highest significant difference between the spatial and the pitch conditions in our sighted group was found to be in close vicinity to the coordinates reported in another study when contrasting a condition of moving sounds to a condition of stationary sounds (Moving > Stationary) in sighted participants [76]. Again, a positive difference was reported in the latter study, but because the mean parameter estimates were not displayed separately for each condition in the sighted participants, such positive difference might also putatively result from less deactivation rather than from more activation in the “spatial” auditory task relative to the “stationary” auditory task [76].

Saenz and colleagues reported specific crossmodal responses to motion in two sight-recovery subjects and not in six control sighted subjects based on the observation of significant positive differences between auditory moving and auditory static sounds in these subjects ([60], Figures 4(b) and 4(c)). Evidence of coexisting specific auditory and visual responses to motion relative to their static version in these regions in patient MM who lost sight at 3 years old, is

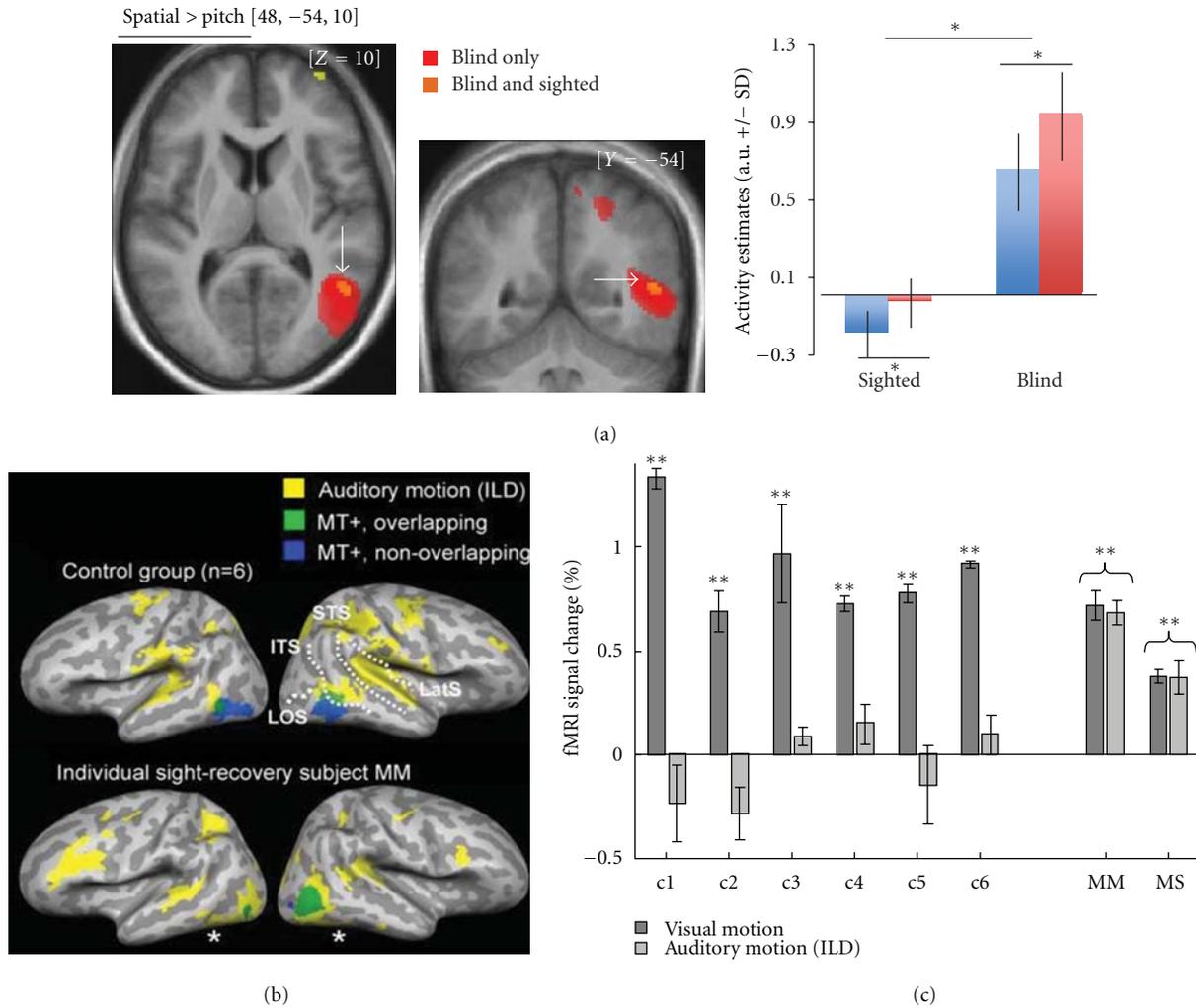


FIGURE 4: (a) The left part of the figure illustrates the activations obtained in the study of Collignon et al. [38] from the contrast testing which regions are more active for the spatial processing rather than the pitch processing of sounds ([Spatial > Pitch]) in early blind subjects only (red), and in both early blind and sighted participants (orange). Functional data are overlaid (uncorrected $P < 0.001$) over a 3D render of the brain (left is left). The right part of the figure shows beta parameter estimates relative to baseline for the spatial and the pitch conditions, in the sighted subjects and in the early blind participants at coordinate [48, -54, 10]. (b) Activations obtained in the study of Saenz et al. [60] in six controls subjects (upper part of the figure) and in sight-recovery individual MM (lower part of the figure). Yellow colored regions show a positive difference when contrasting auditory motion to its static condition. Green and blue regions illustrate the overlap and non-overlap with visually defined MT+ in the same subjects. (c) In the same study, percent signal change in visually defined MT+ ROIs is plotted for moving relative to stationary visual stimuli and for moving relative to stationary auditory stimuli in six control sighted subjects (c1–c6) and in two sight-recovery individuals (MM and MS). Adapted with permission from [60].

contradictive to results reported by Bedny and colleagues where putative homolog of these regions in a blind who lost efficient sight between the ages of 2 and 3 years old did not develop such crossmodal responses. This apparent contradiction was accounted by the possibility of individual differences in the sensitive period for the development of functionally specific crossmodal plasticity [37]. However, because parameter estimates in these regions were reported systematically for the motion (experimental) conditions relative to their respective static (control) conditions (i.e., motion minus static), it is not possible to disentangle whether this positive difference observed for moving sounds relative to stationary sounds is the result of differences in activations or of deactivations [60].

Taking these observations as a whole, it was suggested that both activations and deactivations identified during nonvisual tasks can indicate the presence of nonvisual inputs in the occipital cortex of sighted individuals [77]. In agreement with this, TMS studies have demonstrated that disrupting dorsal extrastriate occipital regions in the sighted brain might impair the processing of auditory spatial information [78] and tactile flow [79, 80]. In the sighted brain, existing cortico-cortical connections between auditory and visual cortices [81–84] might play a role for the integration of spatial information coming from different modalities or for the inhibition of visual cortices during nonvisual tasks in order to minimize potential effect of interference with auditory processing [74, 75]. Understanding how specific

nonvisual tasks may decrease or increase the activity of the occipital cortex in sighted individuals, and how these crossmodal influences relate to the plastic changes observed in early- and late-blind individuals remains one of the most important challenges for future research in the field.

6. Conclusion

Based on the evidence presented here, it could be suggested that early visual experience plays a crucial role for dorsal regions to develop specific visually driven responses to motion/spatial cues, in agreement with the existence of an early and short sensitive period for the normal development of global motion perception in the visual modality. As stressed by studies on cataract-reversal patients, if deprived from normal visual input in the first months after birth, these visual abilities might never develop normally later on [17]. As a direct counterpart, studies on visually deprived individuals have demonstrated that early- but not late-onset blindness drives dorsal regions to develop specific crossmodal responses to spatial/motion cues, maintaining their function for motion/space perception when processing inputs from the remaining modalities [37–43, 45–47]. Finally, the relative preservation of motion perception abilities in sight-recovery patients with functional vision early in life also suggests that such abilities might have developed with early visual experience and do not require prolonged visual experience in order to crystallize [58, 60]. Altogether, these observations are compatible with the idea that the *functional and modality* specificity of the dorsal pathway is set early in development and is quite resistant to later acquired experience.

References

- [1] D. Bavelier and H. J. Neville, “Cross-modal plasticity: where and how?” *Nature Reviews Neuroscience*, vol. 3, no. 6, pp. 443–452, 2002.
- [2] C. Büchel, C. Price, R. S. J. Frackowiak, and K. Friston, “Different activation patterns in the visual cortex of late and congenitally blind subjects,” *Brain*, vol. 121, no. 3, pp. 409–419, 1998.
- [3] H. Burton, “Visual cortex activity in early and late blind people,” *Journal of Neuroscience*, vol. 23, no. 10, pp. 4005–4011, 2003.
- [4] H. Burton, A. Z. Snyder, T. E. Conturo, E. Akbudak, J. M. Ollinger, and M. E. Raichle, “Adaptive changes in early and late blind: a fMRI study of Braille reading,” *Journal of Neurophysiology*, vol. 87, no. 1, pp. 589–607, 2002.
- [5] H. Burton, A. Z. Snyder, J. B. Diamond, and M. E. Raichle, “Adaptive changes in early and late blind: a fMRI study of verb generation to heard nouns,” *Journal of Neurophysiology*, vol. 88, no. 6, pp. 3359–3371, 2002.
- [6] P. Voss, F. Gougoux, R. J. Zatorre, M. Lassonde, and F. Lepore, “Differential occipital responses in early- and late-blind individuals during a sound-source discrimination task,” *NeuroImage*, vol. 40, no. 2, pp. 746–758, 2008.
- [7] J. V. Haxby, C. L. Grady, B. Horwitz et al., “Dissociation of object and spatial visual processing pathways in human extrastriate cortex,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 88, no. 5, pp. 1621–1625, 1991.
- [8] M. A. Goodale and A. David Milner, “Separate visual pathways for perception and action,” *Trends in Neurosciences*, vol. 15, no. 1, pp. 20–25, 1992.
- [9] E. Striem-Amit, O. Dakwar, L. Reich, and A. Amedi, “The large-scale organization of “visual” streams emerges without visual experience,” *Cerebral Cortex*, vol. 22, no. 7, pp. 1698–1709, 2012.
- [10] O. Collignon, P. Voss, M. Lassonde, and F. Lepore, “Cross-modal plasticity for the spatial processing of sounds in visually deprived subjects,” *Experimental Brain Research*, vol. 192, no. 3, pp. 343–358, 2009.
- [11] G. Dormal and O. Collignon, “Functional selectivity in sensory-deprived cortices,” *Journal of Neurophysiology*, vol. 105, no. 6, pp. 2627–2630, 2011.
- [12] P. Voss and R. J. Zatorre, “Organization and reorganization of sensory-deprived cortex,” *Current Biology*, vol. 22, no. 5, pp. R168–R173, 2012.
- [13] D. Maurer, T. L. Lewis, and C. J. Mondloch, “Missing sights: consequences for visual cognitive development,” *Trends in Cognitive Sciences*, vol. 9, no. 3, pp. 144–151, 2005.
- [14] S. Sunaert, P. Van Hecke, G. Marchal, and G. A. Orban, “Motion-responsive regions of the human brain,” *Experimental Brain Research*, vol. 127, no. 4, pp. 355–370, 1999.
- [15] R. B. H. Tootell, J. B. Reppas, K. K. Kwong et al., “Functional analysis of human MT and related visual cortical areas using magnetic resonance imaging,” *Journal of Neuroscience*, vol. 15, no. 4, pp. 3215–3230, 1995.
- [16] J. D. G. Watson, R. Myers, R. S. J. Frackowiak et al., “Area V5 of the human brain: evidence from a combined study using positron emission tomography and magnetic resonance imaging,” *Cerebral Cortex*, vol. 3, no. 2, pp. 79–94, 1993.
- [17] D. Elleberg, T. L. Lewis, D. Maurer, S. Brar, and H. P. Brent, “Better perception of global motion after monocular than after binocular deprivation,” *Vision Research*, vol. 42, no. 2, pp. 169–179, 2002.
- [18] D. E. Mitchell, J. Kennie, and D. Kung, “Development of global motion perception requires postnatal exposure to pattern light,” *Current Biology*, vol. 19, no. 8, pp. 645–649, 2009.
- [19] N. L. Rochefort, M. Narushima, C. Grienberger, N. Marandi, D. N. Hill, and A. Konnerth, “Development of direction selectivity in mouse cortical neurons,” *Neuron*, vol. 71, no. 3, pp. 425–432, 2011.
- [20] Y. Li, D. Fitzpatrick, and L. E. White, “The development of direction selectivity in ferret visual cortex requires early visual experience,” *Nature Neuroscience*, vol. 9, no. 5, pp. 676–681, 2006.
- [21] Y. Li, S. D. Van Hooser, M. Mazurek, L. E. White, and D. Fitzpatrick, “Experience with moving visual stimuli drives the early development of cortical direction selectivity,” *Nature*, vol. 456, no. 7224, pp. 952–956, 2008.
- [22] O. Braddick, J. Atkinson, and J. Wattam-Bell, “Normal and anomalous development of visual motion processing: motion coherence and ‘dorsal-stream vulnerability,’” *Neuropsychologia*, vol. 41, no. 13, pp. 1769–1784, 2003.
- [23] T. Banton, B. I. Bertenthal, and J. Seaks, “Infants’ sensitivity to statistical distributions of motion direction and speed,” *Vision Research*, vol. 39, no. 20, pp. 3417–3430, 1999.
- [24] J. Wattam-Bell, “Coherence thresholds for discrimination of motion direction in infants,” *Vision Research*, vol. 34, no. 7, pp. 877–883, 1994.
- [25] E. E. Parrish, D. E. Giaschi, C. Boden, and R. Dougherty, “The maturation of form and motion perception in school age children,” *Vision Research*, vol. 45, no. 7, pp. 827–837, 2005.

- [26] D. Ellemberg, T. L. Lewis, M. Dirks et al., "Putting order into the development of sensitivity to global motion," *Vision Research*, vol. 44, no. 20, pp. 2403–2411, 2004.
- [27] A. Gunn, E. Cory, J. Atkinson et al., "Dorsal and ventral stream sensitivity in normal development and hemiplegia," *NeuroReport*, vol. 13, no. 6, pp. 843–847, 2002.
- [28] B.-S. Hadad, D. Maurer, and T. L. Lewis, "Long trajectory for the development of sensitivity to global and biological motion," *Developmental Science*, vol. 14, no. 6, pp. 1330–1339, 2011.
- [29] S. Narasimhan and D. Giaschi, "The effect of dot speed and density on the development of global motion perception," *Vision Research*, vol. 62, pp. 102–107, 2012.
- [30] T. L. Lewis and D. Maurer, "Multiple sensitive periods in human visual development: evidence from visually deprived children," *Developmental Psychobiology*, vol. 46, no. 3, pp. 163–183, 2005.
- [31] D. Maurer, C. J. Mondloch, and T. L. Lewis, "Sleeper effects," *Developmental Science*, vol. 10, no. 1, pp. 40–47, 2007.
- [32] S. Geldart, C. J. Mondloch, D. Maurer, S. De Schonen, and H. P. Brent, "The effect of early visual deprivation on the development of face processing," *Developmental Science*, vol. 5, no. 4, pp. 490–501, 2002.
- [33] R. Le Grand, C. J. Mondloch, D. Maurer, and H. P. Brent, "Neuroperception: early visual experience and face processing," *Nature*, vol. 410, no. 6831, p. 890, 2001.
- [34] R. Le Grand, C. J. Mondloch, D. Maurer, and H. P. Brent, "Impairment in holistic face processing following early visual deprivation," *Psychological Science*, vol. 15, no. 11, pp. 762–768, 2004.
- [35] D. Maurer, D. Ellemberg, and T. L. Lewis, "Repeated measurements of contrast sensitivity reveal limits to visual plasticity after early binocular deprivation in humans," *Neuropsychologia*, vol. 44, no. 11, pp. 2104–2112, 2006.
- [36] A. Pascual-Leone, A. Amedi, F. Fregni, and L. B. Merabet, "The plastic human brain cortex," *Annual Review of Neuroscience*, vol. 28, pp. 377–401, 2005.
- [37] M. Bedny, T. Konkle, K. Pelphrey, R. Saxe, and A. Pascual-Leone, "Sensitive period for a multimodal response in human visual motion area MT/MST," *Current Biology*, vol. 20, no. 21, pp. 1900–1906, 2010.
- [38] O. Collignon, G. Vandewalle, P. Voss et al., "Functional specialization for auditory-spatial processing in the occipital cortex of congenitally blind humans," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 11, pp. 4435–4440, 2011.
- [39] C. Poirier, O. Collignon, C. Scheiber et al., "Auditory motion perception activates visual motion areas in early blind subjects," *NeuroImage*, vol. 31, no. 1, pp. 279–285, 2006.
- [40] T. Wolbers, P. Zahorik, and N. A. Giudice, "Decoding the direction of auditory motion in blind humans," *NeuroImage*, vol. 56, no. 2, pp. 681–687, 2011.
- [41] D. Bonino, E. Ricciardi, L. Sani et al., "Tactile spatial working memory activates the dorsal extrastriate cortical pathway in congenitally blind individuals," *Archives Italiennes de Biologie*, vol. 146, no. 3-4, pp. 133–146, 2008.
- [42] E. Ricciardi, N. Vanello, L. Sani et al., "The effect of visual experience on the development of functional architecture in hMT+," *Cerebral Cortex*, vol. 17, no. 12, pp. 2933–2939, 2007.
- [43] I. Matteau, R. Kupers, E. Ricciardi, P. Pietrini, and M. Ptito, "Beyond visual, aural and haptic movement perception: hMT+ is activated by electrotactile motion stimulation of the tongue in sighted and in congenitally blind individuals," *Brain Research Bulletin*, vol. 82, no. 5-6, pp. 264–270, 2010.
- [44] R. T. Born and D. C. Bradley, "Structure and function of visual area MT," *Annual Review of Neuroscience*, vol. 28, pp. 157–189, 2005.
- [45] F. Gougoux, R. J. Zatorre, M. Lassonde, P. Voss, and F. Lepore, "A functional neuroimaging study of sound localization: visual cortex activity predicts performance in early-blind individuals," *PLoS Biology*, vol. 3, no. 2, article e27, 2005.
- [46] L. A. Renier, I. Anurova, A. G. De Volder, S. Carlson, J. VanMeter, and J. P. Rauschecker, "Preserved functional specialization for spatial processing in the middle occipital gyrus of the early blind," *Neuron*, vol. 68, no. 1, pp. 138–148, 2010.
- [47] R. Weeks, B. Horwitz, A. Aziz-Sultan et al., "A positron emission tomographic study of auditory localization in the congenitally blind," *Journal of Neuroscience*, vol. 20, no. 7, pp. 2664–2672, 2000.
- [48] R. B. H. Tootell, J. D. Mendola, N. K. Hadjikhani et al., "Functional analysis of V3A and related areas in human visual cortex," *Journal of Neuroscience*, vol. 17, no. 18, pp. 7060–7078, 1997.
- [49] S. M. Szczepanski, C. S. Konen, and S. Kastner, "Mechanisms of spatial attention control in frontal and parietal cortex," *Journal of Neuroscience*, vol. 30, no. 1, pp. 148–160, 2010.
- [50] O. Collignon, M. Davare, E. Olivier, and A. G. De Volder, "Reorganisation of the right occipito-parietal stream for auditory spatial processing in early blind humans. a transcranial magnetic stimulation study," *Brain Topography*, vol. 21, no. 3-4, pp. 232–240, 2009.
- [51] O. Collignon, M. Lassonde, F. Lepore, D. Bastien, and C. Veraart, "Functional cerebral reorganization for auditory spatial processing and auditory substitution of vision in early blind subjects," *Cerebral Cortex*, vol. 17, no. 2, pp. 457–465, 2007.
- [52] E. Fischer, H. H. Bühlhoff, N. K. Logothetis, and A. Bartels, "Visual motion responses in the posterior cingulate sulcus: a comparison to V5/MT and MST," *Cerebral Cortex*, vol. 22, no. 4, pp. 865–876, 2012.
- [53] J. Larsson and D. J. Heeger, "Two retinotopic visual areas in human lateral occipital cortex," *Journal of Neuroscience*, vol. 26, no. 51, pp. 13128–13142, 2006.
- [54] M. I. Sereno, A. M. Dale, J. B. Reppas et al., "Borders of multiple visual areas in humans revealed by functional magnetic resonance imaging," *Science*, vol. 268, no. 5212, pp. 889–893, 1995.
- [55] O. Collignon, G. Albouy, G. Dormal et al., "Impact of early versus late acquired blindness on the functional organization and connectivity of the occipital cortex," *Journal of Vision*, vol. 12, no. 9, p. 610, 2012.
- [56] R. L. Gregory and J. Wallace, *Recovery from Early Blindness: A Case Study*, Experimental Psychology Society Monograph No. 2, Heffers, Cambridge, UK, 1963.
- [57] R. Gregory, *Concepts and Mechanisms of Perception*, Duckworth, London, 1974.
- [58] I. Fine, A. R. Wade, A. A. Brewer et al., "Long-term deprivation affects visual perception and cortex," *Nature Neuroscience*, vol. 6, no. 9, pp. 915–916, 2003.
- [59] N. Levin, S. O. Dumoulin, J. Winawer, R. F. Dougherty, and B. A. Wandell, "Cortical maps and white matter tracts following long period of visual deprivation and retinal image restoration," *Neuron*, vol. 65, no. 1, pp. 21–31, 2010.
- [60] M. Saenz, L. B. Lewis, A. G. Huth, I. Fine, and C. Koch, "Visual motion area MT + /V5 responds to auditory motion in human sight-recovery subjects," *Journal of Neuroscience*, vol. 28, no. 20, pp. 5141–5148, 2008.

- [61] C. Ackroyd, N. K. Humphrey, and E. K. Warrington, "Lasting effects of early blindness, a case study," *Quarterly Journal of Experimental Psychology*, vol. 26, no. 1, pp. 114–124, 1974.
- [62] Y. Ostrovsky, A. Andalman, and P. Sinha, "Vision following extended congenital blindness," *Psychological Science*, vol. 17, no. 12, pp. 1009–1014, 2006.
- [63] Y. Ostrovsky, E. Meyers, S. Ganesh, U. Mathur, and P. Sinha, "Visual parsing after recovery from blindness," *Psychological Science*, vol. 20, no. 12, pp. 1484–1491, 2009.
- [64] A. Alink, W. Singer, and L. Muckli, "Capture of auditory motion by vision is represented by an activation shift from auditory to visual motion cortex," *Journal of Neuroscience*, vol. 28, no. 11, pp. 2690–2697, 2008.
- [65] S. Sadaghiani, J. X. Maier, and U. Noppeney, "Natural, metaphoric, and linguistic auditory direction signals have distinct influences on visual motion processing," *Journal of Neuroscience*, vol. 29, no. 20, pp. 6490–6499, 2009.
- [66] M. C. Hagen, O. Franzén, F. McGlone, G. Essick, C. Dancer, and J. V. Pardo, "Tactile motion activates the human middle temporal/V5 (MT/V5) complex," *European Journal of Neuroscience*, vol. 16, no. 5, pp. 957–964, 2002.
- [67] E. Ricciardi, D. Bonino, C. Gentili, L. Sani, P. Pietrini, and T. Vecchi, "Neural correlates of spatial working memory in humans: a functional magnetic resonance imaging study comparing visual and tactile processes," *Neuroscience*, vol. 139, no. 1, pp. 339–349, 2006.
- [68] K. Sathian, "Visual cortical activity during tactile perception in the sighted and the visually deprived," *Developmental Psychobiology*, vol. 46, no. 3, pp. 279–286, 2005.
- [69] A. Pascual-Leone and R. Hamilton, "The metamodal organization of the brain," *Progress in Brain Research*, vol. 134, pp. 427–445, 2001.
- [70] E. Ricciardi and P. Pietrini, "New light from the dark: what blindness can teach us about brain function," *Current Opinion in Neurology*, vol. 24, no. 4, pp. 357–363, 2011.
- [71] S. M. Kosslyn, N. M. Alpert, W. L. Thompson et al., "Visual mental imagery activates topographically organized visual cortex: PET investigations," *Journal of Cognitive Neuroscience*, vol. 5, no. 3, pp. 263–287, 1993.
- [72] S. D. Slotnick, W. L. Thompson, and S. M. Kosslyn, "Visual mental imagery induces retinotopically organized activation of early visual areas," *Cerebral Cortex*, vol. 15, no. 10, pp. 1570–1583, 2005.
- [73] S. M. Kosslyn and W. L. Thompson, "When is early visual cortex activated during visual mental imagery?" *Psychological Bulletin*, vol. 129, no. 5, pp. 723–746, 2003.
- [74] P. J. Laurienti, J. H. Burdette, M. T. Wallace, Y. F. Yen, A. S. Field, and B. E. Stein, "Deactivation of sensory-specific cortex by cross-modal stimuli," *Journal of Cognitive Neuroscience*, vol. 14, no. 3, pp. 420–429, 2002.
- [75] L. B. Merabet, J. D. Swisher, S. A. McMains et al., "Combined activation and deactivation of visual cortex during tactile sensory processing," *Journal of Neurophysiology*, vol. 97, no. 2, pp. 1633–1641, 2007.
- [76] C. Poirier, O. Collignon, A. G. DeVolder et al., "Specific activation of the V5 brain area by auditory motion processing: an fMRI study," *Cognitive Brain Research*, vol. 25, no. 3, pp. 650–658, 2005.
- [77] A. A. Ghazanfar and C. E. Schroeder, "Is neocortex essentially multisensory?" *Trends in Cognitive Sciences*, vol. 10, no. 6, pp. 278–285, 2006.
- [78] O. Collignon, M. Davare, A. G. De Volder, C. Poirier, E. Olivier, and C. Veraart, "Time-course of posterior parietal and occipital cortex contribution to sound localization," *Journal of Cognitive Neuroscience*, vol. 20, no. 8, pp. 1454–1463, 2008.
- [79] D. Basso, A. Pavan, E. Ricciardi et al., "Touching Motion : rTMS on human middle temporal complex interferes with tactile speed perception," *Brain Topography*. In press.
- [80] E. Ricciardi, D. Basso, L. Sani et al., "Functional inhibition of the human middle temporal cortex affects non-visual motion perception: a repetitive transcranial magnetic stimulation study during tactile speed discrimination," *Experimental Biology and Medicine*, vol. 236, no. 2, pp. 138–144, 2011.
- [81] C. Cappe and P. Barone, "Heteromodal connections supporting multisensory integration at low levels of cortical processing in the monkey," *European Journal of Neuroscience*, vol. 22, no. 11, pp. 2886–2902, 2005.
- [82] S. Clavagnier, A. Falchier, and H. Kennedy, "Long-distance feedback projections to area V1: implications for multisensory integration, spatial awareness, and visual consciousness," *Cognitive, Affective and Behavioral Neuroscience*, vol. 4, no. 2, pp. 117–126, 2004.
- [83] A. Falchier, S. Clavagnier, P. Barone, and H. Kennedy, "Anatomical evidence of multimodal integration in primate striate cortex," *Journal of Neuroscience*, vol. 22, no. 13, pp. 5749–5759, 2002.
- [84] K. S. Rockland and H. Ojima, "Multisensory convergence in calcarine visual areas in macaque monkey," *International Journal of Psychophysiology*, vol. 50, no. 1-2, pp. 19–26, 2003.

Research Article

Early Hearing-Impairment Results in Crossmodal Reorganization of Ferret Core Auditory Cortex

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Received 16 December 2011; Accepted 11 April 2012

Academic Editor: Steve Lomber

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Numerous investigations of cortical crossmodal plasticity, most often in congenital or early-deaf subjects, have indicated that secondary auditory cortical areas reorganize to exhibit visual responsiveness while the core auditory regions are largely spared. However, a recent study of adult-deafened ferrets demonstrated that core auditory cortex was reorganized by the somatosensory modality. Because adult animals have matured beyond their critical period of sensory development and plasticity, it was not known if adult-deafening and early-deafening would generate the same crossmodal results. The present study used young, ototoxically-lesioned ferrets ($n = 3$) that, after maturation (avg. = 173 days old), showed significant hearing deficits (avg. threshold = 72 dB SPL). Recordings from single-units ($n = 132$) in core auditory cortex showed that 72% were activated by somatosensory stimulation (compared to 1% in hearing controls). In addition, tracer injection into early hearing-impaired core auditory cortex labeled essentially the same auditory cortical and thalamic projection sources as seen for injections in the hearing controls, indicating that the functional reorganization was not the result of new or latent projections to the cortex. These data, along with similar observations from adult-deafened and adult hearing-impaired animals, support the recently proposed brainstem theory for crossmodal plasticity induced by hearing loss.

1. Introduction

Neural plasticity affords the brain the remarkable capacity for adapting to features of its sensory environment. This same mechanism, however, also renders the brain vulnerable to altered or deprived developmental experiences. Under these conditions, the neural representation of a damaged sensory system can be replaced by inputs from the intact sensory modalities, and this substitution of one sensory modality with another is referred to as crossmodal plasticity. To date, most examples of crossmodal plasticity have been observed in subjects that experienced sensory deprivation/loss either congenitally, or early in life [1–3]. For example, following early-deafness, visual crossmodal effects have been documented within secondary or auditory association areas [2, 4–8]. In addition, crossmodal plasticity has been shown to convey supranormal performance in

the remaining modalities, such as in tasks of visual spatial localization [8–10] or visual motion detection [10] in early-deaf subjects. Recently it has been shown that the neural bases for these perceptual enhancements in the early-deaf were not distributed homogeneously across the “vacated” auditory cortex, but were dependent on specific subregions of visually reorganized auditory cortex [10, 11]. Because each affected neural area houses the circuitry for a specific behavioral program (localization, movement detection) that is the same in deaf or hearing subjects, and enhanced performances are based on stimulus features that are common to both the auditory and visual modalities (e.g., stimulus location or movement velocity), these observations suggest a supramodal basis for enhanced crossmodal performance, an effect now regarded as Lomber’s Law [10]. These same experiments also indicated that some areas of vacated auditory cortex, in particular A1, were not involved in

any of the many visual crossmodal tasks examined, an effect that was consistent with numerous studies of early-deaf subjects [2, 4–7, 12]. Thus, early-deafness can lead to supranormal crossmodal performance on specific tasks mediated by particular subregions of auditory cortex, while other regions seem to be unaffected by crossmodal plasticity.

In contrast to the considerable attention that early sensory loss has received, very few studies have examined the crossmodal effects of late, or adult, sensory loss. Instead, the neural bases for deafness-induced adult crossmodal plasticity were virtually unexplored until recently. However, Allman et al. [13] demonstrated that ferrets, deafened as mature adults, exhibited a robust somatosensory reorganization of core auditory cortex, which included both the primary and anterior auditory fields. This single-unit recording study showed that neurons in the reorganized core auditory cortices were cutaneously driven, exhibited receptive fields located bilaterally on the head or head and neck, and lacked a global somatotopy. Thus, the core auditory cortex, described by so many studies as lacking visual crossmodal inputs (see above), is crossmodally innervated by somatosensory inputs following adult hearing loss. In fact, somatosensory reorganization of core auditory cortex was observed not only in response to profound deafening in adults [13], but in adult animals with incomplete hearing loss as well [14, 15]. However, because these mature animals were deafened long after their critical period for sensory development and plasticity (which ends near postnatal day 60 [16]), it could not be predicted whether the same crossmodal effects on core auditory cortex would also occur after early hearing loss. Therefore, the present experiments were designed to use the same experimental approaches as in the examination of the effects of adult-deafness, except that hearing deficits were induced early in the developmental sequence.

2. Materials and Methods

All procedures were performed in compliance with the Guide for Care and Use of Laboratory Animals (National Institutes of Health, publication 86-23), the National Research Council's Guidelines for Care and Use of Mammals in Neuroscience and Behavioral Research (2003), and approved by the Institutional Animal Care and Use Committee at Virginia Commonwealth University. These procedures are the same as those used by Allman et al. [13] for examining the crossmodal properties of auditory cortical neurons in adult-deafened ferrets.

2.1. Deafening. Ferrets begin to hear by the end of the first postnatal month [17], and primary auditory cortex reaches maturity one month later at approximately 60 days postnatally [16]. Therefore, to damage the functioning auditory system [18] before it matures, kanamycin (300 mg/kg, s.c.) and ethacrynic acid (25 mg/kg, i.v.; after protocol of [19]) were coadministered to ferrets ($n = 3$) on postnatal day 49 (see Table 1 for vital statistics). At approximately four weeks after ototoxic treatment, auditory brainstem responses (ABRs) were assessed for each ear separately, as shown

in Figure 1. The auditory stimulus was a calibrated click (2000 trials each, 0.1 ms square-wave click, rarefaction), delivered through a speaker positioned in front of one ear. Subdermal recording leads were inserted over the right and left mastoid processes, at midcranium and on midback. Evoked electrical activity was signal averaged, and threshold response levels were determined using a descending (5–10 dB SPL increments) sequence of sound intensity for each ear of each animal. Bilateral ABRs were also tested on hearing animals (threshold ~ 15 dB SPL; reported in [13]).

2.2. Electrophysiology. At three to five months after the ototoxic procedure (see Table 1), the animals were at or near sexual maturity (150–180 days of age) and well beyond the critical period of auditory cortical development [16]. The early hearing-impaired ferrets were surgically prepared for electrophysiological recording. Under pentobarbital anesthesia (40 mg/kg, i.p.) and aseptic surgical conditions, a craniotomy was made over the left cortical hemisphere to expose the auditory cortices. Next, a stainless-steel well/head-support device was implanted using screws and dental acrylic, and the incision was closed around the implant. A standard postoperative antibiotic and analgesic regimen was administered, and the recording experiment occurred 2–4 days after implantation. Procedures and data from four, age-matched normal hearing ferrets (mean = 199 ± 4 DPN) also provided controls for another published study and are fully described there [13].

Electrophysiological recordings were initiated by anesthetizing the animal (35 mg/kg Ketamine; 2 mg/kg Acepromazine i.m.) and fixing the implanted well to a supporting bar. The animal was intubated through the mouth and ventilated with expired CO_2 monitored and maintained at $\sim 4.5\%$. Fluids, supplemental anesthetics (8 mg/kg/h Ketamine; 0.5 mg/kg/h Acepromazine), and a muscle relaxant (Pancuronium bromide 0.2 mg/kg/h i.p.) were continuously infused. This drug regimen was necessary to prevent spontaneous eye and body movements during the lengthy sensory/multisensory tests. The implant was opened, and the recording electrode (glass-insulated tungsten; $< 1 \text{ M}\Omega$ impedance at 1000 Hz) was inserted into core auditory cortex guided by gyral/sulcal landmarks and the functional map published by [20].

With the electrode inserted into auditory cortex, neuronal activity was amplified and routed to a computer. Neurons were identified by their spontaneous activity and their responses to an extensive battery of manually presented auditory (claps, clicks, whistles, and hisses), visual (flashed or moving dark or light stimuli) somatosensory (strokes and taps using brushes and calibrated Semmes-Weinstein filaments; air puffs), manual pressure, and joint rotation) stimuli. Thus, at each location, the sensory response modality of the neuron (auditory, visual, somatosensory, multisensory, and unresponsive) was identified and tabulated, and the sensory receptive field(s) were mapped and graphically recorded. To reduce sampling bias during single-unit recording penetrations, neurons were studied at $250 \mu\text{m}$ intervals. Due to their significant hearing loss, standard

TABLE 1: Hearing and age statistics for the ferrets with early hearing loss induced by ototoxic lesion (OT); all animals were male. The age of hearing onset in ferrets is ~ 32 days postnatally, and auditory critical period closure for A1 is 80 days [16].

ID no.	Weight	Age at OT lesion	ABR hear threshold	Age at recording	Impairment duration	BDA volume
DJF 1	1.7 kg	49 days	65 dB SPL	148 days	99 days	0.5 μL
DJF 3	2.3 kg	49 days	65 dB SPL	182 days	133 days	0.5 μL
DJF 4	1.9 kg	49 days	85 dB SPL	190 days	141 days	0.5 μL

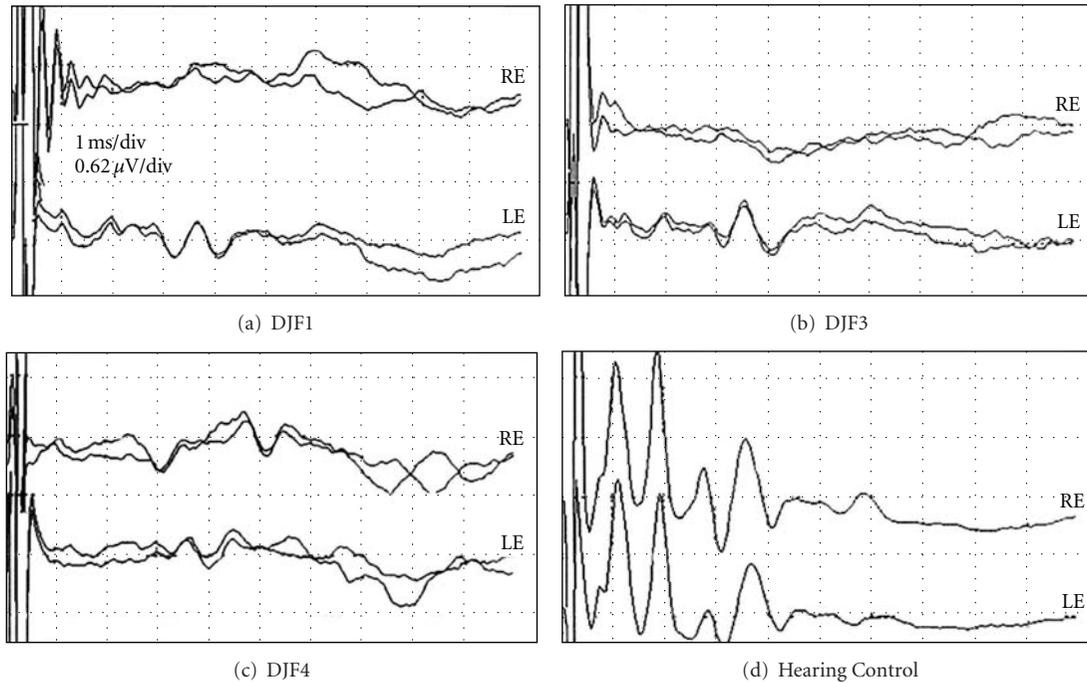


FIGURE 1: Auditory brainstem response (ABR) data for ferrets with early hearing loss (a–c) or normal hearing (d). For each panel, the auditory stimulus was a calibrated click (90 dB SPL; 0.1 ms square-wave click, rarefaction), delivered through a speaker positioned directly in front of one ear (RE = right; LE = left). Each waveform represents the average of 2000 trials; overlapped dual waveforms indicate that the test was repeated. Scale, indicated in panel (a), is the same for each panel. As evidenced by comparison of panels “a–c” with that of the hearing control in (d), all ferrets with early hearing loss demonstrated profoundly reduced ABRs to 90 dB SPL stimuli. However, some residual auditory response was apparent in each of the treated animals, as demonstrated by the small but repeatable peaks at approximately 3 ms latency. Further tests (not depicted) indicated that hearing thresholds for the treated animals ranged from 65 to 85 dB SPL (see Table 1).

measures of auditory frequency-response tuning were not attempted in these experiments.

Additional, quantitative sensory/multisensory tests were performed at selected recording sites. Quantitative sensory tests consisted of computer-triggered auditory, visual, and somatosensory stimuli, presented alone and in combination. Free-field auditory cues were electronically generated white-noise bursts (100 ms, >80 dB SPL) from a hoop-mounted speaker 44 cm from the head (45° azimuth/ 0° elevation). Visual cues were bars of light projected onto a translucent hemisphere (92 cm diameter) whose size, direction, velocity, and amplitude were independently controlled. Somatosensory stimulation was achieved using an electronically-driven, modified shaker with independently programmable amplitude, and velocity settings to indent skin/deflect hairs. When no receptive field could be identified, a standard stimulus configuration was presented: auditory stimulus as described above; a large light bar ($5 \times 15^\circ$) that moved

across the contralateral visual field from nasal to temporal at $200^\circ/\text{s}$; the somatosensory probe was positioned to stimulate the contralateral cheek. Each stimulus presentation was separated by 3–7 s, and each condition was presented 25 times. Neuronal responses were digitized (rate > 25 kHz), and individual waveforms were templated and routed to a computer for analysis. For each waveform (i.e., single neuron), a peristimulus-time histogram was constructed for each of the test conditions from which the response (mean spikes per trial) was measured. Unisensory neurons (auditory, visual, and somatosensory) were identified as those which were activated or influenced by only one sensory modality. Multisensory neurons activated by two different sensory modalities were defined as bimodal (e.g., auditory-somatosensory); those activated by three were classified as trimodal neurons. Multisensory neurons activated by one modality but whose response could be modulated (suppressed or facilitated) by a second modality that was

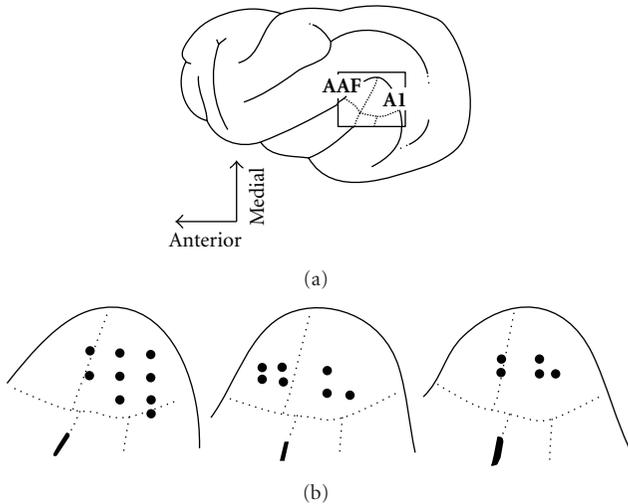


FIGURE 2: Single-unit recording penetrations in core (AAF and A1) auditory cortex of the three ferrets with early hearing loss. Part (a) shows a lateral view of the ferret cortex with the location of the core auditory areas of anterior auditory field (AAF) and primary auditory cortex (A1) indicated (boxed). Part (b) shows tracings of surface photographs of the positions of the recording penetrations ($n = 21$) made in each of the hearing impaired animals. Dotted lines approximate the borders of the auditory fields (after [20]).

ineffective alone were categorized as subthreshold neurons [15].

The depth of each neuron within a penetration was noted and, in a data table, correlated with its sensory response type: unisensory auditory, visual, somatosensory, and multisensory combinations thereof, or unresponsive. Several recording penetrations were made in each animal, and their location was plotted on a digital photograph of the cortical surface. Each recording penetration was marked at its terminus with a small electrolytic lesion to facilitate its histological reconstruction. At the conclusion of the recording experiment, the animal received a barbiturate overdose (pentobarbital, 120 mg/kg, i.v.) and was perfused transcardially with saline followed by fixative (4% paraformaldehyde). The brain was removed from the cranium, and the auditory cortical regions were stereotaxically blocked and serially sectioned (50 μm thick). The sections were processed using standard histological procedures, and a projecting microscope was used to make scaled reconstructions of the recording penetrations. Auditory cortical fields were approximated using sulcal landmarks according to the criteria of [20]. However, the sulcal borders of these fields have not been mapped. Furthermore, the adjoining sulci are known to contain non-auditory representations, such as the anterior and posterior lateral suprasylvian visual regions [21] or the pseudosylvian somatosensory region [22]. Therefore, a conservative approach was adopted to exclude data from these potentially non-auditory regions, whereby the core auditory cortices were defined by a border at the lip of the relevant sulcus.

2.3. Neuroanatomy. The cortical connectivity of the three early ototoxically-treated ferrets (the right hemisphere from

the recorded animals) and four hearing adult ferrets was examined. Approximately 1 week prior to the terminal recording session and using pentobarbital anesthesia (40 mg/kg i.p.) and aseptic surgical conditions, a craniotomy was made to expose the right auditory cortices. Biotinylated dextran amine (BDA, 3k MW, 10% in citrate buffer) was pressure injected (0.8–1.5 μL volume) from a 5 μL Hamilton syringe into the A1 region using gyral/sulcal landmarks and the criteria of [20]. After a period for tracer transport, the animal was euthanized, perfused (saline) and fixed (4% paraformaldehyde). The brain was removed, blocked stereotaxically, and sectioned (50 μm thick) in the coronal plane. Sections were processed using the protocol of [23] with metal intensification, mounted on slides, and coverslipped without counterstain. The processed sections were digitized using Neurolucida software (MBF Biosciences, Williston, VT, USA) to document the tissue outline, grey-white border, and the location of retrogradely labeled neuronal cell bodies. Functional maps of ferret cortex [20, 21, 24–26] and thalamus [27] were used to identify the regions containing retrogradely labeled neurons. For display, the sections were graphically arranged, and neural regions were identified using accepted sulcal/gyral/cytoarchitectonic landmarks [20, 26–28].

3. Results

Young ferrets ($n = 3$), ototoxically treated after hearing onset but well before the end of their critical period of auditory development, showed significant levels of hearing impairment as adults, as illustrated in Figure 1. Each treated animal showed an elevated hearing threshold of 65–85 dB SPL, which is summarized in Table 1. In contrast, the untreated hearing controls exhibited hearing thresholds of approximately 15 dB SPL [13].

To evaluate the sensory responsiveness of auditory cortical neurons in the hearing-impaired animals, single-unit recordings were made at sites across the upper/medial aspects of the middle ectosylvian gyrus (MEG), as depicted in Figure 2. This figure summarizes the cortical location of recording penetrations plotted from surface photographs for each animal. A total of 21 effective recording penetrations were made into the AAF or A1 regions. Because the animals were significantly hearing-impaired, however, the functional border between AAF and A1 could not be mapped, and the auditory cytoarchitectonic borders have not yet been described in this species. Hence, recordings were regarded as samples of the core (inclusive of AAF and A1) auditory cortices based on gyral/sulcal landmarks, histological reconstruction (see below), and published functional maps [20].

Recording penetrations in the core auditory cortices of the early hearing-impaired ferrets revealed neurons that were responsive to somatosensory and/or auditory stimulation. As evidenced in the raster/histogram of Figure 3(a), some neurons were unresponsive to acoustic stimulation, but were robustly and reliably activated by a tactile stimulus presented within its receptive field. Furthermore, the tactile response was not significantly influenced when somatosensory and auditory stimulation was combined. Other neurons, like that

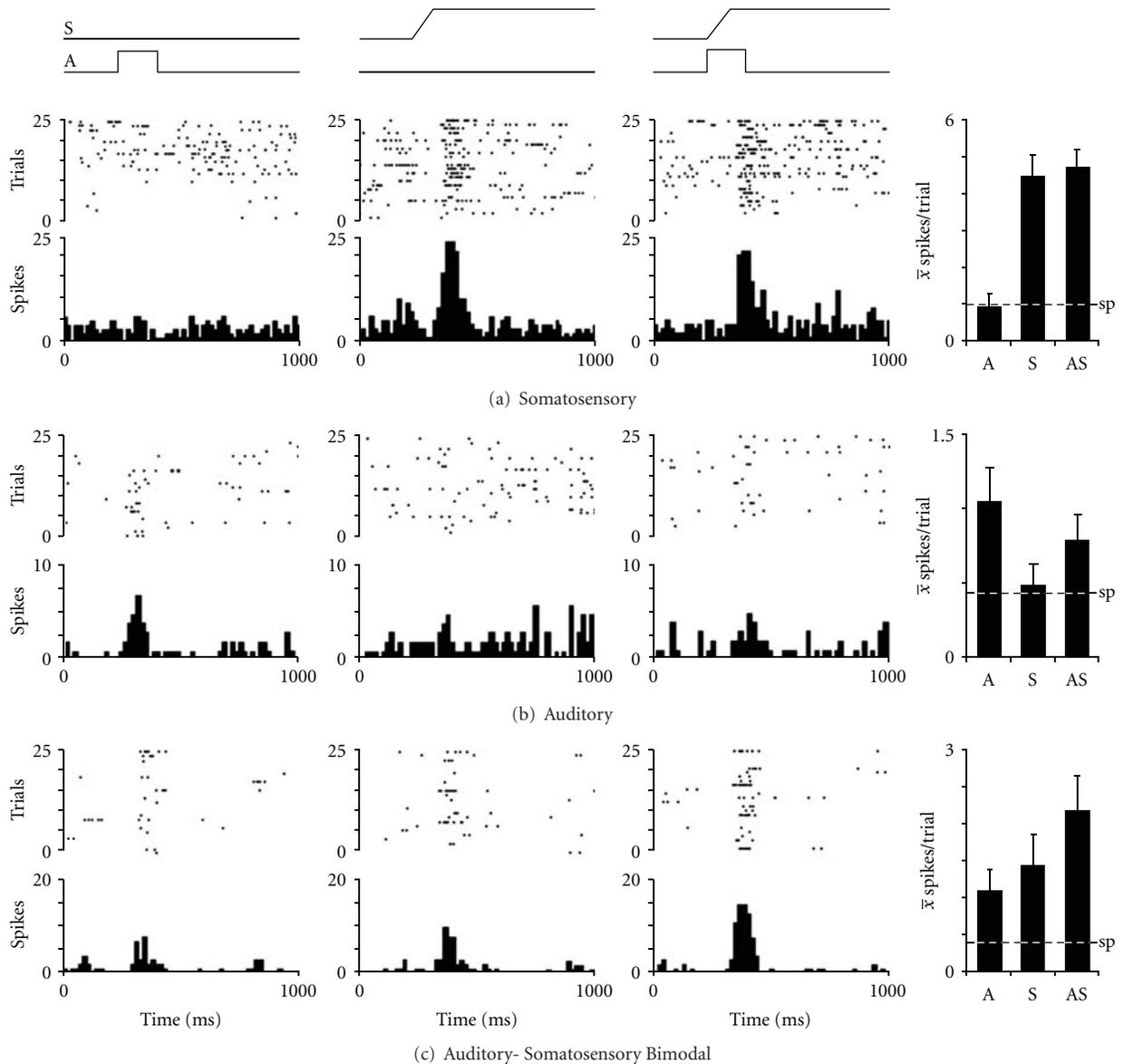


FIGURE 3: Sensory responses of neurons from core auditory cortex of ferrets with early hearing impairment. Single-unit recordings revealed that neurons were responsive to auditory (square wave, white noise), somatosensory (ramp; 1 g filament displacement of skin/hair) or combined auditory-somatosensory stimulation as displayed in the raster/histogram rows. In (a), the neuron was unresponsive to the auditory stimulus, but was vigorously activated by the somatosensory cue, and this response was not significantly altered when the two stimuli were combined. These responses are summarized by the bar graph (far right; error = standard error; sp = spontaneous activity), and are characteristic of a unisensory somatosensory neuron. The same conventions are used in the subsequent rows where activity indicative of a neuron with unisensory auditory properties (b) and a neuron with bimodal auditory-somatosensory response features (c) are illustrated. None of the depicted responses to combined stimulation were significantly (paired t -test) different from their best unisensory responses. These data show that core auditory cortical neurons in these hearing-impaired animals exhibit vigorous somatosensory-evoked activity.

represented in Figure 3(b), were responsive to acoustic stimulation, but were not significantly affected by tactile cues. In addition, many of the identified neurons were multisensory because they were activated by auditory stimulation alone as well as by independent somatosensory stimulation. An example of such a bimodal neuron is shown in Figure 3(c). All neurons were also tested for responsiveness to visual stimulation (see below); some neurons were encountered

that were unresponsive to all the different stimuli and their combinations (not illustrated).

Neurons responsive to auditory as well as somatosensory cues were encountered across the MEG, as well as through the full thickness of the cortical mantle. Histological reconstructions of the recording sites (summarized in Figure 4) depict the location of each recording penetration and the neuron types that were encountered. Even though the

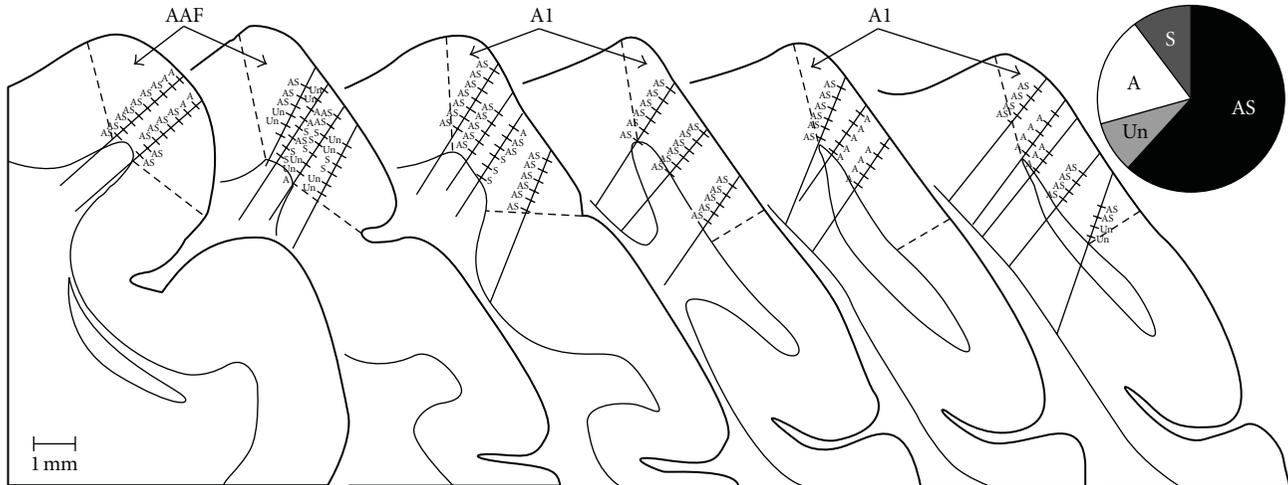


FIGURE 4: Coronal sections through core auditory cortex (AAF and A1) summarize each of the single-unit recording penetrations ($n = 21$) in ferrets with early hearing loss. For each penetration, the hashmarks indicate the location of an identified neuron ($n = 132$) whose functional properties are indicated: A = auditory; S = somatosensory; AS = auditory-somatosensory; Un = unresponsive. The pie chart (top right) summarizes the proportions of encountered neuron types: AS = 62%; S = 10%; A = 19%; and Un = 9%. The coronal sections are serially arranged (anterior : left) with the thin contour representing the gray-white border and the dashed lines indicating the presumed borders of AAF and A1. Because the sulcal borders of these regions have not been mapped, it was assumed that the sulcal extent of each area terminated at the lip of the sulcus. Only those neurons whose location plotted within the depicted borders of AAF/A1 were included in this study. Not all neurons are plotted due to overlap.

recording penetrations clearly continued into white matter or the adjoining lateral bank of the suprasylvian sulcus, only those recording sites within the gyral aspects of the MEG were included for this study (see Section 2). When all neurons identified in 21 penetrations were tallied ($n = 132$), the overwhelming majority showed bimodal auditory-somatosensory properties (were independently activated by separate auditory and somatosensory stimulation; $62 \pm 19\%$ sem; see pie chart in Figure 4). In addition, another 10% (± 8 sem) were activated by somatosensory stimulation alone (e.g., unisensory somatosensory). Combining these values indicates that nearly 3/4ths of core auditory cortex in early hearing-impaired ferrets was responsive to somatosensory stimulation. In contrast, only 1 (1%; 1/100) somatosensory neuron was identified in similar recordings from normal hearing controls (from [13]).

In each case, every neuron was tested not only for auditory and somatosensory activation, but for visual inputs as well. In 6 of the penetrations, a total of 19 neurons responsive to visual stimulation were also identified. However, upon reconstruction of the recording tracks, these neurons were uniformly located at the deepest points of the penetration and within the bank of the suprasylvian sulcus that corresponds with the proposed lateral suprasylvian visual area of normal animals [21]. These visually-responsive neurons did not meet the criterion for residing within the gyral portion of core auditory cortex [20] and, therefore, were excluded from further analysis.

Somatosensory responses were recorded in the overwhelming majority ($>80\%$; 17/21) of recording penetrations in the early hearing-impaired cortices, as shown in

Figure 4. Somatosensory responses were observed through the full thickness of the cortical mantle, and representative somatosensory receptive fields observed for a given track are plotted in Figure 5. These data show for each animal that somatosensory receptive fields always included the face and often extended into other adjoining regions of the neck and/or forelimb and represented inputs carried by trigeminal and cervical nerves. In addition, the somatosensory receptive fields often represented bilateral aspects of the body surface including the standard contralateral representation as well as ipsilateral features. As also shown in the bilateral receptive fields depicted in Figure 5, their contralateral and ipsilateral distributions were usually symmetrical. Thus, as depicted in Figure 5(a), receptive fields that included the contralateral forepaw also included the same ipsilateral region, or in Figure 5(c) where the contralateral and ipsilateral pinnae are represented together in the same neuron. In addition, within a given recording penetration, somatosensory receptive fields tended to cluster around representation of a particular segment of the body surface. Two examples of this effect are provided in Figure 6. The recording penetration illustrated in Figure 6(a) occurs orthogonal to the pial surface and shows a recording sequence in which somatosensory receptive fields on the cheek, pinna, and neck were consistent along the depth of the recording penetration. However, the receptive fields shifted from bilateral to contralateral for the neurons located the deepest in the penetration. A similar vertically-oriented penetration depicted in Figure 6(b) also shows that somatosensory receptive fields were nested on the representation of the face, head, and neck, but abruptly expanded onto the bilateral forelimbs at the deepest recording sites.

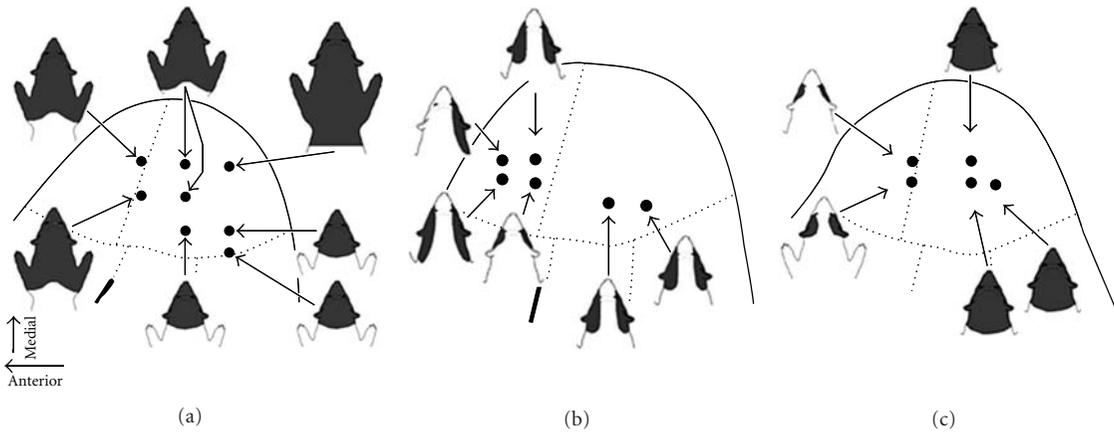


FIGURE 5: Distribution of somatosensory receptive fields in core auditory cortex of early hearing-impaired ferrets. For each of the surface plots for the different hearing-impaired animals (a–c), the associated schematic of the ferret’s body surface shows the somatosensory receptive field(s) (shaded dark gray) encountered in that recording penetration. Note that in each case somatosensory receptive fields were located on the anterior aspect of the body (head, neck, and forelimb) and that they were predominantly bilateral (included both ipsi- and contralateral body surface).

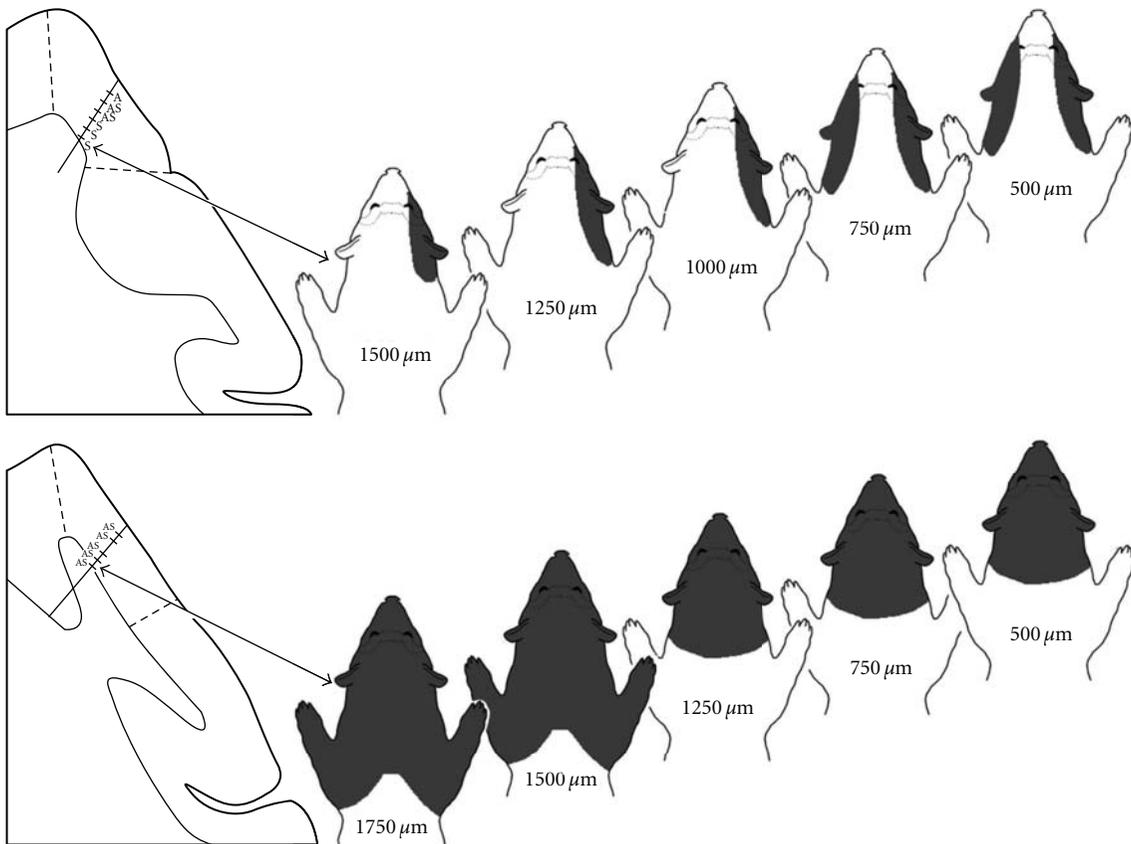


FIGURE 6: Distribution of somatosensory receptive fields across the cortical mantle (gray matter) of core auditory cortex in early hearing-impaired ferrets. Segments of coronal sections through core auditory cortex (left) show the location of a particular recording penetration and the sensory responsiveness of the neurons identified (A = auditory, S = somatosensory; AS = auditory and somatosensory). To the right, the series of ferret body surface depictions indicate the location of the somatosensory receptive field (shaded dark gray) and depth (in microns) corresponding to the neuron and recording penetration plotted on the tissue section. These data show that somatosensory reorganization of core auditory cortex in early hearing impaired ferrets was robust across the fullthickness of the cortex, represented the anterior segment of the body, and was often bilateral (on the ipsi- and contralateral body surface).

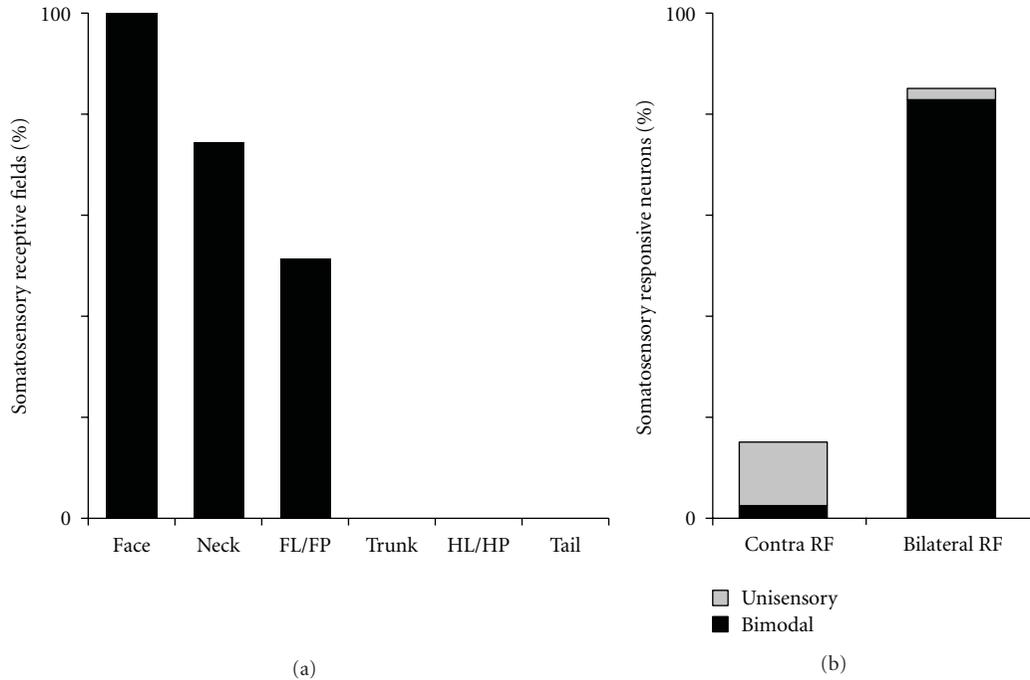


FIGURE 7: Somatosensory receptive field properties of neurons in core auditory cortex of early hearing-impaired ferrets. Part (a) shows that all somatosensory receptive fields included representation of the face and that these often extended into the neck or forelimb/forepaw (FL/FP) regions. Conversely, no somatosensory receptive fields were observed representing the posterior portion of the body (trunk, hindlimb/hindpaw (HL/HP), tail). Part (b) illustrates that the overwhelming majority of somatosensory receptive fields were bilateral (included ipsi- and contralateral body surface) of which 97% occurred in bimodal auditory-somatosensory neurons (black region of bar). In contrast, comparatively few neurons exhibited exclusively contralateral somatosensory receptive fields, most of which (86%; grey region of bar) occurred in unisensory somatosensory neurons.

When all of the mapped somatosensory receptive fields were analyzed, it became apparent that all somatosensory-responsive neurons had receptive fields that included the face, as is summarized in Figure 7(a). It was also revealed that many somatosensory receptive fields could also include the neck, or neck and forelimb/forepaw. However, no receptive fields that included the trunk, hindlimb/paw or tail were encountered. Also a laterality component of receptive field distribution that strongly correlated with the unisensory/multisensory nature of the parent neuron was observed. As shown in Figure 7(b), bilateral receptive fields predominated over those with purely contralateral representations (85:15 ratio). Furthermore, 86% of unisensory somatosensory neurons exhibited receptive fields with contralateral distributions, while 97.5% of bimodal neurons demonstrated bilateral receptive fields. Most (65%) somatosensory neurons were excited by very low force threshold stimulation of ≤ 1 gram, which is consistent with activation of peripheral hair receptors.

These recordings in early hearing-impaired ferrets indicate that approximately 72% of core auditory cortical neurons are responsive to somatosensory stimulation. In contrast, using the same recording methods, only 1% of neurons from hearing controls showed the same somatosensory sensitivity [13]. Therefore, it seemed possible that during and following postnatal development, a large contingent of

novel somatosensory inputs reached the auditory cortices of the early hearing-impaired animals. To examine whether these crossmodal inputs arrive from somatosensory cortical sources, the core auditory cortex of these same early hearing-impaired ferrets ($n = 3$) received tracer (BDA) injections, and the loci of the resulting retrogradely labeled neurons were plotted. A representative example is presented in Figure 8(a), which shows that sources of inputs to early hearing-impaired arose largely from other auditory cortices, and the somatosensory and visual regions were essentially devoid of label. This pattern of labeling was consistent for 2 of the 3 cases. In the third case, the injection was more extensive and included not only the subadjacent white matter, but also aspects of the medial bank of the pseudosylvian sulcus known to receive visual inputs [22] and contains the anterior ectosylvian visual area [26]. In this case, the same auditory cortical areas revealed retrograde labeling, but visual areas 19, 20a, and 20b (after [28]) were also labeled. However, and none of the three cases were more than a few labeled neurons found in any of the somatosensory cortical areas. Collectively, these data demonstrated that cortical projections to core auditory cortex of early hearing-impaired ferrets were almost entirely from other auditory cortical areas, just like they were for normal hearing ferrets (see Figure 8(b) of [13]).

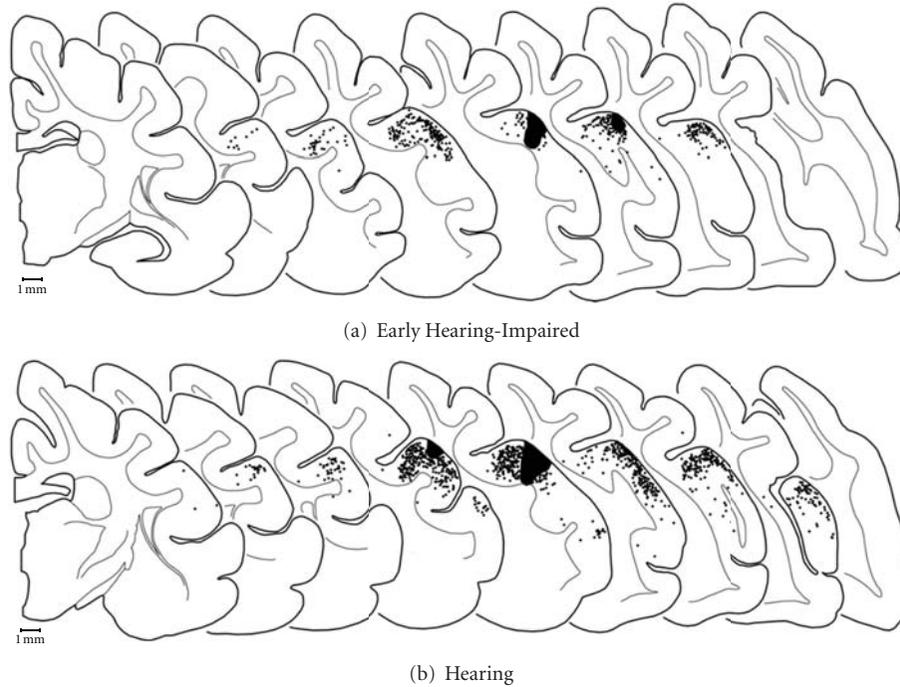


FIGURE 8: Sources of cortical inputs to core auditory cortices for early hearing-impaired (a) and normal hearing (b) ferrets. On serially arranged (anterior = left) coronal sections through one cortical hemisphere, tracer injection (BDA; solid black area) into core auditory cortex produced retrogradely labeled neurons (1 dot = 1 neuron) primarily within the regions regarded as auditory cortex: on the middle ectosylvian gyrus (MEG) and the posterior aspect of the anterior ectosylvian gyrus (AEG), as well as within the lateral bank of the suprasylvian sulcus (SSS) and into the bank of the pseudosylvian sulcus (PSS). The distribution of labeled neurons is essentially coextensive for hearing-impaired and hearing animals, with no difference in labeling found within somatosensory regions on the suprasylvian (SSG) and anterior ectosylvian gyri. Hearing controls replotted from [13].

To assess whether crossmodal inputs to auditory cortex of early hearing-impaired ferrets might arise from somatosensory thalamus, the same cases described above were used to plot the location of labeled thalamocortical neurons. As illustrated in Figure 9(a), a small injection into core auditory cortex of an early hearing-impaired animal almost exclusively yielded retrogradely labeled neurons within the medial geniculate nucleus of the auditory thalamus. This pattern of labeling was consistent for 2 of the 3 cases. In the third case, the injection was more extensive and extended into the visual area of the pseudosylvian sulcus (AEV; [22, 26]). In this case, labeled thalamocortical neurons were more frequent within the LP and Po regions, and a few were observed in the visual LGN or in the posterior-lateral aspects of the somatosensory VB. In all early hearing-impaired cases, however, the overwhelming preponderance of thalamic projections to core auditory cortex was from the medial geniculate, which was consistent with that observed in normal hearing ferrets (Figure 9(b) of [13]).

4. Discussion

These data show that early hearing loss results in cross-modal reorganization of core auditory cortex, such that neurons normally driven by auditory inputs respond to somatosensory stimulation. The novelty and importance of

this observation resides in the context of the literature of crossmodal plasticity following sensory loss. First, many studies have reported a lack of crossmodal innervation of core auditory cortex in early-deaf subjects [2, 4–7, 12]. However, these studies tested for visual responses and, hence, established only that visual crossmodal effects were not observed. In contrast, when early-deaf subjects were examined using somatosensory stimulation, crossmodal plasticity of the core auditory areas was observed in humans [29] and animals (present study). In addition, the present results from early hearing loss closely correspond with that observed in animals with late, or adult, hearing loss [13–15]. In both conditions (early- and late-hearing loss), the preponderance of single-unit recordings from neurons in core auditory cortex was demonstrated to exhibit robust responses to somatosensory inputs. Therefore, rather than being immune to the plasticity that occurs elsewhere in the auditory cortices following hearing loss, core auditory cortex actually exhibits crossmodal effects in the form of somatosensory reorganization.

Single-unit studies of crossmodal plasticity provide unique insights into the features of the reorganizing modality. Not only do neurons in core auditory cortex of early hearing-impaired animals respond to somatosensory stimulation, the majority of them are activated by low force-threshold receptors corresponding to hair-type receptors. In

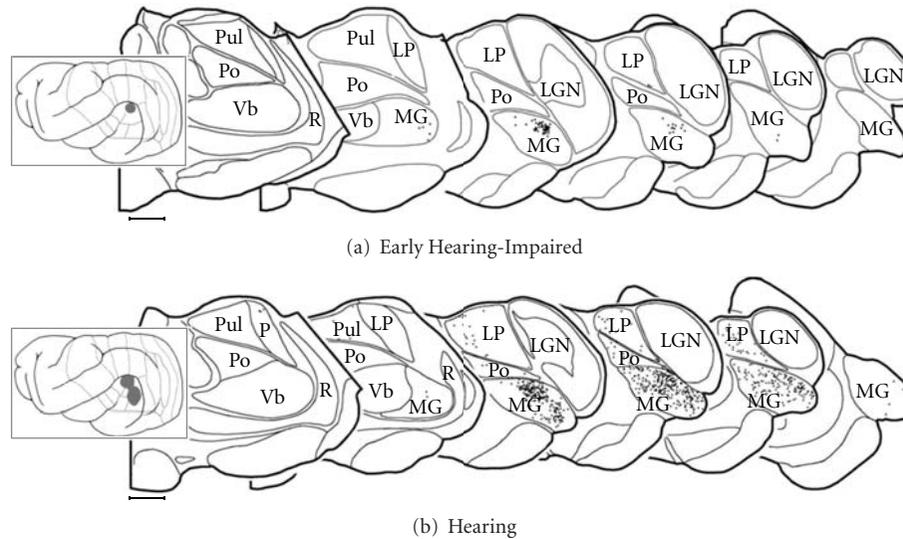


FIGURE 9: Sources of thalamic inputs to core auditory cortices for early hearing-impaired (a) and normal hearing (b) ferrets. On serially arranged (anterior:left) coronal sections through the thalamus of an early hearing-impaired ferret, tracer injection (BDA; solid grey area) into core auditory cortex produced retrogradely labeled neurons (1 dot = 1 neuron) essentially within the regions regarded as auditory thalamus: in the medial geniculate nucleus (MG). No retrgradely labeled neurons were identified in somatosensory (Vb) or visual (LGN) thalamus although a few were found in the border of the lateral posterior (LP) and posterior (Po) nuclei. Although a larger injection in a hearing animal encroached on the secondary auditory fields, the thalamic labeling was focused on the MG, with lesser connectivity with the LP and Po; no labeled neurons were observed in somatosensory (Vb) or visual (LGN) thalamic nuclei. Hearing controls replotted from [13].

addition, the receptive fields of the individual, crossmodally innervated neurons, could be mapped. These data showed that all such receptive fields included the head/face, and many also extended onto the neck and/or forelimb. Thus, there was a strong preference for representation of the anterior aspects of the body surface conveyed by trigeminal/cervical nerves, and no receptive fields were encountered that included the hindlimb or tail. This information suggests that the fMRI study of congenitally deaf humans may have activated even larger proportions of core auditory cortex had the test stimuli been applied to the face rather than the hands [29]. Furthermore, over 80% of the receptive fields were bilateral in distribution, such that they included both contralateral and ipsilateral portions of the body surface. The bilateral arrangement of these receptive fields, and their preponderance in the present sample, are quite unlike the contralateral distribution of receptive fields encountered in somatosensory cortical areas of ferret SI–SIII [24, 25, 30] and the medial rostral suprasylvian somatosensory area [31]. Collectively, it is difficult to discern a pattern of somatotopy from these results, given that the same receptive field locations are represented at widely different locations within core auditory cortex (e.g., see Figure 5). Ultimately, these somatosensory features of reorganized auditory cortex in early hearing-impaired ferrets are fundamentally similar to those observed in late-, or adult-onset deaf [13] or hearing-impaired ferrets [14, 15]. Consistent among each of these types of hearing loss, core auditory cortical crossmodal plasticity was characterized by somatosensory reorganization that was activated largely by low force-threshold hair type

receptors and represented the head/anterior body surface, often bilaterally, without apparent global somatotopy. At present, it is difficult to postulate how such reorganization might convey “adaptive” or “compensatory” advantages at neuronal or perceptual levels, but instead may contribute to the growing literature that interprets some forms of crossmodal plasticity as “maladaptive,” such as tinnitus (reviewed in [32]).

Given the overwhelming presence of somatosensory-responsive neurons in core auditory cortex of these early hearing-impaired animals, it would be expected that robust connections with somatosensory brain regions would be established in these altered animals. In addition, because a large proportion of crossmodal studies have identified changes specifically in cortical function (reviewed in [2, 33]), novel cortical connections between core auditory cortex and portions of somatosensory cortex would seem most likely to occur. However, tracer injections placed in core auditory cortex of early hearing-impaired animals revealed few, if any, inputs from somatosensory cortex and did not appear to be different from core auditory cortical connections in hearing control animals. Furthermore, thalamocortical connections did not reorganize in the early hearing-impaired animals as the somatosensory thalamic nuclei were essentially devoid of label. These data indicate that changes in regional cortical and thalamocortical connectivity were not sufficient to underlie the wholesale functional changes observed in core auditory cortex of early hearing-impaired ferrets. Ultimately, these connectional data in early hearing-impaired ferrets are essentially identical to those observed for the somatosensory

reorganized core auditory cortex of adult-deafened ferrets [13].

Because the neuroplastic effects that occur during the developmental critical period of young animals or subjects are well known, it might be expected that crossmodal effects in early lesioned subjects would differ from those whose onset occurs during adulthood. However, crossmodal plasticity has only rarely been examined in adults. Sound localization behavior in late visually-deprived cats was observed to be similar to, but not as strong as that demonstrated by early-deprived animals [34]. Likewise, sound localization in early blind [35] and late-blind individuals [36] was behaviorally similar, but appears to involve different components of the neural response [36]. The present study of crossmodal functional and connectional effects in early hearing-impaired ferrets closely resembles the results from adult-deaf [13] or adult hearing-impaired ferrets [14, 15]. Collectively, these studies represent, to the best of our knowledge, among the only single-unit or neuroanatomical studies of cortical crossmodal plasticity precipitated by hearing loss (see also [11]). Despite the different stages of maturity and different severity of hearing loss involved, the results were quite similar; the core auditory cortex exhibited a functional somatosensory reorganization that was not accompanied by regional changes in connectivity. As discussed below, only a new theory of cortical crossmodal plasticity can account for these combined observations.

The mechanism underlying crossmodal plasticity has been of considerable interest. More than a decade ago, Rauschecker [33] summarized the known possibilities as follows: “[Crossmodal] plasticity might involve any or all of these neural mechanisms: unmasking of silent inputs; stabilization of normally transient connections, axonal sprouting; or a combination of them.” However, recent data is difficult to reconcile with these earlier postulates. With regard to unmasking of silent inputs, such crossmodal inputs should be revealed by connectional studies of auditory cortex in normal hearing animals. In neither the ferret (present studies) nor the cat [37–39] is there sufficient connectivity from somatosensory structures to underlie the robust somatosensory reorganization of the entire auditory area although the cortex of rodents appears to have a higher proportion of natural crossmodal connections [40, 41]. Alternatively, if crossmodal plasticity was subserved by the preservation of transient connections, then core auditory connectivity should be different between the early hearing-impaired and the normal hearing animals. The present study demonstrated that they were not fundamentally different. Furthermore, this particular mechanism could not account for the somatosensory reorganization observed in adult-deafened ferrets [13]. Last, in the present experiments, there was no evidence of sprouting of axons (e.g., ingrowth of new connections) from somatosensory cortical or thalamic regions in either the early- or late hearing-impaired animals sufficient to underlie the observed crossmodal effects. Therefore, an alternate hypothesis for the mechanism underlying early and late-deafness-induced crossmodal plasticity has been proposed by Allman et al. [13]. This theory accounts for the functional reorganization of auditory cortex in the

deaf without requiring the actual plasticity to occur within the cortex. As has been known for over a decade now, the auditory brainstem naturally receives crossmodal inputs from the somatosensory system at several critical nodes. Neurons in the dorsal cochlear nucleus [42–44] as well as the inferior colliculus [45] have been demonstrated to be affected by somatosensory stimulation or by activation of the trigeminal nucleus. Furthermore, a recent study [46] showed that hearing loss enhances the level of crossmodal somatosensory innervation of the dorsal cochlear nucleus, where significant decreases in response thresholds as well as latency and duration changes were evident. Thus, cochlear damage results in the loss of functional auditory inputs to the cochlear nucleus and induces somatosensory crossmodal plasticity there. Because the cochlear nucleus is the first node in the ascending auditory projection, any functional changes within that nucleus are reflected throughout the entire auditory pathway, including cortex. This postulate is consistent with the representation of trigeminal and cervical somatosensory regions in deafened auditory cortex. Furthermore, given the highly crossed nature of the ascending auditory projection, it is not surprising that a high proportion of crossmodal somatosensory receptive fields in auditory cortical neurons are bilateral. Thus, the brainstem theory of cortical crossmodal *reorganization* (not *plasticity*), while being a significant departure from earlier postulates, seems well supported by empirical observations from different published sources and points of view. Further studies are necessary to map the specific somatosensory or visual spheres of crossmodal influence and determine how they might be appropriated.

5. Conclusions

Sensory loss, such as deafness, is well known to induce crossmodal changes involving the remaining sensory modalities. Many such studies have documented the presence of visual activation of secondary, but not core, auditory cortices following early deafness. The present study demonstrates that core auditory cortices also exhibit crossmodal reorganization following early hearing loss, but through the somatosensory modality. Because similar core auditory cortical effects occur as a result of early or adult hearing loss that do not conform with assumed mechanisms for crossmodal plasticity, a new brainstem theory of cortical crossmodal reorganization is proposed.

Conflict of Interests

The authors report no conflict of interests with this work.

Acknowledgments

The authors thank L. Keniston and S. Ramoa for assistance in programming and data collection and S. Shapiro and A. Rice for use of ABR equipment. This paper is supported by NIH Grants NS39460 (MAM) and R03DC011374 (BLA).

References

- [1] N. Sadato, A. Pascual-Leone, J. Grafman et al., "Activation of the primary visual cortex by Braille reading in blind subjects," *Nature*, vol. 380, no. 6574, pp. 526–528, 1996.
- [2] D. Bavelier and H. J. Neville, "Cross-modal plasticity: where and how?" *Nature Reviews Neuroscience*, vol. 3, no. 6, pp. 443–452, 2002.
- [3] K. Sathian and R. Stilla, "Cross-modal plasticity of tactile perception in blindness," *Restorative Neurology and Neuroscience*, vol. 28, no. 2, pp. 271–281, 2010.
- [4] H. Nishimura, K. Hashikawa, K. Doi et al., "Sign language "heard" in the auditory cortex," *Nature*, vol. 397, no. 6715, article 116, 1999.
- [5] L. A. Petitto, R. J. Zatorre, K. Gauna, E. J. Nikelski, D. Dostie, and A. C. Evans, "Speech-like cerebral activity in profoundly deaf people processing signed languages: implications for the neural basis of human language," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 25, pp. 13961–13966, 2000.
- [6] E. M. Finney, I. Fine, and K. R. Dobkins, "Visual stimuli activate auditory cortex in the deaf," *Nature Neuroscience*, vol. 4, no. 12, pp. 1171–1173, 2001.
- [7] N. Lambertz, E. R. Gizewski, A. De Greiff, and M. Forsting, "Cross-modal plasticity in deaf subjects dependent on the extent of hearing loss," *Cognitive Brain Research*, vol. 25, no. 3, pp. 884–890, 2005.
- [8] D. Bavelier, M. W. G. Dye, and P. C. Hauser, "Do deaf individuals see better?" *Trends in Cognitive Sciences*, vol. 10, no. 11, pp. 512–518, 2006.
- [9] P. Voss, F. Gougoux, R. J. Zatorre, M. Lassonde, and F. Lepore, "Differential occipital responses in early- and late-blind individuals during a sound-source discrimination task," *NeuroImage*, vol. 40, no. 2, pp. 746–758, 2008.
- [10] S. G. Lomber, M. A. Meredith, and A. Kral, "Cross-modal plasticity in specific auditory cortices underlies visual compensations in the deaf," *Nature Neuroscience*, vol. 13, no. 11, pp. 1421–1427, 2010.
- [11] M. A. Meredith, J. Kryklywy, A. J. McMillan, S. Malhotra, R. Lum-Tai, and S. G. Lomber, "Crossmodal reorganization in the early deaf switches sensory, but not behavioral roles of auditory cortex," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 21, pp. 8856–8861, 2011.
- [12] A. Kral, J. H. Schröder, R. Klinke, and A. K. Engel, "Absence of cross-modal reorganization in the primary auditory cortex of congenitally deaf cats," *Experimental Brain Research*, vol. 153, no. 4, pp. 605–613, 2003.
- [13] B. L. Allman, L. P. Keniston, and M. A. Meredith, "Adult deafness induces somatosensory conversion of ferret auditory cortex," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 14, pp. 5925–5930, 2009.
- [14] L. P. Keniston, B. L. Allman, and M. A. Meredith, "Hearing impairment in adult ferrets induces partial crossmodal conversion of core auditory cortex," *Society for Neuroscience*, vol. 41, article 476.13.
- [15] M. A. Meredith, L. P. Keniston, and B. L. Allman, "Multisensory dysfunction accompanies crossmodal plasticity following adult hearing impairment," *Neuroscience*, vol. 214, pp. 136–148, 2012.
- [16] T. D. Mrsic-Flogel, H. Versnel, and A. J. King, "Development of contralateral and ipsilateral frequency representations in ferret primary auditory cortex," *European Journal of Neuroscience*, vol. 23, no. 3, pp. 780–792, 2006.
- [17] D. R. Moore and J. E. Hine, "Rapid development of the auditory brainstem response threshold in individual ferrets," *Developmental Brain Research*, vol. 66, no. 2, pp. 229–235, 1992.
- [18] R. K. Shepherd and R. L. Martin, "Onset of ototoxicity in the cat is related to onset of auditory function," *Hearing Research*, vol. 92, no. 1-2, pp. 131–142, 1995.
- [19] S. A. Xu, R. K. Shepherd, Y. Chen, and G. M. Clark, "Profound hearing loss in the cat following the single co-administration of kanamycin and ethacrynic acid," *Hearing Research*, vol. 70, no. 2, pp. 205–215, 1993.
- [20] J. K. Bizley, F. R. Nodal, I. Nelken, and A. J. King, "Functional organization of ferret auditory cortex," *Cerebral Cortex*, vol. 15, no. 10, pp. 1637–1653, 2005.
- [21] P. R. Manger, G. Engler, C. K. E. Moll, and A. K. Engel, "Location, architecture, and retinotopy of the anteromedial lateral suprasylvian visual area (AMLS) of the ferret (*Mustela putorius*)," *Visual Neuroscience*, vol. 25, no. 1, pp. 27–37, 2008.
- [22] A. M. Ramsay and M. A. Meredith, "Multiple sensory afferents to ferret pseudosylvian sulcal cortex," *NeuroReport*, vol. 15, no. 3, pp. 461–465, 2004.
- [23] C. L. Veenman, A. Reiner, and M. G. Honig, "Biotinylated dextran amine as an anterograde tracer for single- and double-labeling studies," *Journal of Neuroscience Methods*, vol. 41, no. 3, pp. 239–254, 1992.
- [24] F. L. Rice, C. M. Gomez, S. S. Leclerc, R. W. Dykes, J. S. Moon, and K. Pourmoghadam, "Cytoarchitecture of the ferret suprasylvian gyrus correlated with areas containing multiunit responses elicited by stimulation of the face," *Somatosensory and Motor Research*, vol. 10, no. 2, pp. 161–188, 1993.
- [25] S. S. Leclerc, F. L. Rice, R. W. Dykes, K. Pourmoghadam, and C. M. Gomez, "Electrophysiological examination of the representation of the face in the suprasylvian gyrus of the ferret: a correlative study with cytoarchitecture," *Somatosensory and Motor Research*, vol. 10, no. 2, pp. 133–159, 1993.
- [26] P. R. Manger, G. Engler, C. K. E. Moll, and A. K. Engel, "The anterior ectosylvian visual area of the ferret: a homologue for an enigmatic visual cortical area of the cat?" *European Journal of Neuroscience*, vol. 22, no. 3, pp. 706–714, 2005.
- [27] P. R. Manger, I. Masiello, and G. M. Innocenti, "Areal organization of the posterior parietal cortex of the ferret (*Mustela putorius*)," *Cerebral Cortex*, vol. 12, no. 12, pp. 1280–1297, 2002.
- [28] J. Homman-Ludiye, P. R. Manger, and J. A. Bourne, "Immunohistochemical parcellation of the ferret (*Mustela putorius*) visual cortex reveals substantial homology with the cat (*Felis catus*)," *Journal of Comparative Neurology*, vol. 518, no. 21, pp. 4439–4462, 2010.
- [29] E. T. Auer, L. E. Bernstein, W. Sungkarat, and M. Singh, "Vibrotactile activation of the auditory cortices in deaf versus hearing adults," *NeuroReport*, vol. 18, no. 7, pp. 645–648, 2007.
- [30] W. A. Foxworthy and M. A. Meredith, "An examination of somatosensory area SIII in ferret cortex," *Somatosensory and Motor Research*, vol. 28, no. 1-2, pp. 1–10, 2011.
- [31] L. P. Keniston, B. L. Allman, M. A. Meredith, and H. R. Clemon, "Somatosensory and multisensory properties of the medial bank of the ferret rostral suprasylvian sulcus," *Experimental Brain Research*, vol. 196, no. 2, pp. 239–251, 2009.
- [32] S. E. Shore, "Plasticity of somatosensory inputs to the cochlear nucleus—implications for tinnitus," *Hearing Research*, vol. 281, no. 1-2, pp. 38–46, 2011.
- [33] J. P. Rauschecker, "Compensatory plasticity and sensory substitution in the cerebral cortex," *Trends in Neurosciences*, vol. 18, no. 1, pp. 36–43, 1995.

- [34] J. P. Rauschecker and U. Kniepert, "Auditory localization behaviour in visually deprived cats," *European Journal of Neuroscience*, vol. 6, no. 1, pp. 149–160, 1994.
- [35] B. Röder, W. Teder-Sälejärvi, A. Sterr, F. Rösler, S. A. Hillyard, and H. J. Neville, "Improved auditory spatial tuning in blind humans," *Nature*, vol. 400, no. 6740, pp. 162–166, 1999.
- [36] A. Fieger, B. Röder, W. Teder-Sälejärvi, S. A. Hillyard, and H. J. Neville, "Auditory spatial tuning in late-onset blindness in humans," *Journal of Cognitive Neuroscience*, vol. 18, no. 2, pp. 149–157, 2006.
- [37] C. C. Lee and J. A. Winer, "Connections of cat auditory cortex: I. Thalamocortical system," *Journal of Comparative Neurology*, vol. 507, no. 6, pp. 1879–1900, 2008.
- [38] C. C. Lee and J. A. Winer, "Connections of cat auditory cortex: II. Commissural system," *Journal of Comparative Neurology*, vol. 507, no. 6, pp. 1901–1919, 2008.
- [39] C. C. Lee and J. A. Winer, "Connections of cat auditory cortex: III. Corticocortical system," *Journal of Comparative Neurology*, vol. 507, no. 6, pp. 1920–1943, 2008.
- [40] E. Budinger, P. Heil, A. Hess, and H. Scheich, "Multisensory processing via early cortical stages: connections of the primary auditory cortical field with other sensory systems," *Neuroscience*, vol. 143, no. 4, pp. 1065–1083, 2006.
- [41] D. L. Hunt, E. N. Yamoah, and L. Krubitzer, "Multisensory plasticity in congenitally deaf mice: how are cortical areas functionally specified?" *Neuroscience*, vol. 139, no. 4, pp. 1507–1524, 2006.
- [42] S. E. Shore and J. Zhou, "Somatosensory influence on the cochlear nucleus and beyond," *Hearing Research*, vol. 216–217, no. 1–2, pp. 90–99, 2006.
- [43] S. E. Shore, Z. Vass, N. L. Wys, and R. A. Altschuler, "Trigeminal ganglion innervates the auditory brainstem," *Journal of Comparative Neurology*, vol. 419, no. 3, pp. 271–285, 2000.
- [44] P. O. Kanold and E. D. Young, "Proprioceptive information from the pinna provides somatosensory input to cat dorsal cochlear nucleus," *The Journal of Neuroscience*, vol. 21, no. 19, pp. 7848–7858, 2001.
- [45] L. M. Aitkin, C. E. Kenyon, and P. Philpott, "The representation of the auditory and somatosensory systems in the external nucleus of the cat inferior colliculus," *Journal of Comparative Neurology*, vol. 196, no. 1, pp. 25–40, 1981.
- [46] S. E. Shore, S. Koehler, M. Oldakowski, L. F. Hughes, and S. Syed, "Dorsal cochlear nucleus responses to somatosensory stimulation are enhanced after noise-induced hearing loss," *European Journal of Neuroscience*, vol. 27, no. 1, pp. 155–168, 2008.

Review Article

Visual Cortex Plasticity: A Complex Interplay of Genetic and Environmental Influences

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Received 16 December 2011; Revised 11 May 2012; Accepted 31 May 2012

Academic Editor: Ron Kupers

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The central nervous system architecture is highly dynamic and continuously modified by sensory experience through processes of neuronal plasticity. Plasticity is achieved by a complex interplay of environmental influences and physiological mechanisms that ultimately activate intracellular signal transduction pathways regulating gene expression. In addition to the remarkable variety of transcription factors and their combinatorial interaction at specific gene promoters, epigenetic mechanisms that regulate transcription have emerged as conserved processes by which the nervous system accomplishes the induction of plasticity. Experience-dependent changes of DNA methylation patterns and histone posttranslational modifications are, in fact, recruited as targets of plasticity-associated signal transduction mechanisms. Here, we shall concentrate on structural and functional consequences of early sensory deprivation in the visual system and discuss how intracellular signal transduction pathways associated with experience regulate changes of chromatin structure and gene expression patterns that underlie these plastic phenomena. Recent experimental evidence for mechanisms of cross-modal plasticity following congenital or acquired sensory deprivation both in human and animal models will be considered as well. We shall also review different experimental strategies that can be used to achieve the recovery of sensory functions after long-term deprivation in humans.

1. Introduction

As development proceeds, the nervous system begins to process information from the external world thus creating neuronal representations of the environment, which are continuously modified by sensory experience. Interactions with the external world, mediated by sensory input, update and modify the structural and functional architecture of the central nervous system, particularly during short-term periods in early life (known as critical periods) as experience drives the consolidation of synaptic circuitries [1, 2]. However, the reorganization of neuronal representations continues in adult life, as, for instance, in response to learning, loss of sensory input, trauma, or disease. The basis of the continuous and dynamic change in neuronal representations of sensory functions is a hot area of current neuroscience research with potential applications in the fields of neuronal regeneration, brain plasticity, and repair.

Long regarded as a rather static and unchanging structure, the adult brain has increasingly been recognized as a

system that retains a degree of plasticity that allows for a rewiring of neural networks over the entire life course. The visual system is a classical neurobiological paradigm in this context. Structural and functional modifications of neural circuitries in the visual cortex rely on a complex interplay between long-distance neuromodulatory systems [3–17], together with experience-dependent neuronal activity mediated by local inhibitory and excitatory neurotransmission [18–24], neurotrophic factors [15, 16, 25–29], extracellular matrix molecules [30–35], hormones [36, 37], and endocannabinoids [38]. A complex interaction between these physiological processes seems to set in motion intracellular signal transduction pathways [39–41] that eventually promote the expression of transcription factors and downstream target genes that mediate phenomena of plasticity. The result is a highly dynamic architecture of the brain that is continuously modified by sensory experience. In fact, the reorganization of cortical circuitries persists late in life, at least to some extent (for review see [42]). Achieving a

fundamental understanding of physiological processes that lie behind neuronal plasticity may be of clinical relevance in pathological states where the reorganization of neuronal networks would be beneficial in adult life.

The importance of sensory experience in development of the human brain is well exemplified by cases of strabismic or anisometric children that underwent no clinical treatment during early development. In either pathological condition proper visual experience is altered, causing a marked impairment of normal visual functions (amblyopia) that is irreversible if not treated before 8 years of age [43, 44]. A similar phenomenon is described by clinical observations of children born with congenital cataracts, a pathological condition in which the lens of the eye becomes milky and therefore no longer permits images to form on the retina [45, 46]. Although amblyopia can be prevented by different therapeutic strategies that restore the formation of proper retinal images or by eye patching in early life, such treatments are normally ineffective in adults [47, 48]. Therefore, the recovery of normal visual functions after long-term sensory deprivation has long been a subject of study with the prospect of finding therapies for human amblyopia in adulthood. These observations indicate that proper sensory experience during early stages of development is necessary for normal sensory perception and also point towards the enhancement of neuronal plasticity as a strategy for brain repair in adult life.

2. Early Sensory Deprivation and Visual Cortical Plasticity

Although intrinsic factors drive the initial assembly of synaptic circuitries in the nervous system, neuronal networks are shaped by experience during early postnatal life. Once basic patterns of neural connections are formed, an experience-dependent organization of eye-specific inputs is the major mechanism by which synaptic connectivity is achieved in the developing visual cortex.

The monocular deprivation paradigm is a classic model to assess neuronal plasticity in the visual system. Pioneering electrophysiological studies in cats and monkeys clearly demonstrated that short periods of visual deprivation by unilateral eye closure during early development cause structural and functional modifications in visual cortical circuitries. Visual cortex responsiveness actually shifts in favour of the normal eye after monocular deprivation during the critical period [49–52]. Furthermore, the deprived eye becomes amblyopic: its visual acuity (spatial resolution) and contrast sensitivity are markedly impaired [53–55]. At structural level, unilateral eyelid suture causes a reduction in the arborisation of geniculocortical terminals that serve the deprived eye, which parallels an increased spread of terminals serving the open eye [51] and is consistent with the fact that monocular deprivation impairs the spatial resolution of geniculate neurons [56]. Because this type of deprivation does not cause amblyopia in adulthood, this early temporal window characterized by an enhanced brain susceptibility to sensory experience is a typical example of a critical period. It is interesting to note that most cortical cells remain

responsive to both eyes following a period of binocular deprivation in juvenile age. Thus, it appears that afferents from the two eyes compete for cortical territory and that the relative amount of activity in the two eyes determines the outcome of this competitive process.

3. Nonvisual Components of the Environment Alter Visual System Development

Another classical paradigm used to assess the impact of experience in the functional maturation of the visual system is dark rearing (i.e., rearing animals in total darkness from birth). Total absence of visual experience delays the functional maturation of the striate cortex [57–59]. This event seems to be mediated by a downregulation of BDNF expression in early life [60], which results in a retarded maturation of GABAergic circuitries that control functional development of the visual system (for review see [20]). The spatial resolution of visual cortical neurons is actually reduced in dark-reared animals, this phenomenon being accompanied by longer latencies of responses to visual stimuli [58, 59]. Additionally, rearing animals in complete darkness extends the critical period far beyond its normal limits [61].

Although early studies of the visual system showed that sensory experience after eye opening is necessary for the functional maturation of the visual cortex [49–52], the notion that nonvisual components of the environment influence development of the visual system has been increasingly appreciated in the last few years. Experiments that combine dark rearing and electrophysiology as a functional readout, in transgenic mice overexpressing BDNF in forebrain regions, revealed a remarkable and unexpected finding: visual cortical neurons in these animals responded normally to visual stimuli, indicating that they could see well despite the lack of visual experience during the critical period [62]. These observations are in consonance with the fact that environmental enrichment, an experimental condition characterized by an increased exploratory behavior and sensory-motor stimulation, prevents the effects of dark rearing in the visual system [63]. This effect has been ascribed to an increased BDNF signaling and enhanced GABAergic inhibition during early stages of development [63]. Of note, environmental enrichment in normally reared animals accelerates the functional development of the visual cortex, this phenomenon being accompanied by alterations in the expression of BDNF and GABA synthesizing enzymes well before eye opening [64]. These findings further suggest that environmental influences on visual system development are, at least, partially independent of visual experience.

4. Physiological Mechanisms That Regulate Developmental Plasticity in the Visual System

4.1. The Maturation of Inhibitory Circuitries Controls the Time Course of the Critical Period. The experience-dependent maturation of GABA-mediated inhibition during development establishes the beginning of the critical period for

plasticity in the visual system [18–20]. This was demonstrated by seminal electrophysiological studies in transgenic mice that lack one isoform of the GABA synthesizing enzyme GAD65 and therefore show reduced levels of intracortical inhibition. No variation of visual cortex responsiveness was observed after monocular deprivation during early life in GAD65 transgenic animals, whereas enhancing inhibition by means of GABA-A receptor agonists rescued the impairment of plasticity [18, 19]. Therefore, a reduction of inhibitory transmission in early life halts the onset of the critical period for visual cortex plasticity (for review see [20]). A second inhibitory threshold that causes the end of the critical period is reached over postnatal development as well. Transgenic animals that overexpress BDNF in forebrain regions display an accelerated maturation of intracortical inhibitory circuitries, which, in turn, causes a precocious development of the visual system and therefore a fast end of the critical period for ocular dominance plasticity [29]. In summary, an initial threshold of inhibition triggers a sensitive period in which neuronal networks in the visual system are highly susceptible to sensory experience, whereas a second inhibitory threshold signals the end of this phase of enhanced plasticity.

Inhibition triggers plasticity through GABA-A receptors containing the alpha-1 subunit [[21], reviewed in [20]]. These receptors are enriched at somatic synapses on pyramidal neurons made by large basket cells (a class of parvalbumin-positive GABAergic interneurons that extend horizontally across ocular dominance columns). Recent studies indicate that visual experience controls the time course of the critical period by promoting the transfer of the homeoprotein Otx2 from the retina to the visual cortex, where it appears to promote the maturation of parvalbumin-positive GABAergic interneurons [22]. Indeed, a reduction of inhibitory transmission is observed in visual cortical slices from Otx2-knockout animals, suggesting that Otx2 is a retinal messenger that triggers the critical period by enhancing levels of inhibition. Moreover, intracortical delivery of the recombinant Otx2 protein in animals before the onset of the critical period (in which no shift of ocular dominance is observed after eyelid suture due to low levels of inhibition) renders the visual cortex sensitive to monocular deprivation [22]. Accordingly, the impairment of plasticity in Otx2-knockout animals is rescued by enhancing GABA-A receptor currents by benzodiazepine treatments [22]. These findings suggest that visual experience signals the time course of the critical period by activating the retinogeniculocortical transfer of the protein Otx2 in the visual pathway.

4.2. Extracellular Matrix Molecules Restrict Plasticity in the Developing Visual System. An emerging view in neuronal plasticity research is that the effects caused by early sensory experience in the remodeling of visual cortical circuitries are actively preserved throughout life by the late appearance of molecular factors in the extracellular milieu that restrict plasticity. The establishment of neuronal connectivity may be, at least in part, under control of structural factors such as myelin-associated proteins (NgR, PirB) and chondroitin sulphate proteoglycans (CSPGs), which all are inhibitory

for axonal sprouting [30–32, 65, 66]. Different experimental findings support this notion. The maturation of intracortical myelination, for instance, correlates with the end of the critical period and ocular dominance plasticity persists well into adulthood in NgR-knockout mice [30]. Knockout animals lacking the NgR ligand Nogo-A also display plastic phenomena in adult life, thus confirming that NgR-dependent mechanisms restrict plasticity in the visual system. Additionally, the paired immunoglobulin-like receptor B (PirB) shows high affinity for Nogo-A, the signaling of which is inhibitory for axonal regeneration [31]. In keeping with this, PirB restricts ocular dominance plasticity in the developing visual cortex [32].

Likewise, the condensation of CSPGs around the soma and dendrites of parvalbumin-positive GABAergic interneurons parallels the time course of the critical period, whereas CSPGs degradation by exogenous administration of the enzyme chondroitinase-ABC reactivates visual cortex plasticity in the adult [34, 35]. This is consistent with the notion that removing extracellular matrix components that are inhibitory for axonal growth [65, 66] provides a permissive environment for structural plasticity (e.g., by modifying dendritic spine dynamics) and associated functional modifications in the visual cortex. It is important to remark that degradation of CSPGs may alter the ratio of inhibitory/excitatory transmission in the visual cortex as these glycoproteins condense in perineuronal nets (PNNs) mainly around parvalbumin-positive GABAergic interneurons. So far, however, the impact of chondroitinase-ABC treatment on the intracortical inhibitory/excitatory balance in the visual system remains to be explored.

4.3. Long-Distance Neuromodulatory Systems Regulate Visual Cortical Plasticity. The major modulatory systems in the brain (i.e., adrenaline, noradrenaline, dopamine, acetylcholine, and serotonin) regulate complex functions of the central nervous system such as different forms of brain plasticity, cognitive processes, and behavior. Experience-dependent modifications of cortical circuitries are not determined solely by local correlations of electrical activity but are also influenced by attentional mechanisms. Sensory signals, for instance, promote marked modifications of neural circuitries mainly when animals attend to the sensory input and use this information for the control of behavior (reviewed in [67, 68]). Accordingly, early studies performed in kittens demonstrated that changes of visual cortical circuitries in response to experience are lessened when noradrenergic [3, 4], cholinergic [4, 5], and serotonergic [6–8] projections to the cortex are inactivated. Moreover, there is evidence that these neuromodulatory systems mediate forms of visual cortex plasticity late in life both in cats [9–13] and rodents [14–17].

Advances in the understanding of mechanisms by which neuromodulatory systems regulate experience-dependent plasticity derive from *in vitro* studies of synaptic plasticity. There is evidence that noradrenaline, acetylcholine, and serotonin modulate two different forms of activity-dependent synaptic modifications: long-term potentiation (LTP) and long-term depression (LTD). In the visual system,

LTP and LTD can be induced by different patterns of electrical stimulation. Brief and strong episodes of high frequency stimulation promote LTP while prolonged low-frequency stimulation yields LTD. In the rodent visual cortex, upon administration of noradrenaline and acetylcholine, weaker tetanic stimulation is required to induce LTP and shorter episodes of low frequency stimulation are needed to drive LTD [69, 70]. Likewise, serotonin facilitates the induction of both LTP and LTD in layer IV of the kitten visual system [8]. These findings are consistent with a role for neuromodulatory systems as enabling factors for visual cortical plasticity and indicate that activation of noradrenergic, cholinergic, and serotonergic receptors lowers the threshold of activity required for the induction of LTP and LTD. Intracellular mechanisms whereby neuromodulatory systems facilitate these forms of synaptic plasticity have been subject of extensive study. The induction of LTP and LTD requires the activation of N-methyl-D-aspartate (NMDA) receptors together with a postsynaptic rise in intracellular calcium. The available evidence is consistent with a model in which the magnitude and duration of the calcium signal determines the magnitude of the synaptic modification [71]. Brief and large calcium influxes induce LTP, whereas smaller and prolonged calcium increases yield LTD. Of note, receptors of these three major neuromodulatory systems are able to activate the IP3 second messenger pathway, which can induce calcium release from intracellular stores and therefore modulate plasticity. Because the intracortical inhibitory/excitatory balance regulates experience-dependent plasticity in the visual system (for review see [20, 24]), the neuromodulators-mediated fine-tuning of the inhibitory/excitatory ratio is likely to play a key role in the induction of plastic phenomena. Accordingly, it has been recently demonstrated that the enhanced signaling of either serotonin or acetylcholine shifts the inhibitory/excitatory balance in favour of excitation in the rodent visual cortex [72, 73].

The critical period for the induction of LTP evoked by stimulation of thalamocortical connections almost overlaps the duration of the critical period for ocular dominance plasticity. In addition to thalamocortical connections, LTP and LTD can be elicited by stimulation of intrinsic connections both during postnatal development and in adulthood and these forms of plasticity are only partially dependent on NMDA receptors [74]. LTP in layer II/III cells is facilitated by concomitant application of muscarinic and noradrenergic agonists but not by the single application of each neurotransmitter [69]. Accordingly, exogenous administration of acetylcholine in visual cortical slices induces LTP through stimulation of muscarinic acetylcholine receptors (mAChRs) [75]. Moreover, LTP is impaired in visual cortex slices from transgenic mice with reduced cortical cholinergic innervation due to the expression of an anti-NGF antibody [76]. Exogenous application of acetylcholine, however, rescues LTP suggesting an essential role of this neurotransmitter in cortical synaptic plasticity. In agreement with this notion, immune depletion of basal forebrain cholinergic neurons with IgG-192 saporin impairs LTP in the visual cortex [77]. Furthermore, it has been reported that visual cortex LTP and LTD are modulated by the activation of different mAChRs

[78]. Using single and double muscarinic receptor knockout mice, it has been demonstrated that normal LTP is expressed when M2 and M4 are coactivated while LTD relies more on M1 and M3 receptor. Moreover, while prolonged low-frequency stimulation normally induces LTD, it does yield LTP in M1-knockout animals. These findings suggest that the direction of synaptic plasticity can be modulated by the combined activity of different mAChRs, possibly by regulating the threshold for synaptic modification.

5. The Reinstatement of Plasticity in the Adult Visual System

The identification of molecular and cellular mechanisms at the basis of brain plasticity and the enhancement of plasticity as a strategy for brain repair in adult life are hot areas of current neuroscience research. As previously described, the developmental maturation of intracortical inhibitory circuitries causes the end of plasticity in the visual system (reviewed in [20]). In keeping with this notion, it is possible to restore plasticity in adult life by reducing levels of inhibition. A direct demonstration that GABAergic signaling is a crucial brake limiting visual cortex plasticity derives from the observation that a pharmacological decrease of inhibitory transmission effectively restores ocular dominance plasticity in adulthood [23]. Accordingly, experimental paradigms such as dark exposure [79, 80], environmental enrichment [17, 81, 82], food restriction [36], long-term fluoxetine treatment [15, 16], and exogenous IGF-I administration [83] all promote plasticity late in life by reducing the intracortical inhibitory/excitatory ratio (Figure 1). This has prompted the search for endogenous factors with the potential to enhance plasticity in adult life by modulating the intracortical I/E balance.

Previous studies have demonstrated that the process of plasticity reactivation in the adult visual system is a multifactorial event that comprises the action of different cellular and molecular mechanisms, working in parallel or in series, the sum of which results in the activation of intracellular signal transduction pathways regulating the expression of plasticity genes [14–17, 34–36, 83–86], [for review see [42]]. In rodents, experimental paradigms based upon the enhancement of environmental stimulation levels, genetic manipulations, and pharmacological treatments have revealed that the enhanced action of either long-distance projection systems (e.g., serotonergic and cholinergic transmission) or IGF-I signaling seems to modulate the intracortical inhibitory/excitatory balance in favour of excitation [72, 73, 83], which in turn, sets in motion cellular and molecular events that eventually mediate the expression of genes associated with functional modifications in the adult visual system (Figure 2). The reinstatement of plasticity caused by enhanced serotonergic transmission, for instance, is mediated by 5-HT_{1A} receptors signaling and accompanied by increased BDNF expression [16]. This is paralleled by heightened histone acetylation status at the activity-dependently regulated BDNF promoter regions and by decreased expression of histone deacetylase enzymes (HDACs) [16]. In keeping with this, increasing histone

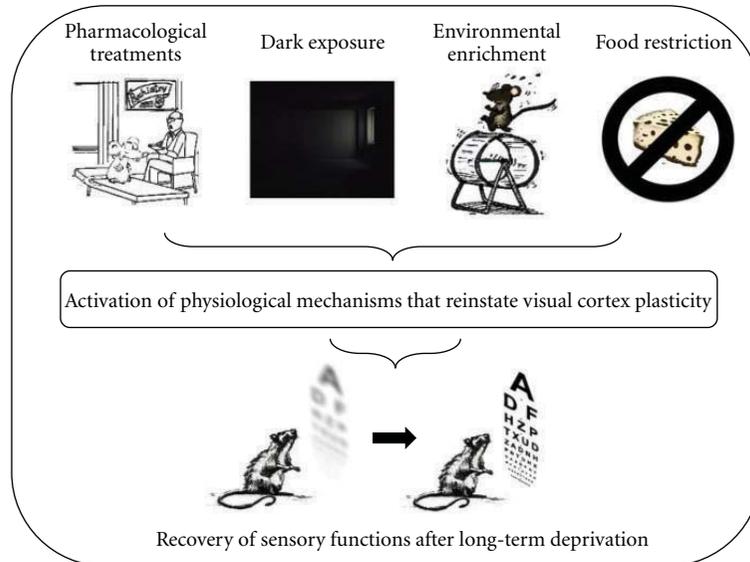


FIGURE 1: Experimental paradigms that restore neuronal plasticity in adult life. Environmental enrichment [17, 81, 82], long-term fluoxetine administration [15, 16], visual deprivation by dark exposure [79, 80], food restriction [36], and IGF-1 treatment [83] are noninvasive experimental approaches that promote adult visual cortical plasticity by altering the balance of inhibition and excitation in the visual system. The potential for the reactivation of plasticity caused by some of these paradigms to promote the recovery of sensory functions after long-term sensory deprivation has been reported using amblyopia as a paradigmatic model [15, 36, 80, 81, 83].

acetylation levels by long-term treatment with HDACs inhibitors (e.g., trichostatin-A, valproic acid, and sodium butyrate) not only reinstates ocular dominance plasticity in adulthood [16, 84] but also promotes full recovery of visual functions in adult amblyopic animals [85]. Accordingly, environmental enrichment, long-term fluoxetine treatment, and food restriction all increase acetylation of histones in the hippocampus and cortex in adult life [16, 36, 87].

6. Structural Plasticity in the Visual Cortex

Experience-dependent functional modifications in the visual system are accompanied by a structural remodeling of synaptic connectivity, in terms of growth and loss of dendritic spines. Dendritic spines in pyramidal neurons are markedly sensitive to experience. Total lack of visual experience in early life induces modifications in spine morphology and density, both of which are partially reversible by light exposure [88]. Accordingly, monocular deprivation in early life alters the motility, turnover, number, and morphology of dendritic spines in the visual cortex [89–92].

Does structural plasticity contribute to experience-dependent modifications of neural circuitries? Structural plasticity *in vivo* studies, using two-photon imaging, indicate that dendritic spine dynamics is high during early postnatal life but decreases thereafter, in parallel to the time course of critical period plasticity over development (reviewed in [93]). This suggests that, despite the absence of large-scale structural remodeling, the reorganization of cortical connections in terms of growth and loss of dendritic spines may be the structural substrate for experience-dependent plasticity.

Notably, chronic imaging experiments have recently demonstrated that changes in visual cortex responsiveness after monocular deprivation during the critical period correlate with dendritic spines structural modifications across the visual cortex, these two features being reversed when the deprived eye is reopened. After brief periods of monocular deprivation, spine turnover increases significantly, with a larger percentage of spines being lost rather than gained, whereas after a 24-hour period of recovery (visual experience) the total number of dendritic spines is reestablished [94]. Accordingly, increasing the density and dynamics of spines by intracortical infusion of the bacterial toxin CNF1 restores a degree of plasticity in the mature cortex that is similar to that observed during early postnatal life [95].

It is worth mentioning that new synapses formation may increase memory storage capacity of the brain and that new dendritic spines may serve as structural traces for earlier memories, enabling the brain for faster adaptations to similar future experiences [91, 96]. Recent experiments carried out using the monocular deprivation paradigm seem to confirm this notion. Modifications of dendritic spines caused by a first experience of unilateral eyelid suture persist even after restoration of binocular vision and may therefore be involved in the enhancement of plasticity observed after a second episode of visual deprivation [91].

The imaging studies mentioned above raise the question of whether structural modifications of dendritic spines represent functional changes of synaptic transmission. Electrophysiological experiments in hippocampal slice cultures indicate that AMPA- and NMDA-type glutamate receptor currents of newborn spines resemble those of mature synaptic contacts [97]. It has been recently demonstrated that dynamics of dendritic spine development regulates

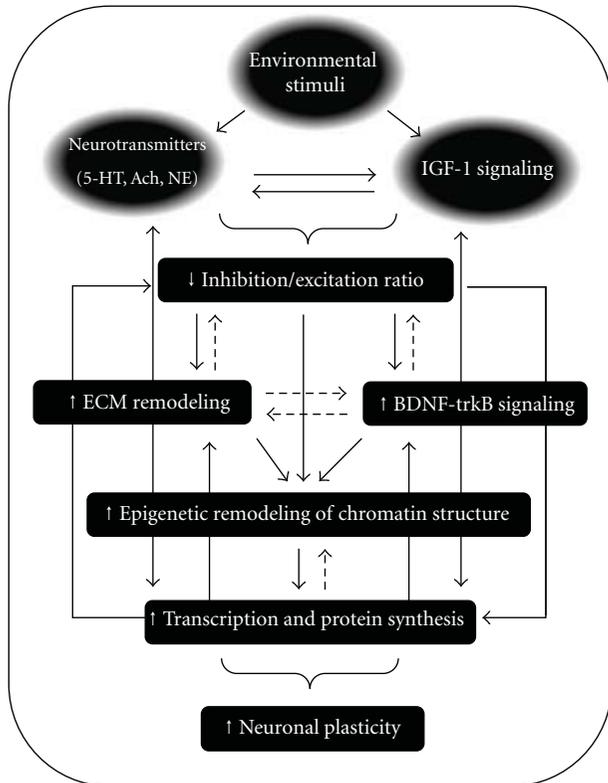


FIGURE 2: The reinstatement of ocular dominance plasticity in adulthood is associated with signal transduction pathways that involve the enhanced action of either neuromodulatory projection systems (e.g., serotonin and acetylcholine) or IGF-1 signaling, which all set in motion physiological processes that modulate the inhibitory/excitatory ratio in favour of excitation [14–17, 72, 73, 83]. A shift of the inhibitory/excitatory balance may directly activate intracellular mechanisms that eventually promote epigenetic modifications of chromatin structure (e.g., changes of DNA methylation patterns and/or posttranslational modifications of histones), which in turn allow for the expression of genes that act as downstream effectors of plastic phenomena in adult life. A pharmacological reduction of intracortical inhibition enhances plasticity while promoting the activity-dependent BDNF expression (unpublished data) and degradation of extracellular matrix (ECM) components that are inhibitory for plasticity [23]. BDNF-trkB signaling might upregulate the expression of additional genes associated with functional modifications in the visual cortex. Degradation of ECM components (e.g., CSPGs) may modify the inhibition/excitation ratio in the visual system. The interaction between BDNF-trkB signaling and ECM reorganization has yet to be explored. Continuous arrows represent established interactions between the molecular and cellular processes mentioned (boxes). Dashed lines represent interactions that remain to be ascertained.

the stability of synaptic plasticity. The relationship between calcium influx and spine size actually determines the long-term synaptic stability and synaptic strength distribution in synapses of hippocampal CA3-CA1 pyramidal neurons [98].

7. Epigenetic Mechanisms of Plasticity

Long-term functional modifications of neural circuitries are mediated by a complex interplay between cellular and

molecular mechanisms that activate intracellular signal transduction pathways regulating gene expression. Besides the remarkable diversity of transcription factors and their combinatorial interaction at gene promoter areas, the role of epigenetic mechanisms that control chromatin susceptibility to transcription in response to experience has been increasingly appreciated [99]. Growing experimental evidence indicates that chromatin structure is highly dynamic within the nervous system and that it is recruited as a target of plasticity-associated signal transduction pathways. The remodeling of chromatin structure is actively involved in activity-dependent neuronal plasticity in different brain areas via regulation of gene expression [100, 101].

Processes of chromatin remodeling that modulate gene transcription are conserved mechanisms by which the mammalian nervous system accomplishes adaptive behavioral responses upon environmental demands. In rodents, maternal care seems to influence behavioral and endocrine responses to stress in the offspring by modifying chromatin susceptibility to gene expression. It has been demonstrated that rat pups that are most licked and groomed during postnatal development, display, later in life, better performance in tests of learning and memory than pups that get licked less [102]. Interestingly, high licking/grooming pups are less anxious than low licked/groomed counterparts, and this behavior seems to be epigenetic rather than inherited. The genetic reprogramming by maternal behavior actually emerges over the first week of postnatal life and can be reversed by cross-fostering: if high licking/grooming dams rear the biological offspring of low licking/grooming ones, the offspring actually behave as high licking/grooming pups [103]. At molecular level, maternal care promotes the expression of the glucocorticoid receptor, this phenomenon being accompanied by a decreased DNA methylation status at the glucocorticoid receptor gene promoter area in the hippocampus [103]. Moreover, changes in the pattern of DNA methylation correlate with modifications at the level of histones, as high licking/grooming pups show enhanced levels of acetylation of lysine 9 on histone 3 (H3K9), which is a marker of gene transcription activation. This is consistent with the observation that increasing histones acetylation by hippocampal infusion of the histone deacetylase (HDAC) inhibitor Trichostatin-A in low licking/grooming pups changes the methylation pattern to that of pups brought up by high licking/grooming dams. Furthermore, low licking/grooming pups treated with Trichostatin-A are also less anxious than vehicle-treated counterparts and show no difference at behavioral level as compared to high licking/grooming pups [103]. These findings illustrate the notion that sensory experience in early life drives epigenetic mechanisms of neuronal plasticity that underlie behavior.

Similarly, phenomena of plasticity in the visual cortex of cats and rodents during the critical period require the activation of different intracellular protein kinases (e.g., PKA, ERK1/2, and CamKII) [39–41]. The activation of these intracellular signal transduction pathways promotes the upregulation of transcription factors that, in turn, mediate gene expression. A very well-known activity-dependent mechanism is the activation of the transcription factor

CREB, which triggers the expression of genes under control of the cAMP-response element (CRE) promoter, thus allowing phenomena of plasticity to occur [104, 105]. These plastic events involve processes of chromatin remodeling. Visual experience during early life promotes modifications of chromatin structure that are permissive for transcription, whereas a developmental downregulation of histone post-translational modifications regulates the closure of the critical period in the mouse visual system [84]. Accordingly, directly increasing acetylation of histones by long-term treatment with HDACs inhibitors effectively reactivates plasticity in the adult visual system [16, 84, 85].

8. Short Noncoding mRNAs and the Regulation of Plasticity

In addition to the function of transcription factors and modifications of chromatin structure, growing experimental evidence supports a critical role for short noncoding RNAs (microRNAs), which interact with and control translation of mRNA targets, in the regulation of gene expression patterns at the basis of plastic phenomena in the mammalian nervous system [106, 107]. MicroRNAs are powerful regulators of gene expression and act by binding to the 3'-untranslated region (3'-UTR) of the target mRNA, making it possible for a single microRNA to control expression of multiple genes that possess the same sequence in this region of the mRNA.

The brain-specific microRNA, miR-134, for instance, has been found to localize in the synaptodendritic compartment of rat hippocampal neurons and negatively regulates the size and density of dendritic spines [108]. This effect seems to be achieved by miR-134 posttranscriptional inhibition of the mRNA that encodes the protein kinase, Limk1, which controls dendritic spine development. This was demonstrated by refined experiments in which miRNA-134 was overexpressed in hippocampal neurons together with constructs expressing either a wild-type Limk1 mRNA or a mutant Limk1 mRNA that is incapable of interacting with miRNA-134. The study of spine morphology revealed that coexpression of the wild-type Limk1 mRNA, which is still subject to miRNA-134 translational inhibition, caused a decreased spine size phenotype. In contrast, expression of the mutant Limk1 mRNA that is incapable of interacting with miRNA-134 rescued the spine defect [108]. Hence, both overexpression of miRNA-134 and disruption of Limk1 function lead to decreased spine size. Accordingly, exposure of neurons to neurotrophins such as BDNF, which promotes synaptic development, maturation, and plasticity, relieves miRNA-134 inhibition of Limk1 mRNA translation [108]. These findings indicate that miR-134 disrupts dendritic and synaptic development by repressing Limk1 mRNA translation.

Recent experimental evidence points toward a key role for another microRNA, miRNA-132, as a molecular transducer of neuronal plasticity. It has been reported that synaptic activity promotes a CREB-dependent miRNA-132 expression and that miRNA-132 induction is necessary for the activity-dependent dendritic growth [109]. The effect of miRNA-132 on dendrite morphology seems to be mediated

by the activation of the Rac1-PAK actin-remodeling pathway that is due to the miRNA-132 translational inhibition of the mRNA that encodes the protein p250GAP, which is a Rho family GTPase activating protein [110]. This is consistent with the observation that overexpression of miRNA-132 in neuronal cultures promotes neuronal morphogenesis [111] and is in line with the fact that transgenic mice overexpressing miRNA-132 in forebrain regions display an increased spine density [112]. Interestingly, downstream target genes regulated by miRNA-132 mediate phenomena of chromatin remodeling and protein translation in the suprachiasmatic nucleus of rodents [113], these two molecular processes being critically involved in the occurrence of neuronal plasticity.

Electrophysiological studies have addressed the role of microRNAs as mediators of synaptic plasticity at hippocampal level. LTP of synaptic transmission in the dentate gyrus of rodents is accompanied by an upregulation of miRNA-132 [114], while its overexpression in cortical neurons regulates short-term plasticity [115]. More recently, evidence for the role of miRNA-132 as a mediator of visual cortical plasticity has been obtained *in vivo* by using the experience-dependent monocular deprivation paradigm. It has been reported that miRNA-132 is rapidly upregulated after eye opening in normally reared animals. This phenomenon is delayed by dark rearing, whereas monocular deprivation in early life results in a decrease of miRNA-132 expression. Remarkably, reducing miRNA-132 neonatal expression by lentiviral infection [116] or counteracting the miRNA-132 downregulation in response to monocular deprivation [117] effectively prevents ocular dominance plasticity in the developing visual system. These data highlight the notion that optimal physiological levels of miRNA-132 are critical for plasticity to occur during the critical period. Interestingly, neonatal blockade of miRNA-132 expression in early life results in an immature state of dendritic spines [116], whereas counteracting the miRNA-132 downregulation after monocular occlusion increases the percentage of mushroom-stubby dendritic spines that represent the more stable state of spines [117]. These findings suggest that miRNA-132 is a molecular transducer of the action of visual experience on developing visual circuitries, possibly acting through modulation of dendritic spines plasticity [118].

9. Cross-Modal Plasticity: Adaptive Reorganization of Neural Networks in Early Life

Sensory deprivation in one modality during early stages of development can have marked effects on the development of the remaining modalities. This phenomenon is known as cross-modal plasticity and is particularly epitomized by cases of congenital blindness or deafness from birth. In such instances, processes of cross-modal plasticity strengthen other sensory systems to compensate for the lack of vision or hearing.

Although clinical studies of deaf and blind humans have clearly demonstrated increased functional capabilities and

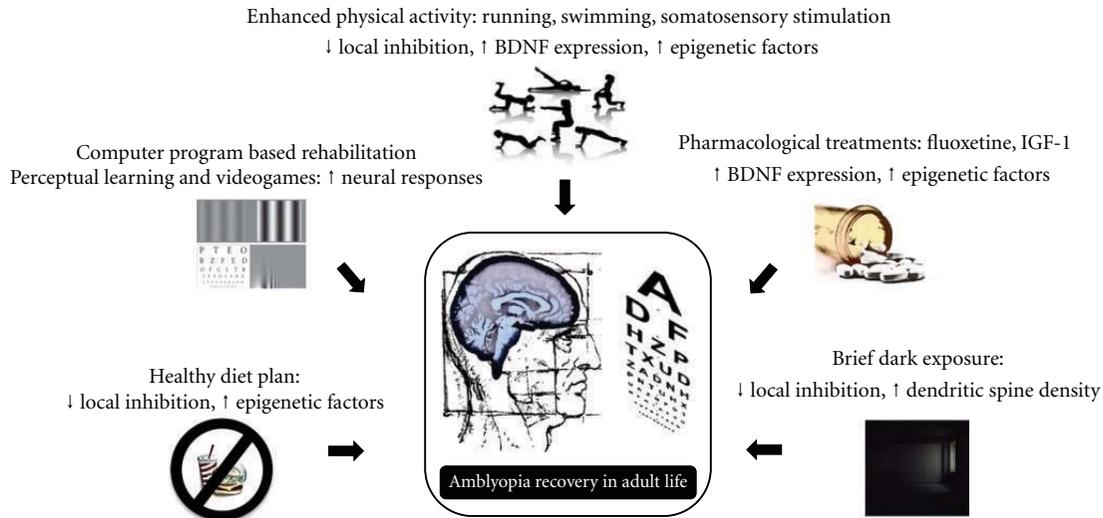


FIGURE 3: Potential strategies for the treatment of human amblyopia in adult life. The recent findings that environmental enrichment [17, 81, 82, 137, 138], long-term administration of fluoxetine [15, 16], dark exposure [79, 80], food restriction [36], and IGF-1 signaling [83] all promote full recovery of visual acuity and binocularity in adult amblyopic animals, emphasize the potential of different pharmacological and/or behavioral interventions as complementary strategies for current therapies of human amblyopia in adult life. In particular, an enhanced sensory-motor activity together with a healthy diet planning, brief periods of visual deprivation by dark exposure, and pharmacological treatments (long-term antidepressant treatment or exogenous IGF-1 administration) may enhance plasticity by shifting the I/E ratio while increasing BDNF expression and epigenetic factors. These therapeutic interventions could be coupled to video game playing or computer-program-based training of the amblyopic eye in order to rescue normal visual functions after long-term sensory deprivation in humans.

compensatory expansion in the remaining sensory modalities (reviewed in [119]), the neurological bases for these plastic phenomena remain poorly understood. It has been reported that congenitally blind subjects show better sound localization abilities as compared to sighted individuals [120] and display better two-point tactile discrimination skills as well [119]. Studies that combine Braille reading and functional brain imaging revealed that early blind individuals show a strong activation of the occipital cortex during the reading task [121, 122], this phenomenon being independent of attentional mechanisms [122]. Activation of the visual cortex has also been reported during the tactile object recognition task [123]. Remarkably, the inactivation of the visual cortex by means of transcranial electrical stimulation in blind people during Braille reading not only distorts tactile perceptions of blind subjects but also induces errors in Braille reading [124]. Furthermore, the visual cortex in blind subjects is recruited by language processing (e.g., semantic and phonological tasks) [125–127]. There is also evidence that congenital blindness enables visual circuitries to contribute to olfactory processing [128].

The question of whether there is a critical period for cross-modal plasticity has also been addressed by examining the activation of visual cortical areas by Braille reading in early and late-onset blind individuals. It has been reported that visual cortex responsiveness to somatosensory stimuli (Braille reading) is higher in congenitally blind and early-onset subjects as compared to the late-onset blind group [129–132]. These data indicate that there is a critical period for the visual cortex to be recruited to a role in the

processing of somatosensory information, which does not extend beyond 14 years of age in humans. An important question that remains to be answered concerns structural and functional mechanisms whereby phenomena of cross-modal plasticity occur. It has been reported that stabilization of long-range cortico-cortical connections between sensory modalities may mediate, at least, some aspects of these plastic phenomena [133]. Such cross-modal connections have been described in several species. Anatomical evidence for direct connections between primary auditory cortex and primary visual cortex in adult monkeys has been previously reported [133].

Phenomena of cross-modal plasticity have also been observed in the brain of deaf subjects. Functional magnetic resonance imaging studies have demonstrated that early deaf individuals use the primary auditory cortex alongside the visual system when they observe sign language [134]. Although there is no hearing component to sign language, the auditory cortex is instead used to assist with visual and language processing. The effects of cochlear implants also provide another strategy to assess cross-modal plasticity in the deaf. Early deaf individuals, but not late-onset deaf subjects, actually display impairments in their ability to process language using a cochlear implant in adult life as the auditory cortex has been reshaped to deal with visual information and therefore it cannot deal as well with the new sensory input that the implant provides [135].

A recent series of experiments using environmental enrichment [136] as a strategy to investigate the influence of sensory experience on brain development, and in particular

the somatosensory stimulation in terms of body massage, provide evidence that mechanisms of cross-modal plasticity are likely to underlie the beneficial effects of enhancing somatosensory activity in development of another sensory modality, the visual system [137]. It has been reported that an enriched environment accelerates the structural and functional development of the rodent visual system [64] and that enriching the environment in terms of tactile stimulation (body massage with a soft toothbrush) in rat pups effectively mimics the effects of enrichment on visual system development [137]. The massage protocol in the offspring of rats accelerated the maturation of visual functions and increased circulating levels of IGF-1, whereas antagonizing IGF-1 signaling by systemic injections of JB1 (IGF-1 receptor antagonist) prevented the effects of massage [137]. Remarkably, enriching the environment in terms of body massage in human preterm infants accelerates the maturation of the visual system as indicated by an enhanced development of spatial acuity, this effect being correlated to high IGF-1 serum levels [137]. Taken together, these findings indicate that processes of cross-modal plasticity may be involved in the effects caused by environmental enrichment in visual cortex development and portray the well-characterized visual system as a model to understand the functional integration of two or more sensory modalities.

10. Can We Treat Human Amblyopia in Adult Life?

The potential clinical application of experimental strategies that promote plasticity in the adult visual system has long been explored with the prospect of treating human amblyopia, a pathological condition that arises from an abnormal visual experience during development and is refractory treatment in the adult (for review see [43, 47]). The data on animal models reported along this paper suggest that an enhanced sensory-motor activity [17, 137, 138], a healthy diet planning [36], brief periods of dark exposure [79, 80], fluoxetine administration [15, 16], and IGF-1 treatment [83] may be used as complementary strategies to current therapies for human amblyopia (Figure 3). Clinical trials that include pharmacological and behavioral interventions by long-term fluoxetine treatment together with a computer-program-based training of the amblyopic eye to rescue amblyopia in adult life are underway in Finland and New Zealand (L. Maffei, personal communication. <http://www.hermopharma.com/news-a-publication/120-first-patients-completed-the-amblyopia-phase-2a-study>).

In this context, perceptual learning has long been used to improve spatial acuity in adult amblyopic patients (for review see [139]). Systematic training of patients with unilateral amblyopia (secondary to strabismus and anisometropia) in simple visual tasks revealed a 2-fold increase of contrast sensitivity and improved performance in letter-recognition tests [140]. Likewise, Snellen acuities in anisometric amblyopes improved after intensive training in a Vernier acuity task. Moreover, video game playing seems to promote a significant rescue of visual functions in adult amblyopic patients. Playing video games (both action and nonaction

games) for a short period of time using the amblyopic eye results in a substantial improvement in a wide range of fundamental visual functions, including visual acuity, positional acuity, spatial attention, and stereopsis [141].

The improvement of performance seen in perceptual learning is proportional to the number of trials taken, although performance eventually reaches an asymptote of no further progress [142]. Unfortunately, the extent to which acuity improvements occur is limited by the task specificity of perceptual learning [143]. It is worth mentioning that in most instances of perceptual learning, attention to the trained stimulus is necessary for improvements of vision to occur [144]. This observation is particularly important as it epitomizes the role of long-distance neuromodulatory systems in the physiological state of arousal, which regulates mechanisms of attention and information processing that may contribute to functional changes of neural circuitries in the adult brain. This points toward the possibility to pharmacologically enhance plasticity as a strategy for brain repair in a variety of pathological states where reorganization of neuronal networks would be beneficial in adult life. This, for instance, could facilitate restructuring of mature circuitries impaired by damage or disease. Long-term pharmacologically induced serotonergic transmission actually enhances the effects of rehabilitation in the recovery from motor deficits after ischemic stroke in humans [145].

Acknowledgments

This work was supported by a grant to J. F. Maya-Vetencourt from Progetto di Ricerca di Giovani Ricercatori of the Scuola Normale Superiore. J. F. Maya-Vetencourt is supported by a research fellowship provided by the Scuola Normale Superiore, Pisa, Italy.

References

- [1] L. C. Katz and C. J. Shatz, "Synaptic activity and the construction of cortical circuits," *Science*, vol. 274, no. 5290, pp. 1133–1138, 1996.
- [2] N. Berardi, T. Pizzorusso, and L. Maffei, "Critical periods during sensory development," *Current Opinion in Neurobiology*, vol. 10, no. 1, pp. 138–145, 2000.
- [3] T. Kasamatsu and J. D. Pettigrew, "Depletion of brain catecholamines. Failure of ocular dominance shift after monocular occlusion in kittens," *Science*, vol. 194, no. 4261, pp. 206–209, 1976.
- [4] M. F. Bear and W. Singer, "Modulation of visual cortical plasticity by acetylcholine and noradrenaline," *Nature*, vol. 320, no. 6058, pp. 172–176, 1986.
- [5] Q. Gu and W. Singer, "Effects of intracortical infusion of anticholinergic drugs on neuronal plasticity in kitten striate cortex," *European Journal of Neuroscience*, vol. 5, no. 5, pp. 475–485, 1993.
- [6] Q. Gu and W. Singer, "Involvement of serotonin in developmental plasticity of kitten visual cortex," *European Journal of Neuroscience*, vol. 7, no. 6, pp. 1146–1153, 1995.
- [7] Y. Wang, Q. Gu, and M. S. Cynader, "Blockade of serotonin-2C receptors by mesulergine reduces ocular dominance plasticity in kitten visual cortex," *Experimental Brain Research*, vol. 114, no. 2, pp. 321–328, 1997.

- [8] L. Kojic, Q. Gu, R. M. Douglas, and M. S. Cynader, "Serotonin facilitates synaptic plasticity in kitten visual cortex: an in vitro study," *Developmental Brain Research*, vol. 101, no. 1-2, pp. 299–304, 1997.
- [9] J. D. Pettigrew and T. Kasamatsu, "Local perfusion of nor-adrenaline maintains visual cortical plasticity," *Nature*, vol. 271, no. 5647, pp. 761–763, 1978.
- [10] T. Kasamatsu, J. D. Pettigrew, and M. Ary, "Restoration of visual cortical plasticity by local microperfusion of norepinephrine," *Journal of Comparative Neurology*, vol. 185, no. 1, pp. 163–181, 1979.
- [11] T. Kasamatsu, K. Watabe, P. Heggelund, and E. Scholler, "Plasticity in cat visual cortex restored by electrical stimulation of the locus coeruleus," *Neuroscience Research*, vol. 2, no. 5, pp. 365–386, 1985.
- [12] K. Imamura and T. Kasamatsu, "Ocular dominance plasticity restored by NA infusion to aplastic visual cortex of anesthetized and paralyzed kittens," *Experimental Brain Research*, vol. 87, no. 2, pp. 309–318, 1991.
- [13] N. Mataga, K. Imamura, and Y. Watanabe, "L-threo-3,4-Dihydroxyphenylserine enhanced ocular dominance plasticity in adult cats," *Neuroscience Letters*, vol. 142, no. 2, pp. 115–118, 1992.
- [14] H. Morishita, J. M. Miwa, N. Heintz, and T. K. Hensch, "Lynx1, a cholinergic brake, limits plasticity in adult visual cortex," *Science*, vol. 330, no. 6008, pp. 1238–1240, 2010.
- [15] J. F. Maya-Vetencourt, A. Sale, A. Viegi et al., "The antidepressant fluoxetine restores plasticity in the adult visual cortex," *Science*, vol. 320, no. 5874, pp. 385–388, 2008.
- [16] J. F. Maya-Vetencourt, E. Tiraboschi, M. Spolidoro, E. Castrén, and L. Maffei, "Serotonin triggers a transient epigenetic mechanism that reinstates adult visual cortex plasticity in rats," *European Journal of Neuroscience*, vol. 33, no. 1, pp. 49–57, 2011.
- [17] L. Baroncelli, A. Sale, A. Viegi et al., "Experience-dependent reactivation of ocular dominance plasticity in the adult visual cortex," *Experimental Neurology*, vol. 226, no. 1, pp. 100–109, 2010.
- [18] T. K. Hensch, M. Fagiolini, N. Mataga, M. P. Stryker, S. Baekkeskov, and S. F. Kash, "Local GABA circuit control of experience-dependent plasticity in developing visual cortex," *Science*, vol. 282, no. 5393, pp. 1504–1508, 1998.
- [19] M. Fagiolini and T. K. Hensch, "Inhibitory threshold for critical-period activation in primary visual cortex," *Nature*, vol. 404, no. 6774, pp. 183–186, 2000.
- [20] T. K. Hensch, "Critical period plasticity in local cortical circuits," *Nature Reviews Neuroscience*, vol. 6, no. 11, pp. 877–888, 2005.
- [21] M. Fagiolini, J. M. Fritschy, K. Löw, H. Möhler, U. Rudolph, and T. K. Hensch, "Specific GABAA circuits for visual cortical plasticity," *Science*, vol. 303, no. 5664, pp. 1681–1683, 2004.
- [22] S. Sugiyama, A. A. Di Nardo, S. Aizawa et al., "Experience-dependent transfer of Otx2 homeoprotein into the visual cortex activates postnatal plasticity," *Cell*, vol. 134, no. 3, pp. 508–520, 2008.
- [23] A. Harauzov, M. Spolidoro, G. DiCristo et al., "Reducing intracortical inhibition in the adult visual cortex promotes ocular dominance plasticity," *Journal of Neuroscience*, vol. 30, no. 1, pp. 361–371, 2010.
- [24] A. Sale, N. Berardi, M. Spolidoro, L. Baroncelli, and L. Maffei, "GABAergic inhibition in visual cortical plasticity," *Frontiers in Cellular Neuroscience*, vol. 4, p. 10, 2010.
- [25] L. Maffei, N. Berardi, L. Domenici, V. Parisi, and T. Pizzorusso, "Nerve growth factor (NGF) prevents the shift in ocular dominance distribution of visual cortical neurons in monocularly deprived rats," *Journal of Neuroscience*, vol. 12, no. 12, pp. 4651–4662, 1992.
- [26] R. J. Cabelli, A. Hohn, and C. J. Shatz, "Inhibition of ocular dominance column formation by infusion of NT-4/5 or BDNF," *Science*, vol. 267, no. 5204, pp. 1662–1666, 1995.
- [27] R. J. Cabelli, D. L. Shelton, R. A. Segal, and C. J. Shatz, "Blockade of endogenous ligands of TrkB inhibits formation of ocular dominance columns," *Neuron*, vol. 19, no. 1, pp. 63–76, 1997.
- [28] M. Caleo, C. Lodovichi, and L. Maffei, "Effects of nerve growth factor on visual cortical plasticity require afferent electrical activity," *European Journal of Neuroscience*, vol. 11, no. 8, pp. 2979–2984, 1999.
- [29] Z. J. Huang, A. Kirkwood, T. Pizzorusso et al., "BDNF regulates the maturation of inhibition and the critical period of plasticity in mouse visual cortex," *Cell*, vol. 98, no. 6, pp. 739–755, 1999.
- [30] A. W. McGee, Y. Yang, Q. S. Fischer, N. W. Daw, and S. H. Strittmatter, "Experience-driven plasticity of visual cortex limited by myelin and nogo receptor," *Science*, vol. 309, no. 5744, pp. 2222–2226, 2005.
- [31] J. K. Atwal, J. Pinkston-Gosse, J. Syken et al., "PirB is a functional receptor for myelin inhibitors of axonal regeneration," *Science*, vol. 322, no. 5903, pp. 967–970, 2008.
- [32] J. Syken, T. GrandPre, P. O. Kanold, and C. J. Shatz, "PirB restricts ocular-dominance plasticity in visual cortex," *Science*, vol. 313, no. 5794, pp. 1795–1800, 2006.
- [33] G. Di Cristo, B. Chattopadhyaya, S. J. Kuhlman et al., "Activity-dependent PSA expression regulates inhibitory maturation and onset of critical period plasticity," *Nature Neuroscience*, vol. 10, no. 12, pp. 1569–1577, 2007.
- [34] T. Pizzorusso, P. Medini, N. Berardi, S. Chierzi, J. W. Fawcett, and L. Maffei, "Reactivation of ocular dominance plasticity in the adult visual cortex," *Science*, vol. 298, no. 5596, pp. 1248–1251, 2002.
- [35] T. Pizzorusso, P. Medini, S. Landi, S. Baldini, N. Berardi, and L. Maffei, "Structural and functional recovery from early monocular deprivation in adult rats," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 103, no. 22, pp. 8517–8522, 2006.
- [36] M. Spolidoro, L. Baroncelli, E. Putignano, J. F. Maya-Vetencourt, A. Viegi, and L. Maffei, "Food restriction enhances visual cortex plasticity in adulthood," *Nature Communications*, vol. 2, no. 1, article 320, 2011.
- [37] N. W. Daw, H. Sato, K. Fox, T. Carmichael, and R. Gingerich, "Cortisol reduces plasticity in the kitten visual cortex," *Journal of Neurobiology*, vol. 22, no. 2, pp. 158–168, 1991.
- [38] C. H. Liu, A. J. Heynen, M. G. H. Shuler, and M. F. Bear, "Cannabinoid receptor blockade reveals parallel plasticity mechanisms in different layers of mouse visual cortex," *Neuron*, vol. 58, no. 3, pp. 340–345, 2008.
- [39] G. Di Cristo, N. Berardi, L. Cancedda et al., "Requirement of ERK activation for visual cortical plasticity," *Science*, vol. 292, no. 5525, pp. 2337–2340, 2001.
- [40] C. J. Beaver, Q. Ji, Q. S. Fischer, and N. W. Daw, "Cyclic AMP-dependent protein kinase mediates ocular dominance shifts in cat visual cortex," *Nature Neuroscience*, vol. 4, no. 2, pp. 159–163, 2001.
- [41] S. Taha, J. L. Hanover, A. J. Silva, and M. P. Stryker, "Autophosphorylation of α CaMKII Is required for ocular dominance plasticity," *Neuron*, vol. 36, no. 3, pp. 483–491, 2002.

- [42] D. Bavelier, D. M. Levi, R. W. Li, Y. Dan, and T. K. Hensch, "Removing brakes on adult brain plasticity: from molecular to behavioral interventions," *Journal of Neuroscience*, vol. 30, no. 45, pp. 14964–14971, 2010.
- [43] J. M. Holmes and M. P. Clarke, "Amblyopia," *The Lancet*, vol. 367, no. 9519, pp. 1343–1351, 2006.
- [44] D. S. Friedman, M. X. Repka, J. Katz et al., "Prevalence of amblyopia and strabismus in white and African American children aged 6 through 71 months. The Baltimore Pediatric Eye Disease study," *Ophthalmology*, vol. 116, no. 11, pp. 2128–2134, 2009.
- [45] I. C. Lloyd, J. Ashworth, S. Biswas, and R. V. Abadi, "Advances in the management of congenital and infantile cataract," *Eye*, vol. 21, no. 10, pp. 1301–1309, 2007.
- [46] S. Milazzo, P. Turut, and D. Brémond-Gignac, "Infantile cataract et surgical management," *Journal Francais d'Ophthalmologie*, vol. 34, no. 3, pp. 192–197, 2011.
- [47] D. Maurer and T. K. Hensch, "Amblyopia: background to the special issue on stroke recovery," *Developmental Psychobiology*, vol. 54, no. 3, pp. 224–238, 2012.
- [48] F. Sengpiel, "Experimental models of amblyopia: insights for prevention and treatment," *Strabismus*, vol. 19, no. 3, pp. 87–90, 2011.
- [49] S. Le Vay, T. N. Wiesel, and D. H. Hubel, "The development of ocular dominance columns in normal and visually deprived monkeys," *Journal of Comparative Neurology*, vol. 191, no. 1, pp. 1–51, 1980.
- [50] D. H. Hubel and T. N. Wiesel, "The period of susceptibility to the physiological effects of unilateral eye closure in kittens," *Journal of Physiology*, vol. 206, no. 2, pp. 419–436, 1970.
- [51] A. Antonini and M. P. Stryker, "Rapid remodeling of axonal arbors in the visual cortex," *Science*, vol. 260, no. 5115, pp. 1819–1821, 1993.
- [52] T. N. Wiesel and D. H. Hubel, "Single-cell responses in striate cortex of kittens deprived of vision in one eye," *Journal of neurophysiology*, vol. 26, pp. 1003–1017, 1963.
- [53] C. Blakemore and R. C. Van Sluyters, "Reversal of the physiological effects of monocular deprivation in kittens: further evidence for a sensitive period," *Journal of Physiology*, vol. 237, no. 1, pp. 195–216, 1974.
- [54] C. Blakemore, F. Vital-Durand, and L. J. Garey, "Recovery from monocular deprivation in the monkey. I. Reversal of physiological effects in the visual cortex," *Proceedings of the Royal Society of London*, vol. 213, no. 1193, pp. 399–423, 1981.
- [55] G. T. Prusky and R. M. Douglas, "Developmental plasticity of mouse visual acuity," *European Journal of Neuroscience*, vol. 17, no. 1, pp. 167–173, 2003.
- [56] L. Maffei and A. Fiorentini, "Monocular deprivation in kittens impairs the spatial resolution of geniculate neurones," *Nature*, vol. 264, no. 5588, pp. 754–755, 1976.
- [57] B. Timney, D. E. Mitchell, and F. Giffin, "The development of vision in cats after extended periods of dark-rearing," *Experimental Brain Research*, vol. 31, no. 4, pp. 547–560, 1978.
- [58] C. Blakemore and D. J. Price, "Effects of dark-rearing on the development of area 18 of the cat's visual cortex," *Journal of Physiology*, vol. 384, pp. 293–309, 1987.
- [59] M. Fagiolini, T. Pizzorusso, N. Berardi, L. Domenici, and L. Maffei, "Functional postnatal development of the rat primary visual cortex and the role of visual experience: dark rearing and monocular deprivation," *Vision Research*, vol. 34, no. 6, pp. 709–720, 1994.
- [60] T. Cotrufo, A. Viegi, N. Berardi, Y. Bozzi, L. Mascia, and L. Maffei, "Effects of neurotrophins on synaptic protein expression in the visual cortex of dark-reared rats," *Journal of Neuroscience*, vol. 23, no. 9, pp. 3566–3571, 2003.
- [61] A. Kirkwood, H. K. Lee, and M. F. Bear, "Co-regulation of long-term potentiation and experience-dependent synaptic plasticity in visual cortex by age and experience," *Nature*, vol. 375, no. 6529, pp. 328–331, 1995.
- [62] L. Gianfranceschi, R. Siciliano, J. Walls et al., "Visual cortex is rescued from the effects of dark rearing by overexpression of BDNF," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 100, no. 21, pp. 12486–12491, 2003.
- [63] A. Bartoletti, P. Medini, N. Berardi, and L. Maffei, "Environmental enrichment prevents effects of dark-rearing in the rat visual cortex," *Nature Neuroscience*, vol. 7, no. 3, pp. 215–216, 2004.
- [64] L. Cancedda, E. Putignano, A. Sale, A. Viegi, N. Berardi, and L. Maffei, "Acceleration of visual system development by environmental enrichment," *Journal of Neuroscience*, vol. 24, no. 20, pp. 4840–4848, 2004.
- [65] L. D. F. Moon, R. A. Asher, K. E. Rhodes, and J. W. Fawcett, "Regeneration of CNS axons back to their target following treatment of adult rat brain with chondroitinase ABC," *Nature Neuroscience*, vol. 4, no. 5, pp. 465–466, 2001.
- [66] E. J. Bradbury, L. D. F. Moon, R. J. Popat et al., "Chondroitinase ABC promotes functional recovery after spinal cord injury," *Nature*, vol. 416, no. 6881, pp. 636–640, 2002.
- [67] W. Singer, "The formation of cooperative cell assemblies in the visual cortex," *Journal of Experimental Biology*, vol. 153, pp. 177–197, 1990.
- [68] W. Singer, "Development and plasticity of cortical processing architectures," *Science*, vol. 270, no. 5237, pp. 758–764, 1995.
- [69] S. Brocher, A. Artola, and W. Singer, "Agonists of cholinergic and noradrenergic receptors facilitate synergistically the induction of long-term potentiation in slices of rat visual cortex," *Brain Research*, vol. 573, no. 1, pp. 27–36, 1992.
- [70] A. Kirkwood, C. Rozas, J. Kirkwood, F. Perez, and M. F. Bear, "Modulation of long-term synaptic depression in visual cortex by acetylcholine and norepinephrine," *Journal of Neuroscience*, vol. 19, no. 5, pp. 1599–1609, 1999.
- [71] M. F. Bear, "NMDA-receptor-dependent synaptic plasticity in the visual cortex," *Progress in Brain Research*, vol. 108, pp. 205–218, 1996.
- [72] W. A. Moreau, M. Amar, N. Le Roux, N. Morel, and P. Fossier, "Serotonergic fine-tuning of the excitation-inhibition balance in rat visual cortical networks," *Cerebral Cortex*, vol. 20, no. 2, pp. 456–467, 2010.
- [73] M. Amar, E. Lucas-Meunier, G. Baux, and P. Fossier, "Blockade of different muscarinic receptor subtypes changes the equilibrium between excitation and inhibition in rat visual cortex," *Neuroscience*, vol. 169, no. 4, pp. 1610–1620, 2010.
- [74] Y. Rao and N. W. Daw, "Layer variations of long-term depression in rat visual cortex," *Journal of Neurophysiology*, vol. 92, no. 5, pp. 2652–2658, 2004.
- [75] E. Pesavento, E. Margotti, M. Righi, A. Cattaneo, and L. Domenici, "Blocking the NGF-TrkA interaction rescues the developmental loss of LTP in the rat visual cortex: role of the cholinergic system," *Neuron*, vol. 25, no. 1, pp. 165–175, 2000.
- [76] N. Origlia, S. Capsoni, L. Domenici, and A. Cattaneo, "Time window in cholinomimetic ability to rescue long-term potentiation in neurodegenerating anti-nerve growth factor mice," *Journal of Alzheimer's Disease*, vol. 9, no. 1, pp. 59–68, 2006.

- [77] N. Kuczewski, E. Aztiria, G. Leanza, and L. Domenici, "Selective cholinergic immunolesioning affects synaptic plasticity in developing visual cortex," *European Journal of Neuroscience*, vol. 21, no. 7, pp. 1807–1814, 2005.
- [78] N. Origlia, N. Kuczewski, E. Aztiria, D. Gautam, J. Wess, and L. Domenici, "Muscarinic acetylcholine receptor knockout mice show distinct synaptic plasticity impairments in the visual cortex," *Journal of Physiology*, vol. 577, no. 3, pp. 829–840, 2006.
- [79] H. Y. He, W. Hodos, and E. M. Quinlan, "Visual deprivation reactivates rapid ocular dominance plasticity in adult visual cortex," *Journal of Neuroscience*, vol. 26, no. 11, pp. 2951–2955, 2006.
- [80] H. Y. He, B. Ray, K. Dennis, and E. M. Quinlan, "Experience-dependent recovery of vision following chronic deprivation amblyopia," *Nature Neuroscience*, vol. 10, no. 9, pp. 1134–1136, 2007.
- [81] A. Sale, J. F. Maya-Vetencourt, P. Medini et al., "Environmental enrichment in adulthood promotes amblyopia recovery through a reduction of intracortical inhibition," *Nature Neuroscience*, vol. 10, no. 6, pp. 679–681, 2007.
- [82] A. Sale, N. Berardi, and L. Maffei, "Enrich the environment to empower the brain," *Trends in Neurosciences*, vol. 32, no. 4, pp. 233–239, 2009.
- [83] J. F. Maya-Vetencourt, L. Baroncelli, A. Viegi et al., "IGF-1 restores visual cortex plasticity in adult life by reducing local GABA levels," *Neural Plasticity*, vol. 2012, Article ID 250421, 10 pages, 2012.
- [84] E. Putignano, G. Lonetti, L. Cancedda et al., "Developmental downregulation of histone posttranslational modifications regulates visual cortical plasticity," *Neuron*, vol. 53, no. 5, pp. 747–759, 2007.
- [85] D. Silingardi, M. Scali, G. Belluomini, and T. Pizzorusso, "Epigenetic treatments of adult rats promote recovery from visual acuity deficits induced by long-term monocular deprivation," *European Journal of Neuroscience*, vol. 31, no. 12, pp. 2185–2192, 2010.
- [86] J. L. Chen, W. C. Lin, J. W. Cha, P. T. So, Y. Kubota, and E. Nedivi, "Structural basis for the role of inhibition in facilitating adult brain plasticity," *Nature Neuroscience*, vol. 14, no. 5, pp. 587–596, 2011.
- [87] A. Fischer, F. Sananbenesi, X. Wang, M. Dobbin, and L. H. Tsai, "Recovery of learning and memory is associated with chromatin remodelling," *Nature*, vol. 447, no. 7141, pp. 178–182, 2007.
- [88] W. Wallace and M. F. Bear, "A morphological correlate of synaptic scaling in visual cortex," *Journal of Neuroscience*, vol. 24, no. 31, pp. 6928–6938, 2004.
- [89] A. Majewska and M. Sur, "Motility of dendritic spines in visual cortex in vivo: changes during the critical period and effects of visual deprivation," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 100, no. 26, pp. 16024–16029, 2003.
- [90] N. Mataga, Y. Mizuguchi, and T. K. Hensch, "Experience-dependent pruning of dendritic spines in visual cortex by tissue plasminogen activator," *Neuron*, vol. 44, no. 6, pp. 1031–1041, 2004.
- [91] S. B. Hofer, T. D. Mrsic-Flogel, T. Bonhoeffer, and M. Hübener, "Experience leaves a lasting structural trace in cortical circuits," *Nature*, vol. 457, no. 7227, pp. 313–317, 2009.
- [92] D. Tropea, A. K. Majewska, R. Garcia, and M. Sur, "Structural dynamics of synapses in vivo correlate with functional changes during experience-dependent plasticity in visual cortex," *Journal of Neuroscience*, vol. 30, no. 33, pp. 11086–11095, 2010.
- [93] M. Fu and Y. Zuo, "Experience-dependent structural plasticity in the cortex," *Trends in Neurosciences*, vol. 34, no. 4, pp. 177–187, 2011.
- [94] H. Yu, A. K. Majewska, and M. Sur, "Rapid experience-dependent plasticity of synapse function and structure in ferret visual cortex in vivo," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 52, pp. 21235–21240, 2011.
- [95] C. Cerri, A. Fabbri, E. Vannini et al., "Activation of Rho GTPases triggers structural remodeling and functional plasticity in the adult rat visual cortex," *Journal of Neuroscience*, vol. 31, no. 42, pp. 15163–15172, 2011.
- [96] T. Xu, X. Yu, A. J. Perlik et al., "Rapid formation and selective stabilization of synapses for enduring motor memories," *Nature*, vol. 462, no. 7275, pp. 915–919, 2009.
- [97] K. Zito, V. Scheuss, G. Knott, T. Hill, and K. Svoboda, "Rapid functional maturation of nascent dendritic spines," *Neuron*, vol. 61, no. 2, pp. 247–258, 2009.
- [98] C. O'Donnel, M. F. Nolan, and M. C. W. van Rossum, "Dendritic spine dynamics regulate the long-term stability of synaptic plasticity," *Journal of Neuroscience*, vol. 31, no. 45, pp. 16142–16156, 2011.
- [99] W. Renthall, I. Maze, V. Krishnan et al., "Histone deacetylase 5 epigenetically controls behavioral adaptations to chronic emotional stimuli," *Neuron*, vol. 56, no. 3, pp. 517–529, 2007.
- [100] C. Crosio, E. Heitz, C. D. Allis, E. Borrelli, and P. Sassone-Corsi, "Chromatin remodeling and neuronal response: multiple signaling pathways induce specific histone H3 modifications and early gene expression in hippocampal neurons," *Journal of Cell Science*, vol. 116, no. 24, pp. 4905–4914, 2003.
- [101] J. M. Levenson, K. J. O'Riordan, K. D. Brown, M. A. Trinh, D. L. Molfese, and J. D. Sweatt, "Regulation of histone acetylation during memory formation in the hippocampus," *Journal of Biological Chemistry*, vol. 279, no. 39, pp. 40545–40559, 2004.
- [102] D. Francis, J. Diorio, D. Liu, and M. J. Meaney, "Nongenomic transmission across generations of maternal behavior and stress responses in the rat," *Science*, vol. 286, no. 5442, pp. 1155–1158, 1999.
- [103] I. C. G. Weaver, N. Cervoni, F. A. Champagne et al., "Epigenetic programming by maternal behavior," *Nature Neuroscience*, vol. 7, no. 8, pp. 847–854, 2004.
- [104] T. A. Pham, S. Impey, D. R. Storm, and M. P. Stryker, "Cremediated gene transcription in neocortical neuronal plasticity during the developmental critical period," *Neuron*, vol. 22, no. 1, pp. 63–72, 1999.
- [105] A. F. Mower, D. S. Liao, E. J. Nestler, R. L. Neve, and A. S. Ramoa, "cAMP/Ca²⁺ response element-binding protein function is essential for ocular dominance plasticity," *Journal of Neuroscience*, vol. 22, no. 6, pp. 2237–2245, 2002.
- [106] A. E. West, E. C. Griffith, and M. E. Greenberg, "Regulation of transcription factors by neuronal activity," *Nature Reviews Neuroscience*, vol. 3, no. 12, pp. 921–931, 2002.
- [107] R. J. Kelleher, A. Govindarajan, and S. Tonegawa, "Translational regulatory mechanisms in persistent forms of synaptic plasticity," *Neuron*, vol. 44, no. 1, pp. 59–73, 2004.
- [108] G. M. Schratt, F. Tuebing, E. A. Nigh et al., "A brain-specific microRNA regulates dendritic spine development," *Nature*, vol. 439, no. 7074, pp. 283–289, 2006.

- [109] G. A. Wayman, M. Davare, H. Ando et al., "An activity-regulated microRNA controls dendritic plasticity by down-regulating p250GAP," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 26, pp. 9093–9098, 2008.
- [110] S. Impey, M. Davare, A. Lasiek et al., "An activity-induced microRNA controls dendritic spine formation by regulating Rac1-PAK signaling," *Molecular and Cellular Neuroscience*, vol. 43, no. 1, pp. 146–156, 2010.
- [111] N. Vo, M. E. Klein, O. Varlamova et al., "A cAMP-response element binding protein-induced microRNA regulates neuronal morphogenesis," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 102, no. 45, pp. 16426–16431, 2005.
- [112] K. F. Hansen, K. Sakamoto, G. A. Wayman, S. Impey, and K. Obrietan, "Transgenic miR132 alters neuronal spine density and impairs novel object recognition memory," *PLoS ONE*, vol. 5, no. 11, Article ID e15497, 2010.
- [113] M. Alvarez-Saavedra, G. Antoun, A. Yanagiya et al., "miRNA-132 orchestrates chromatin remodeling and translational control of the circadian clock," *Human Molecular Genetics*, vol. 20, no. 4, Article ID ddq519, pp. 731–751, 2011.
- [114] K. Wibrand, D. Panja, A. Tiron et al., "Differential regulation of mature and precursor microRNA expression by NMDA and metabotropic glutamate receptor activation during LTP in the adult dentate gyrus in vivo," *European Journal of Neuroscience*, vol. 31, no. 4, pp. 636–645, 2010.
- [115] T. J. Lambert, D. R. Storm, and J. M. Sullivan, "MicroRNA132 modulates short-term synaptic plasticity but not basal release probability in hippocampal neurons," *PLoS ONE*, vol. 5, no. 12, Article ID e15182, 2010.
- [116] N. Mellios, H. Sugihara, J. Castro et al., "MiR-132, an experience-dependent microRNA, is essential for visual cortex plasticity," *Nature Neuroscience*, vol. 14, no. 10, pp. 1240–1242, 2011.
- [117] P. Tognini, E. Putignano, A. Coatti, and T. Pizzorusso, "Experience-dependent expression of miR-132 regulates ocular dominance plasticity," *Nature Neuroscience*, vol. 14, no. 10, pp. 1237–1239, 2011.
- [118] P. Tognini and T. Pizzorusso, "MicroRNA212/132 family: molecular transducer of neuronal function and plasticity," *International Journal of Biochemistry and Cell Biology*, vol. 44, no. 1, pp. 6–10, 2012.
- [119] D. Bavelier and H. J. Neville, "Cross-modal plasticity: where and how?" *Nature Reviews Neuroscience*, vol. 3, no. 6, pp. 443–452, 2002.
- [120] B. Röder, W. Teder-Sälejärvi, A. Sterr, F. Rösler, S. A. Hillyard, and H. J. Neville, "Improved auditory spatial tuning in blind humans," *Nature*, vol. 400, no. 6740, pp. 162–166, 1999.
- [121] N. Sadato, A. Pascual-Leone, J. Grafman et al., "Activation of the primary visual cortex by Braille reading in blind subjects," *Nature*, vol. 380, no. 6574, pp. 526–528, 1996.
- [122] R. Kupers, M. Pappens, A. M. De Noordhout, J. Schoenen, M. Ptito, and A. Fumal, "rTMS of the occipital cortex abolishes Braille reading and repetition priming in blind subjects," *Neurology*, vol. 68, no. 9, pp. 691–693, 2007.
- [123] A. Amedi, N. Raz, H. Azulay, R. Malach, and E. Zohary, "Cortical activity during tactile exploration of objects in blind and sighted humans," *Restorative Neurology and Neuroscience*, vol. 28, no. 2, pp. 143–156, 2010.
- [124] L. G. Cohen, P. Celnik, A. Pascual-Leone et al., "Functional relevance of cross-modal plasticity in blind humans," *Nature*, vol. 389, no. 6647, pp. 180–183, 1997.
- [125] H. Burton, A. Z. Snyder, J. B. Diamond, and M. E. Raichle, "Adaptive changes in early and late blind: a fMRI study of verb generation to heard nouns," *Journal of Neurophysiology*, vol. 88, no. 6, pp. 3359–3371, 2002.
- [126] H. Burton, J. B. Diamond, and K. B. McDermott, "Dissociating cortical regions activated by semantic and phonological tasks: a fMRI study in blind and sighted people," *Journal of Neurophysiology*, vol. 90, no. 3, pp. 1965–1982, 2003.
- [127] L. Reich, M. Szwed, L. Cohen, and A. Amedi, "A ventral visual stream reading center independent of visual experience," *Current Biology*, vol. 21, no. 5, pp. 363–368, 2011.
- [128] R. Kupers, M. Beaulieu-Lefebvre, F. C. Schneider et al., "Neural correlates of olfactory processing in congenital blindness," *Neuropsychologia*, vol. 49, no. 7, pp. 2037–2044, 2011.
- [129] M. Bedny, A. Pascual-Leone, S. Dravida, and R. Saxe, "A sensitive period for language in the visual cortex: distinct patterns of plasticity in congenitally versus late blind adults," *Brain and Language*. In press.
- [130] L. G. Cohen, R. A. Weeks, N. Sadato, P. Celnik, K. Ishii, and M. Hallett, "Period of susceptibility for cross-modal plasticity in the blind," *Annals of Neurology*, vol. 45, no. 4, pp. 451–460, 1999.
- [131] C. Büchel, C. Price, R. S. J. Frackowiak, and K. Friston, "Differential activation patterns in the visual cortex of late and congenitally blind subjects," *Brain*, vol. 121, no. 3, pp. 409–419, 1998.
- [132] H. Burton, A. Z. Snyder, T. E. Conturo, E. Akbudak, J. M. Ollinger, and M. E. Raichle, "Adaptive changes in early and late blind: a fMRI study of Braille reading," *Journal of Neurophysiology*, vol. 87, no. 1, pp. 589–607, 2002.
- [133] A. Falchier, S. Clavagnier, P. Barone, and H. Kennedy, "Anatomical evidence of multimodal integration in primate striate cortex," *Journal of Neuroscience*, vol. 22, no. 13, pp. 5749–5759, 2002.
- [134] N. Lambert, E. R. Gizewski, A. De Greiff, and M. Forsting, "Cross-modal plasticity in deaf subjects dependent on the extent of hearing loss," *Cognitive Brain Research*, vol. 25, no. 3, pp. 884–890, 2005.
- [135] M. E. Doucet, F. Bergeron, M. Lassonde, P. Ferron, and F. Lepore, "Cross-modal reorganization and speech perception in cochlear implant users," *Brain*, vol. 129, no. 12, pp. 3376–3383, 2006.
- [136] H. Van Praag, G. Kempermann, and F. H. Gage, "Neural Consequences of environmental enrichment," *Nature Reviews Neuroscience*, vol. 1, no. 3, pp. 191–198, 2000.
- [137] A. Guzzetta, S. Baldini, A. Bancale et al., "Massage accelerates brain development and the maturation of visual function," *Journal of Neuroscience*, vol. 29, no. 18, pp. 6042–6051, 2009.
- [138] L. Baroncelli, J. Bonaccorsi, M. Milanese et al., "Enriched experience and recovery from amblyopia in adult rats: impact of motor, social and sensory components," *Neuropharmacology*, vol. 62, no. 7, pp. 2387–2396, 2012.
- [139] D. M. Levi and R. W. Li, "Perceptual learning as a potential treatment for amblyopia: a mini-review," *Vision Research*, vol. 49, no. 21, pp. 2535–2549, 2009.
- [140] U. Polat, T. Ma-Naim, M. Belkin, and D. Sagi, "Improving vision in adult amblyopia by perceptual learning," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 101, no. 17, pp. 6692–6697, 2004.
- [141] R. W. Li, C. Ngo, J. Nguyen, and D. M. Levi, "Video-game play induces plasticity in the visual system of adults with amblyopia," *PLoS Biology*, vol. 9, no. 8, Article ID e1001135, 2011.

- [142] C. B. Huang, Y. Zhou, and Z. L. Lu, "Broad bandwidth of perceptual learning in the visual system of adults with anisometropic amblyopia," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 10, pp. 4068–4073, 2008.
- [143] R. E. Crist, M. K. Kapadia, G. Westheimer, and C. D. Gilbert, "Perceptual learning of spatial localization: specificity for orientation, position, and context," *Journal of Neurophysiology*, vol. 78, no. 6, pp. 2889–2894, 1997.
- [144] M. Ahissar and S. Hochstein, "Attentional control of early perceptual learning," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 90, no. 12, pp. 5718–5722, 1993.
- [145] F. Chollet, J. Tardy, J. F. Albucher et al., "Fluoxetine for motor recovery after acute ischaemic stroke (FLAME): a randomised placebo-controlled trial," *The Lancet Neurology*, vol. 10, no. 2, pp. 123–130, 2011.

Review Article

Enriched and Deprived Sensory Experience Induces Structural Changes and Rewires Connectivity during the Postnatal Development of the Brain

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Received 16 December 2011; Revised 20 March 2012; Accepted 13 June 2012

Academic Editor: Pietro Pietrini

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During postnatal development, sensory experience modulates cortical development, inducing numerous changes in all of the components of the cortex. Most of the cortical changes thus induced occur during the critical period, when the functional and structural properties of cortical neurons are particularly susceptible to alterations. Although the time course for experience-mediated sensory development is specific for each system, postnatal development acts as a whole, and if one cortical area is deprived of its normal sensory inputs during early stages, it will be reorganized by the nondeprived senses in a process of cross-modal plasticity that not only increases performance in the remaining senses when one is deprived, but also rewires the brain allowing the deprived cortex to process inputs from other senses and cortices, maintaining the modular configuration. This paper summarizes our current understanding of sensory systems development, focused specially in the visual system. It delineates sensory enhancement and sensory deprivation effects at both physiological and anatomical levels and describes the use of enriched environment as a tool to rewire loss of brain areas to enhance other active senses. Finally, strategies to apply restorative features in human-deprived senses are studied, discussing the beneficial and detrimental effects of cross-modal plasticity in prostheses and sensory substitution devices implantation.

1. Introduction

After birth, sensory experience modulates the intrinsic developmental programs to shape both functional and anatomical cortical architecture and function from gene expression to activity patterns across systems [1–4]. The postnatal nervous system responds to stimuli from the outside world to develop and consolidate brain connections. Brain circuits are particularly susceptible to these stimuli during a special time window called critical period [3]. After this period, the brain wiring is mature and modifications are more difficult to be made. This natural process can be disturbed by a loss of these stimuli (deprivation) in the different sensory systems, such as visual system [5], somatosensory

system [6], or auditory system [7]. Deprivation of sensory inputs throughout postnatal development induces a major disturbance of axonal, dendritic, and synaptic connection patterns of neural circuitry [6, 8, 9]. A major regulator of experience-mediated tuning of sensory systems is the balance between excitation and inhibition [10, 11]. Although the time course for experience-mediated sensory development is specific for each system, postnatal development acts as a whole, and if one cortical area is deprived of its normal sensory inputs during early stages, it will be reorganized by the nondeprived senses in a process of cross-modal plasticity [12, 13].

The purpose of this paper is to detail cross-modal effects that can link different sensorial cortices. Particularly review

the effects of enriched environment over different senses, specially focused on visual system and the effects of this environment on visually deprived animals. Enriched environment is a tool with neuroprotective effects over many brain diseases and with restorative effects over sensory systems. Through a deeper understanding of this environment, better strategies can be designed to exert cross-modal effects in order to complement the missing sense with the spared ones.

2. Visual System Development

During postnatal development, specific connections among neurons within the visual cortex as well as its inputs and outputs are established, ultimately leading to a functional network. This process is completed in two stages, since environmental experience modulates the genetically pre-determined roadmap to shape functional and anatomical cortical architecture and function [2, 3, 14]. Experience exerts effects over the three major elements of the brain; increases the number and size of synapses per neuron [15], the neuronal activity [16, 17], and the metabolic demand [18, 19]; increases astroglial population [20]; causes changes of the vascular network [5, 15, 21].

The structure of the visual system follows the basic outlines of sensory systems. It is a hierarchical system that has a sensitive receptor, some intermediate stations, and a specific area in the cerebral cortex. The fact that it is a pathway known in depth and has great accessibility to each of its components makes it the system of choice in most studies of sensory systems or the cerebral cortex [22].

The influence of visual experience begins at eye opening, which in rats occurs in the second week of postnatal life [23]. This is the moment when a major reorganization of all the visual system mediated by visual stimuli begins to be felt; especially during the so-called critical period (maximum period of synaptic reorganization due to experience).

This period exist in many species, from humans to *Drosophila* [24], is specific for each brain area, and after this experience-mediated reorganization of the cortex, the sensory functions reach maturity [3, 25]. The closure of the critical period is completed when anatomical and functional phenomena are established. Structural factors such as perineuronal nets (formed around the neurons) [26, 27] and myelin-related proteins [28] inhibit axonal sprouting. On the other hand, functional changes between excitatory and inhibitory signals, such as intracortical GABAergic inhibition by parvalbumin positive interneurons [29], regulate the termination of the critical period. The exact period of vulnerability for the deprivation of cortical visual stimuli is important for understanding the normal development of the visual cortex. It has been shown that susceptibility to monocular occlusion begins around the end of the third week of postnatal life, peaks during the fourth and fifth weeks of postnatal life, and begins to decline after the end of the fifth week of postnatal life [30].

Although until relatively recent times it was believed that brain lost plasticity after the end of the critical period remaining fixed in adulthood, now it is well accepted that

the adult brain maintains certain degree of plasticity to cope with a changing environment throughout life [14], like an extended critical period. Throughout numerous studies, it has been found that a number of interventions can promote plasticity in adult rodents, including environmental enrichment [31], visual deprivation [32], previous monocular deprivation of the same eye [33], enzymatic degradation of the extracellular matrix [26], stimulation of histone acetylation [34], and the antidepressant fluoxetine [35].

The best studied model of age-dependent cortical plasticity is ocular dominance (OD), achieved by monocular deprivation (MD). Neurons in the binocular visual cortex respond to inputs from both eyes but are dominated by the contralateral eye (in rodents), and monocular deprivation induces a shift in the ocular dominance of binocular neurons towards the open eye. The ocular dominance is most pronounced in young animals during postnatal development (P25), is reduced in young adults (P95), and is absent in fully mature animals older than 110 days of age [36].

To date, most studies and efforts have focused on young animals, but the studies of the last years have opened a new window for studies of plasticity in adults and their therapeutic application.

3. Sensory Deprivation

Modifications of properties of sensory cortices by elimination of its natural sensory inputs (deprivation) serves as a model for studying brain plasticity and his capacity to rewire itself, showing an impressive range of cross-modal plasticity.

3.1. Visual System Deprivation. Although the influence of external experience takes place throughout the central nervous system, most studies on sensory deprivation have been performed on the visual cortex. The absence of visual experience from birth delays normal maturation and maintains the visual cortex in an immature state [20, 30]. In particular, visual connections do not consolidate. They remain plastic well after the closure of the physiological critical period and visual acuity does not develop [26]. The visual system organization facilitates the study of its structures through the interruption of pathways at different stages and through the deprivation of inputs using either invasive methods such as eyelid suturing [30, 37] and unilateral/bilateral enucleation [38], or noninvasive ones, such as dark rearing (DR) [5, 26, 39, 40]. Whereas dark rearing avoids any surgery action and leaves the cortex in an immature state which can be modified by subsequent visual experience [41], eyelid suture requires surgical manipulation and animals receive visual stimulation with diffuse light trough the suture eyelids that result in abnormal binocular interactions in the striate cortex and irreversible development defects on cortical physiology [42]. Enucleation is the most invasive technique that affects either physiologically and at structural level over animals and the effects are not of any greater extent than did dark rearing alone [43]. On the other hand, surgical or invasive techniques allow us to close only one eye so that we can see the effects

of monocular deprivation and compensation effects in the contralateral cortex, while dark rearing does not allow this fact, being deprivation bilateral.

Eliminating visual stimulation by dark rearing, alterations at physiological and morphological levels in 3 of the major components of the brain, neurons, astrocytes, and blood vessels are achieved. Deprivation of visual experience reduces synapse-neuron ratio [43] and alters brain-derived neurotrophic factor (BDNF) signaling affecting normal development of visual cortex neurons [44]. The astrocyte population is also affected. Astrocytic density is reduced in visual and somatosensory cortices [20, 40, 45] and the maturation of astrocytes is restricted [46]. Blood vessels, the third element of the neuroglivascular unit, are also affected by the lack of visual stimulation. There is a delay in the maturation of the microvascular pattern of the visual cortex. During postnatal development including the critical period, the vascular density is lower in rats reared in darkness due to decreased synaptic activity and lower energy requirements which need a lower rate of blood supply to meet demand in the cortex [5, 47]. The vascular area was also decreased and the number of neurons is minor, all related to a decrease in cortical activity [48]. The effects on brain vascularization are reflected in the principal angiogenic factor, vascular endothelial growth factor (VEGF). In the precritical period of the rat visual cortex, DR and control animals showed similar VEGF protein values, while during the critical period difference between the two groups were found, characterized by a reduced protein expression translates in a lower vascular density in visually deprived animals [39].

3.2. Nonvisual Sensory Deprivation. Another widely used system for sensory deprivation is the somatosensory cortex. Tactile information coming from whiskers plays a key role in the perception of the environment of rodents [6, 49, 50]. A major feature of the rodent primary somatosensory cortex (S1) is that layer IV contains a unique topographic representation of each facial whisker called a barrel, that is organized forming discrete cytoarchitectonic units [8, 51, 52]. This cortical organization allows the evaluation of the effects of manipulating single whiskers. The whisker map is established during the critical period that extends along the first postnatal week, and therefore precedes the visual or auditory critical periods [53]. Plasticity of the somatosensory cortex follows the same biochemical pathways of the rest of the senses [53, 54], and the critical period is also characterized by the development, balance, and pruning of excitatory and inhibitory synapses [55]. Impoverishing sensory activity by whisker trimming induces morphological and physiological alterations in the somatosensory barrel cortex when manipulation is performed during the critical period [6, 56–58]. Although the effects and the time course of S1 deprivation on neuronal architecture and function have been widely studied [59], the rest of the elements of the S1 cortex have received much less attention. With regard to vascularization, fMRI studies show a pattern of neurovascular coupling following whisker activity sharing most features of what happens in the visual cortex, where capillary density

is higher in the most active areas [60–62]. In an animal model for ischemia, increasing whisker stimulation after a ischemic injury increases the vascularization of the barrel cortex by upregulating angiogenic factors such as VEGF [63], thus, showing similar vascular effects to increased sensorial activity as we previously reported in the visual cortex [39].

Studies of other senses have also been mostly focused on neuronal structure and function. Studies on the effects of odor or auditory deprivation share similar effects with the visual or somatosensory systems [6, 64]. Nevertheless, the effects are not restricted to neurons, as olfactory deprivation also reduces the organization of astroglial networks [65].

4. Enriched Environment

The first approaches to the effects of environment on development can be traced back to the 19th century with Lamarck or Darwin [66, 67]. The latter reported that rodents raised in nature had bigger brains than caged domestic ones. At the end of the century, both Cajal and Foster advanced the effects of learning on synaptic plasticity [68, 69].

The study of experience-induced modification of brain morphology has been performed by conducting studies in a laboratory setting where environmental conditions can be modified [70].

Although the origin of the studies about effects of environmental enrichment (EE) can be traced back to centuries ago, the first systematic studies can be attributed to Donald Hebb in 1947, when he described how rats taken into his home and cared for as pets performed better on problem-solving tests than rats raised in cages [71]. Rosenzweig, Krech, Bennet, and Diamond, his group of disciples at Berkeley, defined the concept of environmental enrichment as the combination of complex inanimate and social stimulation. From their first studies, the enriched environment has been constantly implemented with cages bigger than standard ones, full of toys of different colors and shapes, tunnels, material to construct the nest, and a shelter, the latter having been recently described as a necessary element of environmental enrichment (Figure 1). These objects have been changed (the best schedule has been established as once every two days) and the placement of food has also been changed on a regular basis. Other elements that have a substantial influence are social interaction, so that wider cages allow rearing a greater number of animals that interchange social stimulation and physical exercise, forced or voluntary, that in rodents is commonly implemented by free access to an exercise wheel or by a treadmill [72, 73]. Some authors doubt whether physical exercise should be included. However, as physical exercise by itself induces brain changes, most enriched environment paradigms, starting from Hebb, have decided to include it.

Environmental enrichment increases sensory, cognitive, and motor stimulation and promotes activation, signaling, and neuronal plasticity in all brain areas, such as sensory ones like visual cortex [74–76], auditory cortex [77], or somatosensory cortex [78] or nonsensory ones like the

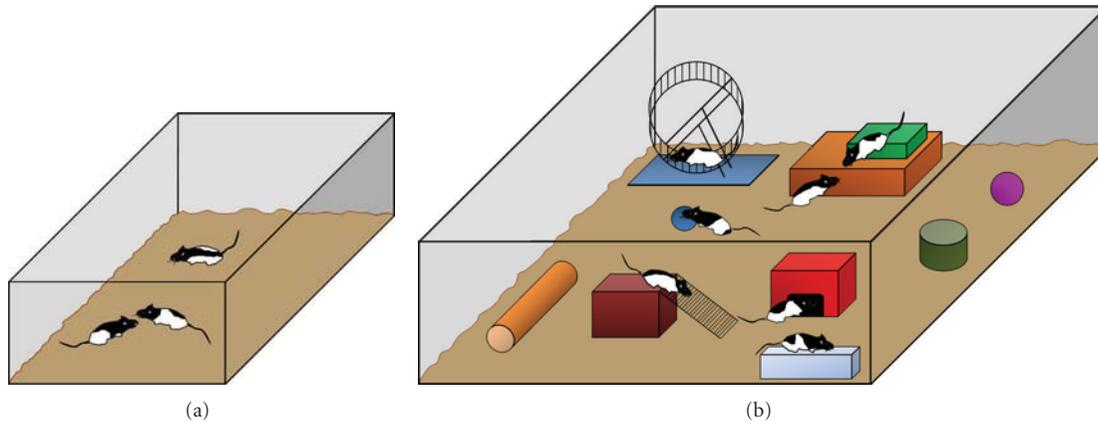


FIGURE 1: (a) Standard laboratory cage for animal rearing; (b) enriched environment (EE), defined as the combination of complex inanimate and social stimulation, formed by bigger cage than standard ones, full of toys of different colors, shapes, tunnels, material to construct the nest, a shelter, and an exercise wheel.

hippocampus [79], the amygdale [80], the basal ganglia [81, 82], and the cerebellar cortex [83].

Enrichment induces effects from cellular, molecular, or genetic levels up to behavioral ones. At anatomical level, initial studies showed that environmental enrichment increases cortical weight and thickness [84, 85]. Posterior works showed that EE increases dendritic branching and length, number of dendritic spines, and size of synapses in some neuronal populations [86, 87]. EE also increases hippocampal neurogenesis, mediated by VEGF [88], inhibits apoptosis [89], and has strong effects on the plasticity of neural connections, especially in the visual cortex [23, 90]. For the rest of the elements of the cortex, similar results have been described. Astrocytic morphology was changed due to exposure to enriched environment [74, 91], and size and density of astrocytes of the visual cortex [92, 93] and the somatosensory cortex were increased [40]. In addition the oligodendroglial density was also increased [92] and the same occurs with vascular density [18, 39, 94].

Most of these changes at the cellular level are in concordance with changes in the expression of genes involved in synaptic function and cell plasticity. Enrichment increases the levels of angioneurins, molecules that affect both the neural and the vascular cell processes [95]. Angioneurins include molecules first described as vascular growth factors, such as the archetypal angioneurin VEGF [39] and molecules first described as neurotrophins such as nerve growth factor (NGF) [96], brain-derived neurotrophic factor (BDNF) [90, 97], and neurotrophin-3 (NT-3) [98]. At the same time, it increases the expression of synapse proteins and induces changes in the expression of the subunits of the NMDA and AMPA receptors [99].

Apart from these increases at cellular and molecular levels, recent studies have reported the acceleration of visual system development as a consequence of environmental enrichment. Rearing animals in enriched environments induces earlier eye opening and has electrophysiological effects such as the early development of visual acuity [23].

Last but not least, rearing in enriched environments improves learning and memory [79, 100], decreases cognitive impairment due to aging [101, 102], diminishes anxiety, and increases exploratory activity [103]. Recent studies have outlined the importance of the duration of environmental enrichment, being relevant to the persistence of its effects on behavior [104, 105].

This wide range of effects exerted by EE over the whole brain made this environment a useful tool to improve their effects in brain disorders. Enhanced sensory, cognitive, and physical stimulation was able to mount neuroprotective responses against neurodegenerative processes, traumatic insults, or other forms of adult-onset neural dysfunction. EE delayed onset of cognitive deficits and depression-like behaviors associated with the Huntington's disease (HD) [106], was neuroprotective against rodent neurodegenerative disorder models like Parkinson's disease [107], or was able to ameliorate behavioral abnormalities in rodent models of psychiatric disorders, like schizophrenia [108].

5. Recovery from Sensory Deprivation

Brain has a great degree of reorganization following sensory deprivation. A common feature is the compensatory cross-modal plasticity that increases performance in the remaining senses when one is deprived [109, 110]. In sensory plasticity maps, the inputs from deprived senses weaken and shrink whereas spared or enriched signals strengthen and expand [11, 111]. Cross-modal plasticity implies not only physiological changes such as a higher activity of the nondeprived systems, but also the recruitment of the deprived area for the compensatory senses [112].

This process also occurs in nature, as happens in blind rats that have decreased visual areas and expanded the remaining sensory ones [113]. Although cross-modal plasticity plays an important role by compensating a deprived sense, it could notably hinder therapies directed at recovering the deprived sense, as the cortical areas devoted to it have been already "occupied" by nondeprived systems.

In principle, most of the studies on the recovery of sensorial deprivation have been developed in the visual cortex, but due to cross plasticity, their effects are not restricted to the deprived system, and most of them show effects all over the cortex. Probably the best-known method to compensate sensorial deprivation is by environmental enrichment, also used to compensate the effects of many brain diseases. As previously mentioned, rearing in complete darkness from birth has major effects on the development of the visual cortex, mainly around the critical period. These effects can be altered or modified if animals are dark-reared in complex environments. The first study about prevention of dark rearing effects by environmental enrichment was performed by Bartoletti et al. [104], showing that environmental enrichment promotes consolidation of visual cortical connections, development of visual acuity in dark-reared rats, and restore the effects of dark rearing in chondroitin sulphate proteoglycans developmental organization into perineuronal nets in the visual cortex. In addition to the effects of enrichment on the maturation of cortical connections, effects of enrichment in rats reared in darkness in two of the major elements of the brain, blood vessels, and astrocytes have been analyzed [40, 114]. In our first studies combining this compensatory system, we observed the effects of EE without an exercise wheel in dark-reared rats analyzing the vascular system of the visual cortex. Enrichment cannot recover the deprivation effect over the vascular density of rats reared in darkness. Effects of enrichment, both at the structural level (vascular density) and the molecular level (the level of VEGF protein), were not sufficient to compensate the effects produced by breeding in darkness, and the values of both groups, dark rearing (DR) and dark rearing in enriched environment cages (DR-EE), were similar, lower than the control group [39]. These results remain if we applied the same paradigm to the study of astrocyte density [40]. When exercise is included as part of the enrichment (DR-EE-Ex), recovery effects are observed. DR-EE-Ex environment deprives visual system and enhances somatosensory (darkness force animals to use whiskers to compensate the lack of visual stimulus) and motor systems. The compensatory role existing between different sensory systems is observed. Without visual excitement, an increase in the stimulation of both motor and somatosensory systems is reflected in the visual cortex, where the density of astrocytes of the DR-EE-Ex group is higher than that of the DR group and the control group (Figure 2) [40].

Another commonly used method to compensate the effects of sensorial deprivation is the exogenous administration of neurotrophins such as BDNF or NGF thus showing their key role in the experience-mediated development of the sensorial systems and controlling the onset and timing of the critical period [115–117]. As exogenous BDNF administration compensates the effects of sensorial deprivation, the previously mentioned effect of exercise on the compensation of visual deprivation could be explained by the fact that exercise upregulates BDNF and thus contributes to the restoration of deprived sensorial systems. In systems other than visual, BDNF administration also compensates at least

in part the effects of sensorial deprivation as in the auditory system following deafness [118].

Most of the studies on cross-modal plasticity in humans have been performed in early blind individuals. This process allows us to integrate information received from different senses to elaborate a complex response [119]. In congenitally or early blind individuals, there is an activation of the occipital cortex in response to tactile or auditive inputs that can be demonstrated by an increase in blood-oxygenation-level-dependent responses (BOLD) [120]. Moreover, the visually deprived occipital cortex maintains a high degree of organization and specificity, as the areas that process spatial information in nonblind humans keep this specific function and process spatial information from the auditory system in blinds [121]. Therefore, the spared senses that overtake the cortical area belonging to the deprived one maintain its modular structure. Once the occipital cortex reorganized to process inputs from nonvisual senses, the behavior will be similar. For example, a lesion of the occipital cortex will have effects on the nondeprived sense, such as alexia for Braille in a congenitally blind patient of stroke that mimics the effects of occipital stroke in no-blinds [122].

Apart from visual system, auditory system is one of the mostly deprived systems. Studies have shown that auditory deprivation leads to the recruitment of auditory areas to visual functions [123] or somatosensory functions [124]. In the same way, auditory stimulation activates the visual cortex of early visually deprived anophthalmic mice [125], and bilaterally enucleated rodents have an expanded somatosensory cortex [126].

The cross-modal interactions between somatosensory and other sensorial systems such as the auditory system also suggest that the effects of recovery strategies for sensorial deprivation are not only circumscribed to the deprived system [59].

Potentially, recovery from auditory deprivation may be closer to practical application in humans than from the other systems. Complete sensorial deprivation is frequent in humans suffering from congenital deafness and as the auditory nerve often is functional, therapies using cochlear implantations have a moderate rate of success, especially if applied before the auditory critical period and if they are combined with auditory enrichment by increasing parent-child interactions [127, 128]. As sometimes happens with handicapped children, the stress produced by a deaf child in hearing parents could lead to an involuntary impoverishing of the language interactions, thus, minimizing the beneficial effects of an early cochlear implantation.

One sense, less studied than the above mentioned is the sense of touch. Body massage and multisensory stimulation are included in neonatal care in human newborns due to their effects on neonates weight gain. In an example of cross-modal effects of early tactile enrichment during development, Guzzetta et al. [129] shows that massage influences the maturation of visual system, accelerating visual acuity development, both in human infants and in rat pups. This effect is exerted by overexpression of IGF in the whole brain. Visual system is not stimulated directly but massage effects have cross-modal effects over the development of this sense.

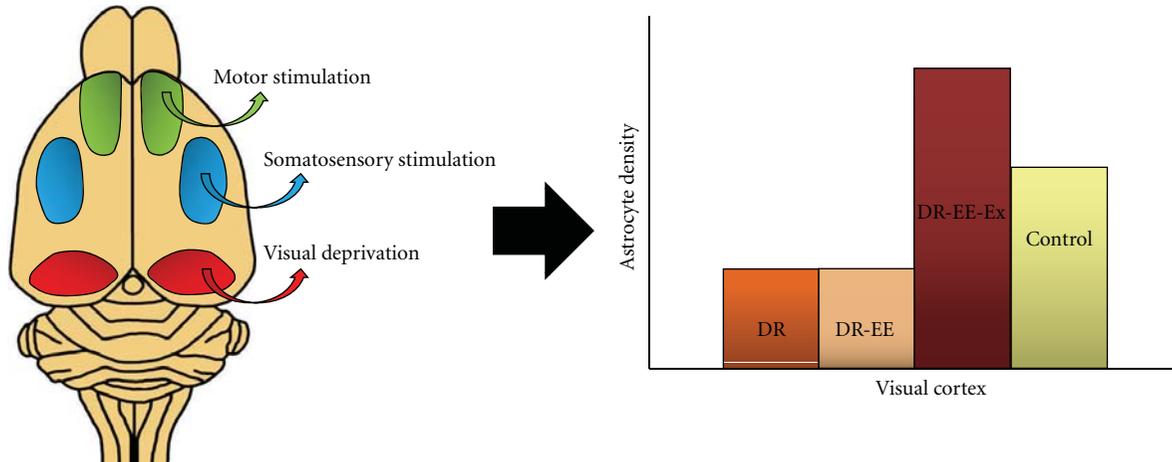


FIGURE 2: Schematic representation of enriched environment effects on dark-reared rats: motor stimulation, somatosensory stimulation, and visual deprivation caused by dark rearing. We have seen the effects on the astrocyte density of the visual cortex, where enrichment completed with exercise (DR-EE-Ex) can help to recover the loss of population caused by dark rearing (DR), reaching even higher level astrocyte density than the control group. And dark rearing with enriched environment (DR-EE) without running wheel shows similar values to DR group (adapted from results of [40]).

As cross-modal plasticity could interfere with the recovery of the original function, strategies intended to promote restorative plasticity should also involve the minimizing of interference from cross-modal inputs [130], which could be considered as a side effect of cross-modal plasticity.

Another point of great interest is the development of sensory substitution devices that could convey information of a deprived system (mostly visual) through other sources such as touch or sounds [131]. Cross-modal plasticity is revealed by electro-tactile stimulation of the tongue in the congenitally blind [132]. Although the use of neuroprostheses to restore vision in patients has been investigated [133], the results have been far from as convincing as cochlea implants, due to the lack of results and the high degree of invasiveness [134]. Another point of doubt on the use of prostheses is that, as happens with cochlea implants, the occipital cortex has been rewired and inputs from other senses have taken a space. In contrast, sensory substitution devices could benefit from the increasing knowledge of cross-modal plasticity and offer a cheaper, less invasive, and more efficient restorative tool. The fact that recent experimental findings show that the rewired occipital cortex maintains most of the modular features of the nonblind cortex, such as the specialization for spatial processing [121], provides a better substrate to convey complex visual experience from spared senses.

6. Conclusions

The lack of sensory inputs from environment during early postnatal development leads to serious consequences that can be reverted reactivating cortical plasticity by physiological strategies such as environmental enrichment, exposure to deprived inputs, or pharmacological solutions as neurotrophin administration. Although the time course of critical period during development is specific for each

sensory system, experience-mediated brain development is a unique event. A major feature of sensory deprivation is the cross-modal plasticity process that leads to a compensatory upregulation of the nondeprived senses that even invade the cortical territory of the deprived one. Thus, rehabilitation can be compromised as sensory inputs from the previously deprived sense have to compete with the sensory circuit neoformed by cross-modal plasticity. In conclusion, all restorative strategies against effects of sensory deprivation must take into account that the cortical areas belonging to the deprived sense have a newly established sensory organization that processes inputs from ectopic senses; thus, neuronal segregation is required to ensure that reactivation of the sense is able to form appropriate neuronal circuits. On the other hand, cross-modal plasticity offers a huge opportunity to develop sensory substitution devices that could enable blind individuals to acquire visual information from spared senses as auditory or tactile. The fact that the occipital cortex keeps the functional organization despite the lack of visual inputs, as happens with spatial processing, provides a higher potentiality of visual restoration by nonvisual inputs.

References

- [1] W. T. Greenough, J. E. Black, and C. S. Wallace, "Experience and brain development," *Child Development*, vol. 58, no. 3, pp. 539–559, 1987.
- [2] L. C. Katz and C. J. Shatz, "Synaptic activity and the construction of cortical circuits," *Science*, vol. 274, no. 5290, pp. 1133–1138, 1996.
- [3] T. K. Hensch, "Critical period regulation," *Annual Review of Neuroscience*, vol. 27, pp. 549–579, 2004.
- [4] T. A. Jones and S. C. Jefferson, "Reflections of experience-expectant development in repair of the adult damaged brain," *Developmental Psychobiology*, vol. 53, no. 5, pp. 466–475, 2011.

- [5] E. G. Argandoña and J. V. Lafuente, "Effects of dark-rearing on the vascularization of the developmental rat visual cortex," *Brain Research*, vol. 732, no. 1-2, pp. 43–51, 1996.
- [6] A. Briner, M. de Roo, A. Dayer, D. Muller, J. Z. Kiss, and L. Vutskits, "Bilateral whisker trimming during early postnatal life impairs dendritic spine development in the mouse somatosensory barrel cortex," *Journal of Comparative Neurology*, vol. 518, no. 10, pp. 1711–1723, 2010.
- [7] A. Kral and J. J. Eggermont, "What's to lose and what's to learn: development under auditory deprivation, cochlear implants and limits of cortical plasticity," *Brain Research Reviews*, vol. 56, no. 1, pp. 259–269, 2007.
- [8] K. Fox and R. O. L. Wong, "A comparison of experience-dependent plasticity in the visual and somatosensory systems," *Neuron*, vol. 48, no. 3, pp. 465–477, 2005.
- [9] M. Butz, F. Wörgötter, and A. van Ooyen, "Activity-dependent structural plasticity," *Brain Research Reviews*, vol. 60, no. 2, pp. 287–305, 2009.
- [10] F. Pouille, A. Marin-Burgin, H. Adesnik, B. V. Atallah, and M. Scanziani, "Input normalization by global feedforward inhibition expands cortical dynamic range," *Nature Neuroscience*, vol. 12, no. 12, pp. 1577–1585, 2009.
- [11] D. R. House, J. Elstrott, E. Koh, J. Chung, and D. E. Feldman, "Parallel regulation of feedforward inhibition and excitation during whisker map plasticity," *Neuron*, vol. 72, no. 5, pp. 819–831, 2011.
- [12] M. A. Meredith and S. G. Lomber, "Somatosensory and visual crossmodal plasticity in the anterior auditory field of early-deaf cats," *Hearing Research*, vol. 280, no. 1-2, pp. 38–47, 2011.
- [13] M. Ptito and S. Desgent, "Sensory input-based adaptation and brain architecture," in *Lifespan Development and the Brain*, P. Baltes, P. Reuter-Lorenz, and F. Rösler, Eds., pp. 111–123, Cambridge University Press, Cambridge, UK, 2006.
- [14] M. Fu and Y. Zuo, "Experience-dependent structural plasticity in the cortex," *Trends in Neurosciences*, vol. 34, no. 4, pp. 177–187, 2011.
- [15] A. M. Sirevaag, J. E. Black, D. Shafron, and W. T. Greenough, "Direct evidence that complex experience increases capillary branching and surface area in visual cortex of young rats," *Brain Research*, vol. 471, no. 2, pp. 299–304, 1988.
- [16] C. D. Gilbert, "Adult cortical dynamics," *Physiological Reviews*, vol. 78, no. 2, pp. 467–485, 1998.
- [17] H. Yao and Y. Dan, "Synaptic learning rules, cortical circuits, and visual function," *Neuroscientist*, vol. 11, no. 3, pp. 206–216, 2005.
- [18] J. E. Black, A. M. Sirevaag, and W. T. Greenough, "Complex experience promotes capillary formation in young rat visual cortex," *Neuroscience Letters*, vol. 83, no. 3, pp. 351–355, 1987.
- [19] R. V. Harrison, N. Harel, J. Panesar, and R. J. Mount, "Blood capillary distribution correlates with hemodynamic-based functional imaging in cerebral cortex," *Cerebral Cortex*, vol. 12, no. 3, pp. 225–233, 2002.
- [20] E. G. Argandoña, M. L. Rossi, and J. V. Lafuente, "Visual deprivation effects on the s100 β positive astrocytic population in the developing rat visual cortex: a quantitative study," *Developmental Brain Research*, vol. 141, no. 1-2, pp. 63–69, 2003.
- [21] S. B. Tieman, S. Möllers, D. G. Tieman, and J. White, "The blood supply of the cat's visual cortex and its postnatal development," *Brain Research*, vol. 998, no. 1, pp. 100–112, 2004.
- [22] B. W. Bakkum, L. A. Benevento, and R. S. Cohen, "Effects of light/dark- and dark-rearing on synaptic morphology in the superior colliculus and visual cortex of the postnatal and adult rat," *Journal of Neuroscience Research*, vol. 28, no. 1, pp. 65–80, 1991.
- [23] L. Cancedda, E. Putignano, A. Sale, A. Viegi, N. Berardi, and L. Maffei, "Acceleration of visual system development by environmental enrichment," *Journal of Neuroscience*, vol. 24, no. 20, pp. 4840–4848, 2004.
- [24] N. Berardi, T. Pizzorusso, and L. Maffei, "Extracellular matrix and visual cortical plasticity: freeing the synapse," *Neuron*, vol. 44, no. 6, pp. 905–908, 2004.
- [25] N. Berardi, T. Pizzorusso, and L. Maffei, "Critical periods during sensory development," *Current Opinion in Neurobiology*, vol. 10, no. 1, pp. 138–145, 2000.
- [26] T. Pizzorusso, P. Medini, N. Berardi, S. Chierzi, J. W. Fawcett, and L. Maffei, "Reactivation of ocular dominance plasticity in the adult visual cortex," *Science*, vol. 298, no. 5596, pp. 1248–1251, 2002.
- [27] D. Nowicka, S. Soulsby, J. Skangiel-Kramska, and S. Glazewski, "Parvalbumin-containing neurons, perineuronal nets and experience-dependent plasticity in murine barrel cortex," *European Journal of Neuroscience*, vol. 30, no. 11, pp. 2053–2063, 2009.
- [28] A. W. McGee, Y. Yang, Q. S. Fischer, N. W. Daw, and S. H. Strittmatter, "Neuroscience: experience-driven plasticity of visual cortex limited by myelin and nogo receptor," *Science*, vol. 309, no. 5744, pp. 2222–2226, 2005.
- [29] T. K. Hensch, "Critical period mechanisms in developing visual cortex," *Current Topics in Developmental Biology*, vol. 69, pp. 215–237, 2005.
- [30] M. Fagiolini, T. Pizzorusso, N. Berardi, L. Domenici, and L. Maffei, "Functional postnatal development of the rat primary visual cortex and the role of visual experience: dark rearing and monocular deprivation," *Vision Research*, vol. 34, no. 6, pp. 709–720, 1994.
- [31] A. Sale, J. F. M. Vetencourt, P. Medini et al., "Environmental enrichment in adulthood promotes amblyopia recovery through a reduction of intracortical inhibition," *Nature Neuroscience*, vol. 10, no. 6, pp. 679–681, 2007.
- [32] H. Y. He, W. Hodos, and E. M. Quinlan, "Visual deprivation reactivates rapid ocular dominance plasticity in adult visual cortex," *Journal of Neuroscience*, vol. 26, no. 11, pp. 2951–2955, 2006.
- [33] S. B. Hofer, T. D. Mrsic-Flogel, T. Bonhoeffer, and M. Hübener, "Prior experience enhances plasticity in adult visual cortex," *Nature Neuroscience*, vol. 9, no. 1, pp. 127–132, 2006.
- [34] E. Putignano, G. Lonetti, L. Cancedda et al., "Developmental downregulation of histone posttranslational modifications regulates visual cortical plasticity," *Neuron*, vol. 53, no. 5, pp. 747–759, 2007.
- [35] J. F. M. Vetencourt, A. Sale, A. Viegi et al., "The antidepressant fluoxetine restores plasticity in the adult visual cortex," *Science*, vol. 320, no. 5874, pp. 385–388, 2008.
- [36] K. Lehmann and S. Löwel, "Age-dependent ocular dominance plasticity in adult mice," *PLoS ONE*, vol. 3, no. 9, Article ID e3120, 2008.
- [37] E. Fifková, "The effect of unilateral deprivation on visual centers in rats," *Journal of Comparative Neurology*, vol. 140, no. 4, pp. 431–438, 1970.
- [38] B. G. Cragg, "Changes in visual cortex on first exposure of rats to light: effect on synaptic dimensions," *Nature*, vol. 215, no. 5098, pp. 251–253, 1967.
- [39] H. Bengoetxea, E. G. Argandoña, and J. V. Lafuente, "Effects of visual experience on vascular endothelial growth factor

- expression during the postnatal development of the rat visual cortex," *Cerebral Cortex*, vol. 18, no. 7, pp. 1630–1639, 2008.
- [40] E. G. Argandoña, H. Bengoetxea, and J. V. Lafuente, "Physical exercise is required for environmental enrichment to offset the quantitative effects of dark-rearing on the S-100 β astrocytic density in the rat visual cortex," *Journal of Anatomy*, vol. 215, no. 2, pp. 132–140, 2009.
- [41] G. D. Mower, D. Berry, J. L. Burchfiel, and F. H. Duffy, "Comparison of the effects of dark rearing and binocular suture on development and plasticity of cat visual cortex," *Brain Research*, vol. 220, no. 2, pp. 255–267, 1981.
- [42] M. T. Sato, A. Tokunaga, Y. Kawai, Y. Shimomura, Y. Tano, and E. Senba, "The effects of binocular suture and dark rearing on the induction of c-fos protein in the rat visual cortex during and after the critical period," *Neuroscience Research*, vol. 36, no. 3, pp. 227–233, 2000.
- [43] K. S. Bedi, "The combined effects of unilateral enucleation and rearing in a "dim" red light on synapse-to-neuron ratios in the rat visual cortex," *Journal of Anatomy*, vol. 167, pp. 71–84, 1989.
- [44] B. K. Bracken and G. G. Tyrrigiano, "Experience-dependent regulation of TrkB isoforms in rodent visual cortex," *Developmental Neurobiology*, vol. 69, no. 5, pp. 267–278, 2009.
- [45] L. Corvetto, E. Aztiria, and L. Domenici, "Reduction of GFAP induced by long dark rearing is not restricted to visual cortex," *Brain Research*, vol. 1067, no. 1, pp. 146–153, 2006.
- [46] C. M. Müller, "Dark-rearing retards the maturation of astrocytes in restricted layers of cat visual cortex," *Glia*, vol. 3, no. 6, pp. 487–494, 1990.
- [47] E. G. Argandoña, H. Bengoetxea, and J. V. Lafuente, "Lack of experience-mediated differences in the immunohistochemical expression of blood-brain barrier markers (EBA and GluT-1) during the postnatal development of the rat visual cortex," *Developmental Brain Research*, vol. 156, no. 2, pp. 158–166, 2005.
- [48] E. G. Argandoña and J. V. Lafuente, "Influence of visual experience deprivation on the postnatal development of the microvascular bed in layer IV of the rat visual cortex," *Brain Research*, vol. 855, no. 1, pp. 137–142, 2000.
- [49] K. Fox, "Anatomical pathways and molecular mechanisms for plasticity in the barrel cortex," *Neuroscience*, vol. 111, no. 4, pp. 799–814, 2002.
- [50] C. S. Wu, C. J. Ballester Rosado, and H. C. Lu, "What can we get from "barrels": the rodent barrel cortex as a model for studying the establishment of neural circuits," *European Journal of Neuroscience*, vol. 34, no. 10, pp. 1663–1676, 2011.
- [51] T. A. Woolsey and H. van der Loos, "The structural organization of layer IV in the somatosensory region (SI) of mouse cerebral cortex. The description of a cortical field composed of discrete cytoarchitectonic units," *Brain Research*, vol. 17, no. 2, pp. 205–242, 1970.
- [52] M. Brecht, "Barrel cortex and whisker-mediated behaviors," *Current Opinion in Neurobiology*, vol. 17, no. 4, pp. 408–416, 2007.
- [53] P. Li, U. Rudolph, and M. M. Huntsman, "Long-term sensory deprivation selectively rearranges functional inhibitory circuits in mouse barrel cortex," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 29, pp. 12156–12161, 2009.
- [54] C. H. Liu, A. J. Heynen, M. G. H. Shuler, and M. F. Bear, "Cannabinoid receptor blockade reveals parallel plasticity mechanisms in different layers of mouse visual cortex," *Neuron*, vol. 58, no. 3, pp. 340–345, 2008.
- [55] H. W. Tao and M. M. Poo, "Activity-dependent matching of excitatory and inhibitory inputs during refinement of visual receptive fields," *Neuron*, vol. 45, no. 6, pp. 829–836, 2005.
- [56] M. Shoykhet, P. W. Land, and D. J. Simons, "Whisker trimming begun at birth or on postnatal day 12 affects excitatory and inhibitory receptive fields of layer IV barrel neurons," *Journal of Neurophysiology*, vol. 94, no. 6, pp. 3987–3995, 2005.
- [57] E. Foeller, T. Celikel, and D. E. Feldman, "Inhibitory sharpening of receptive fields contributes to whisker map plasticity in rat somatosensory cortex," *Journal of Neurophysiology*, vol. 94, no. 6, pp. 4387–4400, 2005.
- [58] S. H. Lee, P. W. Land, and D. J. Simons, "Layer- and cell-type-specific effects of neonatal whisker-trimming in adult rat barrel cortex," *Journal of Neurophysiology*, vol. 97, no. 6, pp. 4380–4385, 2007.
- [59] A. Ghoshal, A. Tomarken, and F. Ebner, "Cross-sensory modulation of primary sensory cortex is developmentally regulated by early sensory experience," *Journal of Neuroscience*, vol. 31, no. 7, pp. 2526–2536, 2011.
- [60] S. B. Cox, T. A. Woolsey, and C. M. Rovainen, "Localized dynamic changes in cortical blood flow with whisker stimulation corresponds to matched vascular and neuronal architecture of rat barrels," *Journal of Cerebral Blood Flow and Metabolism*, vol. 13, no. 6, pp. 899–913, 1993.
- [61] B. de Celis Alonso, A. S. Lowe, J. P. Dear, K. C. Lee, S. C. R. Williams, and G. T. Finnerty, "Sensory inputs from whisking movements modify cortical whisker maps visualized with functional magnetic resonance imaging," *Cerebral Cortex*, vol. 18, no. 6, pp. 1314–1325, 2008.
- [62] J. Li, D. S. Bravo, A. Louise Upton et al., "Close temporal coupling of neuronal activity and tissue oxygen responses in rodent whisker barrel cortex," *European Journal of Neuroscience*, vol. 34, no. 12, pp. 1983–1996, 2011.
- [63] V. R. Whitaker, L. Cui, S. Miller, S. P. Yu, and L. Wei, "Whisker stimulation enhances angiogenesis in the barrel cortex following focal ischemia in mice," *Journal of Cerebral Blood Flow and Metabolism*, vol. 27, no. 1, pp. 57–68, 2007.
- [64] C. J. Angely and D. M. Coppola, "How does long-term odor deprivation affect the olfactory capacity of adult mice?" *Behavioral and Brain Functions*, vol. 25, no. 6, article 26, 2010.
- [65] L. Roux, K. Benchenane, J. D. Rothstein, G. Bonvento, and C. Giaume, "Plasticity of astroglial networks in olfactory glomeruli," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 45, pp. 18442–18446, 2011.
- [66] J. B. Lamarck, *Recherches sur l'Organisation des Corps Vivants*, 1808.
- [67] C. Darwin, *On the Origin of Species by Means of Natural Selection, or the Preservation of Favoured Races in the Struggle for Life*, J. Murray, London, UK, 1859.
- [68] S. R. Cajal, *Les Nouvelles Idées sur la Structure du Système Nerveux: Chez l'Homme et Chez les Vertébrés*, 1894.
- [69] M. S. C. S. Foster, *A Textbook of Physiology, Part Three: The Central Nervous System*, MacMillan & Co., London, UK, 1897.
- [70] J. A. Markham and W. T. Greenough, "Experience-driven brain plasticity: beyond the synapse," *Neuron Glia Biology*, vol. 1, no. 4, pp. 351–363, 2004.
- [71] D. O. Hebb, "The effects of early experience on problem solving at maturity," *American Psychologist*, vol. 2, pp. 306–307, 1947.

- [72] H. van Praag, G. Kempermann, and F. H. Gage, "Neural Consequences of environmental enrichment," *Nature Reviews Neuroscience*, vol. 1, no. 3, pp. 191–198, 2000.
- [73] B. Will, R. Galani, C. Kelche, and M. R. Rosenzweig, "Recovery from brain injury in animals: relative efficacy of environmental enrichment, physical exercise or formal training (1990–2002)," *Progress in Neurobiology*, vol. 72, no. 3, pp. 167–182, 2004.
- [74] M. C. Diamond, D. Krech, and M. R. Rosenzweig, "The effects of an enriched environment on the histology of the rat cerebral cortex," *The Journal of Comparative Neurology*, vol. 123, pp. 111–120, 1964.
- [75] M. C. Diamond, F. Law, H. Rhodes et al., "Increases in cortical depth and glia numbers in rats subjected to enriched environment," *Journal of Comparative Neurology*, vol. 128, no. 1, pp. 117–126, 1966.
- [76] F. R. Volkmar and W. T. Greenough, "Rearing complexity affects branching of dendrites in the visual cortex of the rat," *Science*, vol. 176, no. 4042, pp. 1445–1447, 1972.
- [77] W. T. Greenough, F. R. Volkmar, and J. M. Juraska, "Effects of rearing complexity on dendritic branching in frontolateral and temporal cortex of the rat," *Experimental Neurology*, vol. 41, no. 2, pp. 371–378, 1973.
- [78] J. O. Coq and C. Xerri, "Environmental enrichment alters organizational features of the forepaw representation in the primary somatosensory cortex of adult rats," *Experimental Brain Research*, vol. 121, no. 2, pp. 191–204, 1998.
- [79] C. Rampon, C. H. Jiang, H. Dong et al., "Effects of environmental enrichment on gene expression in the brain," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 23, pp. 12880–12884, 2000.
- [80] E. Nikolaev, L. Kaczmarek, S. W. Zhu, B. Winblad, and A. H. Mohammed, "Environmental manipulation differentially alters c-Fos expression in amygdaloid nuclei following aversive conditioning," *Brain Research*, vol. 957, no. 1, pp. 91–98, 2002.
- [81] T. A. Comery, R. Shah, and W. T. Greenough, "Differential rearing alters spine density on medium-sized spiny neurons in the rat corpus striatum: evidence for association of morphological plasticity with early response gene expression," *Neurobiology of Learning and Memory*, vol. 63, no. 3, pp. 217–219, 1995.
- [82] T. A. Comery, C. X. Stamoudis, S. A. Irwin, and W. T. Greenough, "Increased density of multiple-head dendritic spines on medium-sized spiny neurons of the striatum in rats reared in a complex environment," *Neurobiology of Learning and Memory*, vol. 66, no. 2, pp. 93–96, 1996.
- [83] W. T. Greenough, J. W. McDonald, R. M. Parnisari, and J. E. Camel, "Environmental conditions modulate degeneration and new dendrite growth in cerebellum of senescent rats," *Brain Research*, vol. 380, no. 1, pp. 136–143, 1986.
- [84] E. L. Bennett, M. R. Rosenzweig, and M. C. Diamond, "Rat brain: effects of environmental enrichment on wet and dry weights," *Science*, vol. 163, no. 3869, pp. 825–826, 1969.
- [85] M. C. Diamond, C. A. Ingham, and R. E. Johnson, "Effects of environment on morphology of rat cerebral cortex and hippocampus," *Journal of Neurobiology*, vol. 7, no. 1, pp. 75–85, 1976.
- [86] C. J. Faherty, D. Kerley, and R. J. Smeyne, "A Golgi-Cox morphological analysis of neuronal changes induced by environmental enrichment," *Developmental Brain Research*, vol. 141, no. 1-2, pp. 55–61, 2003.
- [87] M. G. Leggio, L. Mandolesi, F. Federico et al., "Environmental enrichment promotes improved spatial abilities and enhanced dendritic growth in the rat," *Behavioural Brain Research*, vol. 163, no. 1, pp. 78–90, 2005.
- [88] M. J. Doring and L. Cao, "VEGF, a mediator of the effect of experience on hippocampal neurogenesis," *Current Alzheimer Research*, vol. 3, no. 1, pp. 29–33, 2006.
- [89] D. Young, P. A. Lawlor, P. Leone, M. Dragunow, and M. J. Doring, "Environmental enrichment inhibits spontaneous apoptosis, prevents seizures and is neuroprotective," *Nature Medicine*, vol. 5, no. 4, pp. 448–453, 1999.
- [90] A. Sale, E. Putignano, L. Cancedda et al., "Enriched environment and acceleration of visual system development," *Neuropharmacology*, vol. 47, no. 5, pp. 649–660, 2004.
- [91] F. Szeligo and C. P. Leblond, "Response of the 3 main types of glial cells of cortex and corpus callosum in rats handled during suckling or exposed to enriched, control and impoverished environments following weaning," *Journal of Comparative Neurology*, vol. 172, no. 2, pp. 247–263, 1977.
- [92] A. M. Sirevaag and W. T. Greenough, "Differential rearing effects on rat visual cortex synapses. III. Neuronal and glial nuclei, boutons, dendrites, and capillaries," *Brain Research*, vol. 424, no. 2, pp. 320–332, 1987.
- [93] A. M. Sirevaag and W. T. Greenough, "Plasticity of GFAP-immunoreactive astrocyte size and number in visual cortex of rats reared in complex environments," *Brain Research*, vol. 540, no. 1-2, pp. 273–278, 1991.
- [94] J. E. Black, A. M. Zelazny, and W. T. Greenough, "Capillary and mitochondrial support of neural plasticity in adult rat visual cortex," *Experimental Neurology*, vol. 111, no. 2, pp. 204–209, 1991.
- [95] S. Zacchigna, D. Lambrechts, and P. Carmeliet, "Neurovascular signalling defects in neurodegeneration," *Nature Reviews Neuroscience*, vol. 9, no. 3, pp. 169–181, 2008.
- [96] T. M. Pham, B. Winblad, A. C. Granholm, and A. H. Mohammed, "Environmental influences on brain neurotrophins in rats," *Pharmacology Biochemistry and Behavior*, vol. 73, no. 1, pp. 167–175, 2002.
- [97] T. B. Franklin, J. A. Murphy, T. L. Myers, D. B. Clarke, and R. W. Currie, "Enriched environment during adolescence changes brain-derived neurotrophic factor and TrkB levels in the rat visual system but does not offer neuroprotection to retinal ganglion cells following axotomy," *Brain Research*, vol. 1095, no. 1, pp. 1–11, 2006.
- [98] B. R. Ickes, T. M. Pham, L. A. Sanders, D. S. Albeck, A. H. Mohammed, and A. C. Granholm, "Long-term environmental enrichment leads to regional increases in neurotrophin levels in rat brain," *Experimental Neurology*, vol. 164, no. 1, pp. 45–52, 2000.
- [99] F. Naka, N. Narita, N. Okado, and M. Narita, "Modification of AMPA receptor properties following environmental enrichment," *Brain and Development*, vol. 27, no. 4, pp. 275–278, 2005.
- [100] P. K. Dash, S. A. Orsi, and A. N. Moore, "Histone deacetylase inhibition combined with behavioral therapy enhances learning and memory following traumatic brain injury," *Neuroscience*, vol. 163, no. 1, pp. 1–8, 2009.
- [101] J. C. Bennett, P. A. McRae, L. J. Levy, and K. M. Frick, "Long-term continuous, but not daily, environmental enrichment reduces spatial memory decline in aged male mice," *Neurobiology of Learning and Memory*, vol. 85, no. 2, pp. 139–152, 2006.

- [102] G. Segovia, A. del Arco, and F. Mora, "Environmental enrichment, prefrontal cortex, stress, and aging of the brain," *Journal of Neural Transmission*, vol. 116, no. 8, pp. 1007–1016, 2009.
- [103] N. Benaroya-Milshtein, N. Hollander, A. Apter et al., "Environmental enrichment in mice decreases anxiety, attenuates stress responses and enhances natural killer cell activity," *European Journal of Neuroscience*, vol. 20, no. 5, pp. 1341–1347, 2004.
- [104] A. Bartoletti, P. Medini, N. Berardi, and L. Maffei, "Environmental enrichment prevents effects of dark-rearing in the rat visual cortex," *Nature Neuroscience*, vol. 7, no. 3, pp. 215–216, 2004.
- [105] O. B. Amaral, R. S. Vargas, G. Hansel, I. Izquierdo, and D. O. Souza, "Duration of environmental enrichment influences the magnitude and persistence of its behavioral effects on mice," *Physiology & Behavior*, vol. 93, no. 1-2, pp. 388–394, 2008.
- [106] J. Nithianantharajah, C. Barkus, M. Murphy, and A. J. Hannan, "Gene-environment interactions modulating cognitive function and molecular correlates of synaptic plasticity in Huntington's disease transgenic mice," *Neurobiology of Disease*, vol. 29, no. 3, pp. 490–504, 2008.
- [107] C. J. Faherty, K. R. Shepherd, A. Herasimtschuk, and R. J. Smeyne, "Environmental enrichment in adulthood eliminates neuronal death in experimental Parkinsonism," *Molecular Brain Research*, vol. 134, no. 1, pp. 170–179, 2005.
- [108] C. E. McOmish, E. Burrows, M. Howard et al., "Phospholipase C- β 1 knock-out mice exhibit endophenotypes modeling schizophrenia which are rescued by environmental enrichment and clozapine administration," *Molecular Psychiatry*, vol. 13, no. 7, pp. 661–672, 2008.
- [109] J. P. Rauschecker, "Compensatory plasticity and sensory substitution in the cerebral cortex," *Trends in Neurosciences*, vol. 18, no. 1, pp. 36–43, 1995.
- [110] L. B. Merabet and A. Pascual-Leone, "Neural reorganization following sensory loss: the opportunity of change," *Nature Reviews Neuroscience*, vol. 11, no. 1, pp. 44–52, 2010.
- [111] D. E. Feldman and M. Brecht, "Map plasticity in somatosensory cortex," *Science*, vol. 310, no. 5749, pp. 810–815, 2005.
- [112] P. Voss and R. J. Zatorre, "Organization and reorganization of sensory-deprived cortex," *Current Biology*, vol. 22, no. 5, pp. R168–R173, 2012.
- [113] G. Bronchti, P. Heil, R. Sadka, A. Hess, H. Scheich, and Z. Wollberg, "Auditory activation of "visual" cortical areas in the blind mole rat (*Spalax ehrenbergi*)," *European Journal of Neuroscience*, vol. 16, no. 2, pp. 311–329, 2002.
- [114] E. G. Argandoña, R. Ledezma, H. Bengoetxea et al., "Physical exercise increases acute adaptive angiogenic response to moderate altitude," in *Proceedings of the 8th International Brain Research Organization World Congress of Neuroscience*, vol. 2, p. 992, F1000 Posters, July 2011.
- [115] T. Cotrufo, A. Viegi, N. Berardi, Y. Bozzi, L. Mascia, and L. Maffei, "Effects of neurotrophins on synaptic protein expression in the visual cortex of dark-reared rats," *Journal of Neuroscience*, vol. 23, no. 9, pp. 3566–3571, 2003.
- [116] W. C. Lee, H. Huang, G. Feng et al., "Dynamic remodeling of dendritic arbors in GABAergic interneurons of adult visual cortex," *PLoS biology*, vol. 4, no. 2, Article ID e29, 2006.
- [117] Y. Jiao, Z. Zhang, C. Zhang et al., "A key mechanism underlying sensory experience-dependent maturation of neocortical GABAergic circuits in vivo," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 29, pp. 12131–12136, 2011.
- [118] J. M. Miller, C. G. Le Prell, D. M. Prieskorn, N. L. Wys, and R. A. Altschuler, "Delayed neurotrophin treatment following deafness rescues spiral ganglion cells from death and promotes regrowth of auditory nerve peripheral processes: effects of brain-derived neurotrophic factor and fibroblast growth factor," *Journal of Neuroscience Research*, vol. 85, no. 9, pp. 1959–1969, 2007.
- [119] C. C. Chan, A. W. Wong, K. H. Ting, S. Whitfield-Gabrieli, J. He, and T. M. Lee, "Cross auditory-spatial learning in early-blind individuals," *Human Brain Mapping*. In press.
- [120] L. B. Lewis, M. Saenz, and I. Fine, "Mechanisms of cross-modal plasticity in early-blind subjects," *Journal of Neurophysiology*, vol. 104, no. 6, pp. 2995–3008, 2010.
- [121] O. Collignon, F. Champoux, P. Voss, and F. Lepore, "Sensory rehabilitation in the plastic brain," *Progress in Brain Research*, vol. 191, pp. 211–231, 2011.
- [122] R. Hamilton, J. P. Keenan, M. Catala, and A. Pascual-Leone, "Alexia for Braille following bilateral occipital stroke in an early blind woman," *NeuroReport*, vol. 11, no. 2, pp. 237–240, 2000.
- [123] E. M. Finney, I. Fine, and K. R. Dobkins, "Visual stimuli activate auditory cortex in the deaf," *Nature Neuroscience*, vol. 4, no. 12, pp. 1171–1173, 2001.
- [124] A. Sharma, "Special issue on central auditory system development and plasticity," *International Journal of Audiology*, vol. 46, no. 9, p. 459, 2007.
- [125] M. Piché, S. Robert, D. Miceli, and G. Bronchti, "Environmental enrichment enhances auditory takeover of the occipital cortex in anophthalmic mice," *European Journal of Neuroscience*, vol. 20, no. 12, pp. 3463–3472, 2004.
- [126] S. J. Karlen and L. Krubitzer, "Effects of bilateral enucleation on the size of visual and nonvisual areas of the brain," *Cerebral Cortex*, vol. 19, no. 6, pp. 1360–1371, 2009.
- [127] A. M. Robbins, D. B. Koch, M. J. Osberger, S. Zimmerman-Phillips, and L. Kishon-Rabin, "Effect of age at cochlear implantation on auditory skill development in infants and toddlers," *Archives of Otolaryngology—Head & Neck Surgery*, vol. 130, no. 5, pp. 570–574, 2004.
- [128] A. Kral and A. Sharma, "Developmental neuroplasticity after cochlear implantation," *Trends in Neurosciences*, vol. 35, no. 2, pp. 111–122, 2011.
- [129] A. Guzzetta, S. Baldini, A. Bancalè et al., "Massage accelerates brain development and the maturation of visual function," *Journal of Neuroscience*, vol. 29, no. 18, pp. 6042–6051, 2009.
- [130] Y. T. Mao, T. M. Hua, and S. L. Pallas, "Competition and convergence between auditory and cross-modal visual inputs to primary auditory cortical areas," *Journal of Neurophysiology*, vol. 105, no. 4, pp. 1558–1573, 2011.
- [131] T. Ortiz, J. Poch, J. M. Santos et al., "Recruitment of occipital cortex during sensory substitution training linked to subjective experience of seeing in people with blindness," *PLoS One*, vol. 6, no. 8, Article ID e23264, 2011.
- [132] M. Ptito, S. M. Moesgaard, A. Gjedde, and R. Kupers, "Cross-modal plasticity revealed by electrotactile stimulation of the tongue in the congenitally blind," *Brain*, vol. 128, no. 3, pp. 606–614, 2005.
- [133] J. D. Weiland, A. K. Cho, and M. S. Humayun, "Retinal prostheses: current clinical results and future needs," *Ophthalmology*, vol. 118, no. 11, pp. 2227–2237, 2011.
- [134] E. Striem-Amit, U. Hertz, and A. Amedi, "Extensive cochleotopic mapping of human auditory cortical fields obtained with phase-encoding fMRI," *PLoS ONE*, vol. 6, no. 3, Article ID e17832, 2011.

Review Article

Adaptive Neuroplastic Responses in Early and Late Hemispherectomized Monkeys

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Received 13 February 2012; Revised 29 March 2012; Accepted 12 April 2012

Academic Editor: Pietro Pietrini

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Behavioural recovery in children who undergo medically required hemispherectomy showcase the remarkable ability of the cerebral cortex to adapt and reorganize following insult early in life. Case study data suggest that lesions sustained early in childhood lead to better recovery compared to those that occur later in life. In these children, it is possible that neural reorganization had begun prior to surgery but was masked by the dysfunctional hemisphere. The degree of neural reorganization has been difficult to study systematically in human infants. Here we present a 20-year culmination of data on our nonhuman primate model (*Chlorocebus sabeus*) of early-life hemispherectomy in which behavioral recovery is interpreted in light of plastic processes that lead to the anatomical reorganization of the early-damaged brain. The model presented here suggests that significant functional recovery occurs after the removal of one hemisphere in monkeys with no preexisting neurological dysfunctions. Human and primate studies suggest a critical role for subcortical and brainstem structures as well as corticospinal tracts in the neuroanatomical reorganization which result in the remarkable behavioral recovery following hemispherectomy. The non-human primate model presented here offers a unique opportunity for studying the behavioral and functional neuroanatomical reorganization that underlies developmental plasticity.

1. Introduction

1.1. Prologue. Cerebral hemicorticectomy is a form of radical surgical intervention currently used in the treatment of intractable epilepsy [1] and malignant tumors [2] accompanied by infantile hemiplegia [3, 4]. Neurological and behavioral functions are remarkably improved following the removal of the entire cerebral hemisphere, not only in infants but also in adults, with the recovery being greater for the early-lesioned subjects [5, 6]. Although hemispherectomized patients may go on to lead full lives, it is not complete and individuals have lingering behavioral manifestations [7].

The degree of recovery largely depends on the system being investigated. For example, motor functions are improved postoperatively, locomotion is preserved, and the hemiplegia of the contralateral limb is ameliorated with the apparition of simple voluntary movements [4]. Thresholds for touch, pain and temperature are elevated [4, 8, 9],

and localization and discriminative abilities are diminished. Sensory functions are better preserved for the face and the leg and are worsened for the forearm and the hand [4, 9]. At the visual level, there is a persistent contralateral hemianopia similar to the one observed following massive damage to the primary visual area. In clinical settings, functional reorganization may be masked by the dysfunctional hemisphere, providing the illusion of a rapid behavioral recovery [10, 11].

In recent years, we have developed a primate model of human hemispherectomy that allowed us to study behavioral recovery and its underlying anatomical substrates [12, 13]. This model eliminates the potential contamination of residual cortical areas and their projections that can participate in the reorganization process, as is the case in studies using discrete lesions as well as potential presurgery reorganization [10]. Furthermore, by sparing subcortical structures such as striatum, diencephalon, and brainstem, this model provides insights into the mechanisms involved in the magnitude of

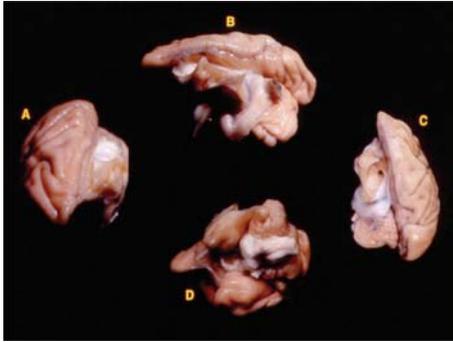


FIGURE 1: The complete removal of the left hemisphere was performed during infancy. Here, we show the results of the near complete removal of the left hemisphere four years after the initial surgery in the axial plane (A and C), lateral view (B), and orbital view (D).

behavioral recovery. We summarize in this paper results on longitudinal behavioral assessment of sensory and motor functions as well as a histological overview of brain reorganization with an emphasis on potential neural substrates for behavioral recovery.

1.2. General Experimental Procedure

1.2.1. Subjects. Eight infants (median age of 9 weeks) and two adult (48 months of age) Vervet monkeys (*Chlorocebus sabaeus*) were used in these studies and underwent the removal of the entire left cerebral hemisphere. An additional 2 adult monkeys without surgical procedures were used as normal controls. All subjects were housed in an enriched naturalistic environment at the facilities of the Behavioral Sciences Foundation, St Kitts as previously described [14]. The experimental protocol was reviewed and approved by the University of Montreal Animal Care and Use Committee.

1.2.2. Surgery. Using previously described surgical procedures [14, 15], a craniotomy was performed under deep general anesthesia and the left hemisphere was gently retracted from the midline and separated from the diencephalon using a suction pipette (Figure 1). After surgery, all monkeys received postoperative injections of antibiotics for a period of 10 days, and the infant monkeys were returned to their mother until the age of six months. Thereafter, they were housed in a nursery setting for an additional 6 months, at which point they were placed in a social group in a large enriched enclosure (3 m × 2 m × 3 m).

1.2.3. Behavioral Assessment. In general the subjects appeared to have normal behaviour within their respective social groups with normal peer-to-peer interactions. Feeding behaviour seemed affected with the subjects holding themselves with their left arm and bending down to pick up food with their mouths instead of their right hand. Hemispherectomized subjects were able to groom and had a normal body weight for their respective ages indicating

that the removal of the left hemisphere did not affect normal growth or health. The feeding behaviour, however, was indicative of a paresis on the left side. A series of sensory and motor assessments was initiated to test recovery following surgery. Visual assessment was conducted one year following hemispherectomy in both the infant- and adult-lesioned subjects. Thermal sensitivity was assessed in infant-lesioned subjects at three years of age [12, 13]. Motor behaviour was evaluated for three consecutive years after surgery for the infant-lesioned subjects and four years following surgery in the adult-lesioned subjects [14]. As a point of reference, for all behavioural and neural recovery, ipsilateral refers to the left side or that of which the hemisphere was removed; contralateral refers to the right side of the body or that of which the hemisphere remained intact. Behavioural assessments were recorded and analyzed frame-by-frame by two independent observers according to previously published reports [12–14].

2. Sensory and Motor Assessment

2.1. Vision. Abnormal environmental inputs, during the critical period of development either through sensory deprivation (e.g., eyelid suturing, dark rearing, and enucleation) or cortical injuries lead to dramatic changes at the cellular level and in brain circuitry (reviewed in [16, 17]). The mechanisms underlying visual recovery from large cortical lesions associated with brain plasticity are still unclear and remains an upmost challenge in understanding human patients with lesions restricted to the primary visual cortex (area V1) and those with massive lesions that include all of the visual cortical areas of one cerebral hemisphere (as in hemispherectomy). Animal models suggest that an early unilateral lesion of the visual cortex induces a loss of the contralateral visual field that subsides with time, leading to a complete visual field recovery (see [17]). Neonate hamsters with induced ectopic retinal projections to nonvisual thalamic targets (auditory nucleus and hence auditory cortex) perform as well as normal controls in visual pattern discriminations [18]. In monkeys and humans, the mechanisms underlying recovery of vision in the blind hemifield are not as clear. Only a few cases of spontaneous visual field recovery have been reported in patients born with neonatal malformations of the occipital lobes [19, 20], with recovery attributed to the intact tissue in or surrounding the lesioned area [19, 21] or the contralateral hemisphere [22]. In adults suffering from acquired visual field loss, intensive training through methods such as visual restoration therapy (VRT) can also lead to the reduction of the blind hemifield [23–27]. Here we summarize data collected on basic visual functions as a function of age.

2.1.1. Perimetry. The extent of the visual field for both eyes was measured according to the technique used in the cat [28, 29] and adapted to the monkey [12, 13] one year after the surgery as previously reported [12]. Briefly, the subject was placed in a restraining chair positioned at the center of a perimeter. The monkey was trained to fixate a target

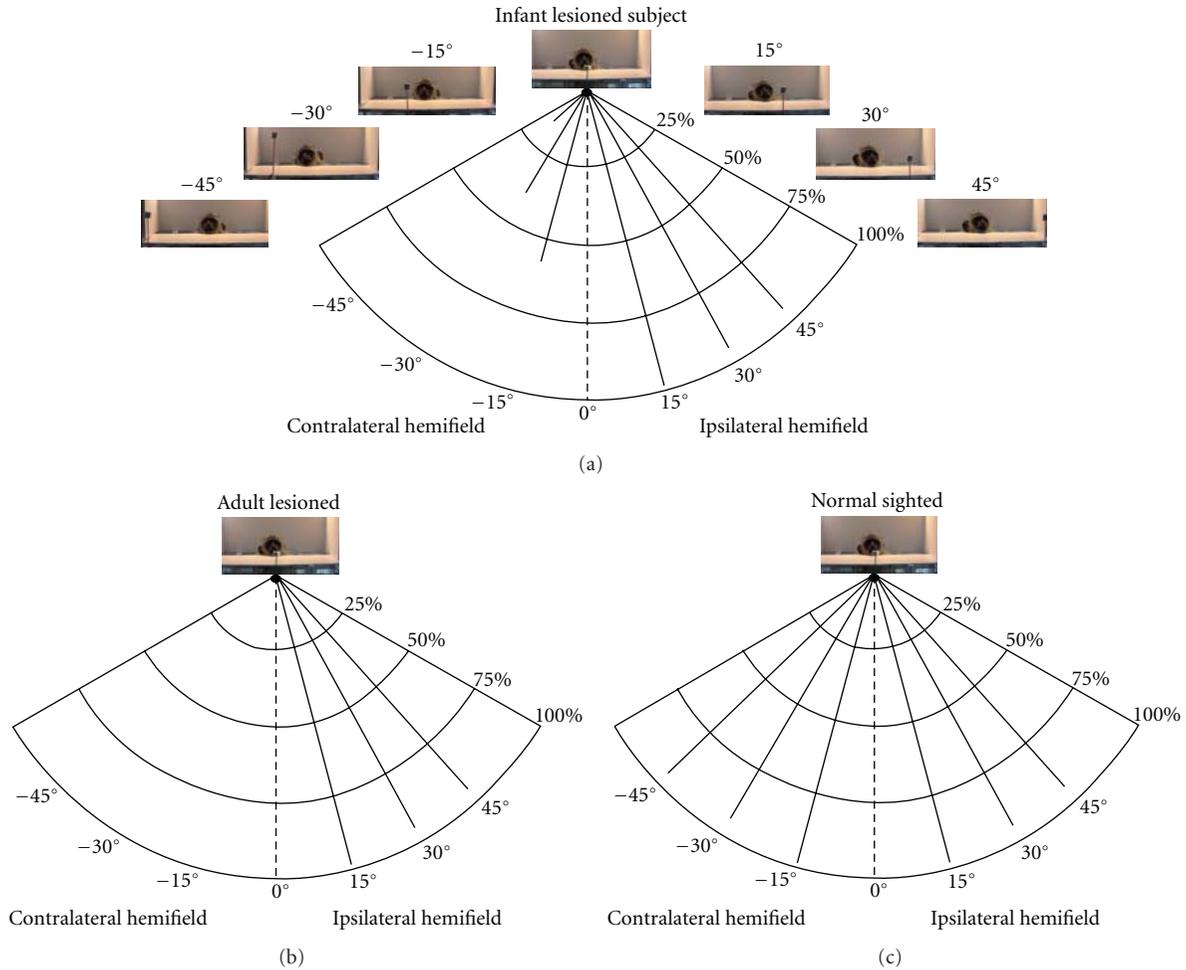


FIGURE 2: Perimetry: infant-lesioned monkeys (a) were able to detect visual stimuli at 45° in the “blind” hemifield, whereas no visual response could be elicited in the adult-lesioned subjects (b) in the contralateral hemifield. Normal-sighted monkeys had a 75% visual perimetry at 90° in both hemifields (c) (adapted from [13]).

(3° of visual angle) at the center of the perimeter positioned at 27 cm from the eyes. A second stimulus (a morsel of fruit on a stick about 1 cm² in size) was then randomly introduced in the visual field at various eccentricities (14 at 15° steps: 0°, 15°, 30°, 45°, 60°, 75°, and 90°, left and right visual fields), and the monkey had to orient its gaze in response to this new stimulus (Figure 2). Visual assessment was performed in three infant monkeys, and the average percent looking behavior is reported here. Adult-lesioned monkeys did not display any type of looking behavior in the blind hemifield in response to visual stimuli. For the control subjects, visual response to stimuli in both hemifields was equal [13].

Orientating responses were apparent in the hemifield contralateral to the lesion in the infant-lesioned subjects. These subjects responded 53%, 35%, and 16% of the time to stimuli presented at 15°, 30°, and 45°, respectively in the blind hemifield (Figure 2(a)). No responses could be elicited beyond 45°. Orientating responses were not seen in the blind hemifield of the adult-lesioned monkeys (Figure 2(b)). In the normal hemifield, occasional errors (e.g., absence of orienting responses to the target) were seen in only the

far periphery, a result usually found in normal animals (Figure 2(c)) [13].

2.1.2. Visual Palpebral Reflex and Visual Pursuit. This reflex was tested, with the monkey in a chair, by a sudden thrust of an object toward the open eyes (first at both eyes from the center and then at each eye independently from the sides), and recording if a blink response occurred. Visual pursuit was assessed while an object was slowly moving in the visual field from left to right or right to left. The visual palpebral reflex was always absent in the contralateral visual field for both infant- and adult-lesioned groups. Visual pursuit, when the stimulus started in the intact field, generally stopped at the body midline (i.e., vertical meridian). Conversely, when the stimulus started in the blind field, visual pursuit began only when it reached the midline for both groups of lesioned subjects. For the controls, visual pursuit was smooth in both directions [13].

2.2. Motor. Clinical and nonhuman primate data indicate that hemispherectomy results in hemiparesis. Subjects may

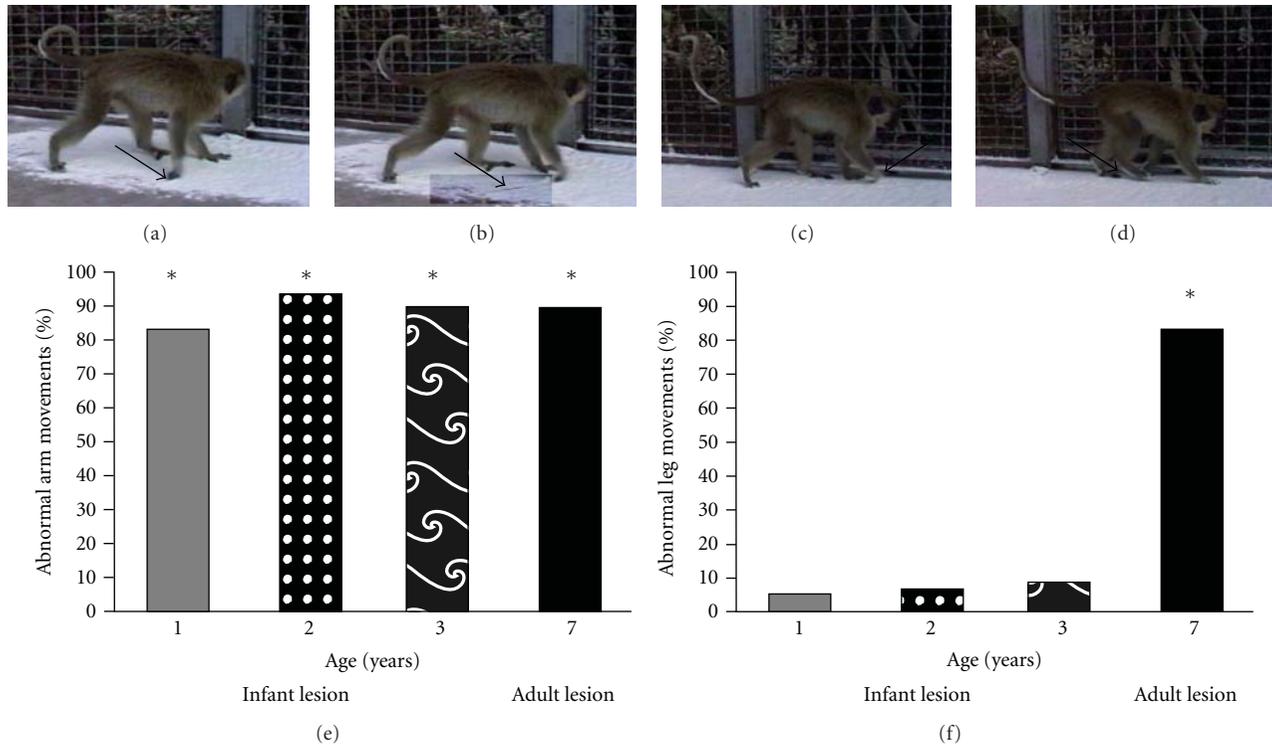


FIGURE 3: Normal gait was significantly affected by the removal of one hemisphere. The ipsilateral side of the body acts as a control for normal motor movements and did not show any paresis, also referred to as the nonparetic side of the subject. Panels (a)–(d) are indicative of a typical gait sequence. As the forelimb moves forward, the contralateral appendages drag on the ground (arrow panel a). The hand dragging continues through the entire forward motion (arrow panels b and c). The hind limb is fully removed from the ground and does not drag (arrow d). At each time point and lesion group there is a significant effect of the hemispherectomy on upper limb movement defined as arm drags/total forward arm movements (e). A number of abnormal leg movements (drags and limps) as a percentage of total leg movements was significantly elevated over the expected rate of zero in the adult-lesioned group only (f). Years 1, 2, and 3 refer to the age of the infant-lesioned subjects at testing which also corresponds to the years after lesion. For the adult-lesioned subjects, testing occurred 3 years after initial surgery when the subjects were 7 years old. * $P < 0.001$ adapted from [14].

regain muscle strength in the leg but continue to show weakness in the contralateral arm and hand [5, 30]. In young subjects, motor functions are improved postoperatively, locomotion is preserved, and the hemiplegia of the contralateral limb is improved and replaced by simple voluntary movements [4, 31]. Hemiparetic recovery following hemispherectomy, especially in younger children, has been associated with reorganization of the remaining cortex [32], prior to surgery [11, 33]. Here we describe a series of gross motor tasks to determine the extent of recovery following infant versus adult hemispherectomy.

2.2.1. Open Field. At yearly intervals beginning one year after surgery, infant-lesioned monkeys were assessed for spontaneous gross motor behavior according to previously published methods [14]. Adult-lesioned monkeys were moved into an empty adjoining enclosure 6 months after surgery and assessed individually for spontaneous behavior three years after surgery. Upper and lower limb movements were scored normal for full lift off the ground or abnormal for dragging of the appendage. Limping behavior was also scored as an abnormal movement but was analyzed separately from appendage dragging [14].

Normal gait was observed on the ipsilateral side with the appendages clearly being lifted off the ground as the limbs moved forward for both age groups at each observation period. Both infant- and adult-lesioned subjects had pervasive upper limb dysfunction with 90% of total arm movements resulting in the hand being dragged along the ground (Figure 3). Lower limb movement difficulties were less frequently observed in infant-lesioned subjects with less than 10% of total movements resulting in dragging. The lower limb in the adult-lesioned subjects was also less affected with 16% of the leg movements being accompanied with a foot drag. However, when taking into account limping as an abnormal movement, the lower limb in the adult-lesioned monkeys was significantly affected. This suggests that lower limb recovery in the young-lesioned animals is much stronger than the upper limb that remains hemiparetic [14].

2.2.2. Horizontal Bar Crossing. The vervet monkey is an agile species with the ability to cross a horizontal bar that is a complex visuo-spatial-motor task, from a very young age. At yearly intervals, infant-lesioned subjects were assessed for their ability to perform bar crossing. Adult-lesioned subjects

were moved into an empty adjoining enclosure 6 months after surgery and assessed individually for spontaneously crossing the bar. Behavior was scored as follows: (1) attempt to grab the bar with the contralateral fore- or hind-limbs was scored either as a successful or unsuccessful grab, with the successful attempt being defined as the contralateral appendage fully grasping the bar and (2) no attempt to grab the bar while walking across the horizontal bar defined as a movement across the bar using only the ipsilateral limbs/appendages. Video was analyzed frame-by-frame for each time point and group (see Burke et al., for more details [14]).

The infant-lesioned subjects were unable to transverse the bar by walking upright as a normal monkey for the first 2 years following surgery; instead, they would transverse the bar hanging upside down from the bar. The subjects would hang onto the bar with the ipsilateral limbs, overreach the bar with the contralateral limb such that the entire arm or leg was completely extended above the bar, and then glide the limb down the bar until either the hand or foot was able to latch onto the bar (Figure 4). The monkeys were typically unable to transverse the entire length of the bar without falling to the ground. By the third year, the monkeys started to use a new strategy for crossing the horizontal bar by not attempting to use the contralateral limb. When the monkeys attempted to use the contralateral arm on the horizontal bar, they displayed a 100% success rate. However, 88% of the movements across the bar did not involve the contralateral hand. The subjects did attempt to use the contralateral leg 79% of the time and had an 88% success rate when the leg was used. The adult-lesioned subjects were able to successfully walk upright the entire length of the horizontal bar. When the monkeys attempted to use the contralateral arm on the horizontal bar, they displayed a 25% success rate; however, the vast majority of movements across the bar did not involve the contralateral hand [14].

2.2.3. Turning Behavior. During open field motor assessment, turning preference was scored as a 90° or 180° turn ipsi- or contralateral to the lesioned side of the body. An average of 172 ± 43 turning behaviors were scored, and at each time point a consistent ipsilateral turning preference was recorded. At 1 year of age, 70% of turns were ipsilateral. This increased to 86% and 89% at the age of 2 and 3, respectively, whereas the adult-lesioned animals had a 94% ipsilateral turning preference. There were no differences in turning behavior between age groups, or infant- versus adult-lesioned subjects indicating that this function never recovered over time, with the animals still showing a marked preference for the nonblind hemifield [14].

2.3. Temperature Sensitivity. Previous studies have shown that hemispherectomy patients are able to perceive tactile stimulation applied to their paretic leg. Functional magnetic resonance imaging (fMRI) studies further showed that this type of stimulation leads to activation of the intact primary and secondary somatosensory cortices, suggesting the existence of ipsilateral, nondecussating pathways from

the periphery [34, 35]. Here we measured thermal sensitivity of the upper and lower limbs as a measure of residual somatosensation in infant-lesioned subjects.

The subjects were placed in a restraining chair with their arms and legs freely moving. The subjects' fingers or toes were randomly immersed into recipients containing water at varying degrees of temperature (0, 10, 20, 30, 40, and 50°C). Whereas the 30°C can be considered as a neutral temperature, the 20 and 40°C are innocuous cold and warm stimuli, respectively. The 10 and 0°C temperatures fall within the noxious cold range whereas the 50°C is a noxious heat stimulus. If the monkey did not withdraw the appendage from the water within 16 seconds, the trial was terminated and counted as a nonresponse (Figures 5(c) and 5(d)). Two years after the surgery both upper (fingers) and lower (toes) limbs (ipsi- and contra-) were tested at each temperature, and the withdrawal reaction time was recorded via frame-by-frame analysis of the video (Figure 5). Appendage withdrawal responses were analyzed in three monkeys, and time of withdrawal was compared by *t*-test between ipsi- and contralateral appendages.

In line with our expectations, withdrawal times of the ipsilateral upper and lower limb for the neutral and innocuous temperatures were longer than for the noxious stimuli (Figure 5). Whereas the animal withdrew the limb in less than 30% of the cases for the neutral temperatures, this increased to 100% for the painful cold and heat stimuli. For the ipsilateral lower limb, the average withdrawal times for neutral, innocuous warm and noxious heat stimuli were 7.33 ± 2.49 , 2.57 ± 0.72 , and 1.36 ± 0.2 , seconds respectively, (Figure 5). A similar response pattern was observed for the contralateral stimuli with this noticeable difference that withdrawal times were significantly longer for all tested temperatures. The percentage of withdrawal responses to noxious cold and heat stimuli remained close to 100% for the contralateral limb. For the contralateral upper limb, average withdrawal times and percentage of withdrawals did not differ for the neutral and innocuous temperatures. However, for the lower limb, withdrawal times were shorter for the innocuous compared to the neutral temperatures. For both the upper and lower limb, withdrawal times for the noxious temperatures were shorter than for the innocuous temperatures. Together, these data suggest that while the contralateral limbs retained thermal sensitivity, there was a clear impairment in the perception of the innocuous warm and cold temperatures, especially for the upper limb.

3. Discussion

The ability of the cerebral cortex to adapt and reorganize following insult early in life is remarkable but has been difficult to study systematically in human infants. Data has been mostly accumulated from case studies and suggest that lesions sustained in early childhood lead to better recovery [36] indicating a prominent anatomical reorganization in human subjects [37–39]. Here we present a 20-year culmination of data on our nonhuman primate model of early-life hemispherectomy. For individuals with medically required hemispherectomy, it is possible that neural reorganization

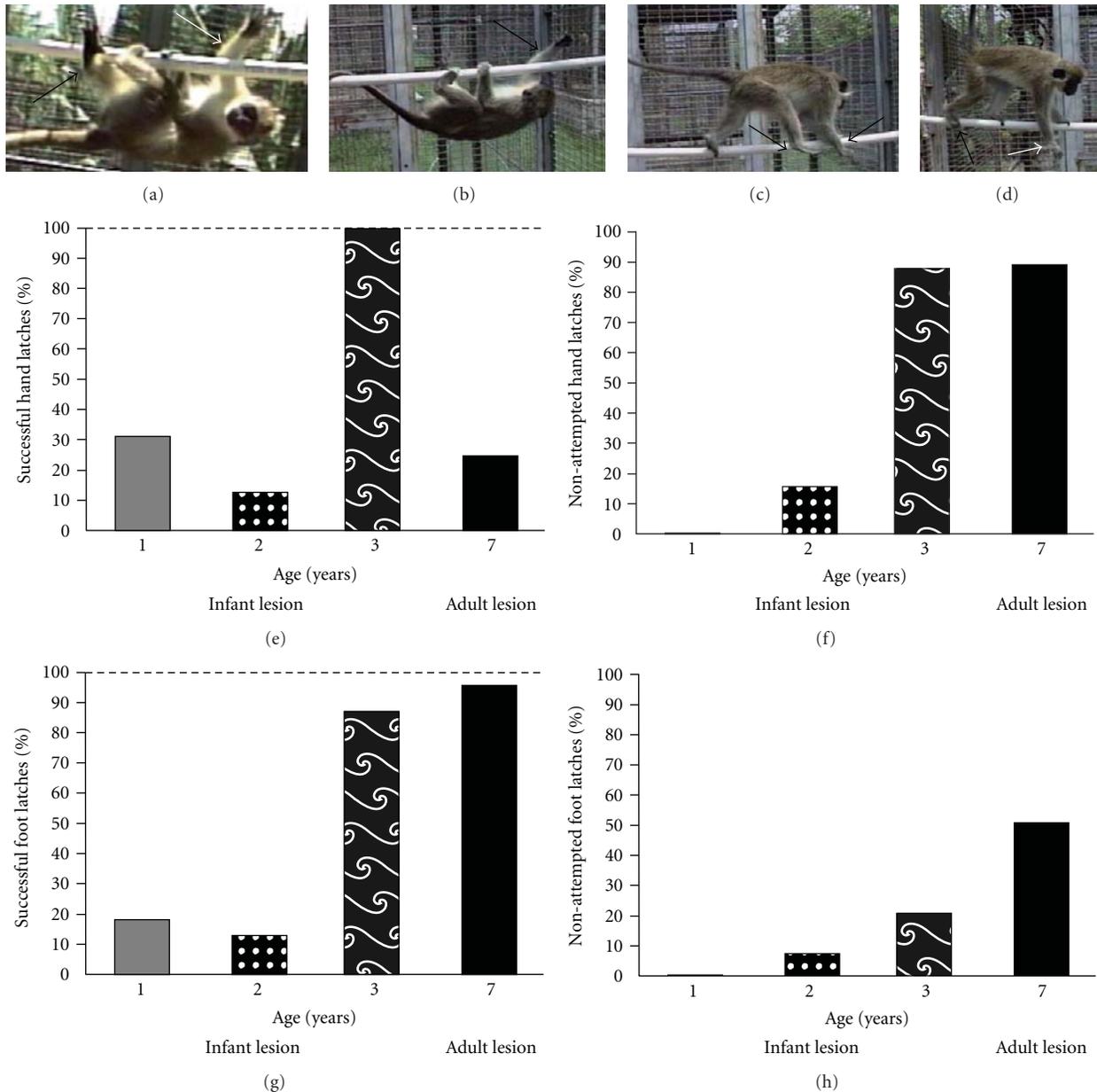


FIGURE 4: At a young age, the subjects made a significant number of unsuccessful attempts to grab the horizontal bar with their hands and would transverse the bar upside down (a and e). The subjects would typically overreach the bar with their arms and glide their arms across the bar until their hands were able to latch on (white arrows in a and b). The lower limb also had difficulty latching onto the bar during the first two postoperative years after the surgery (black arrow in panels a and g). At two years of age, the monkeys began not to attempt to use the contralateral upper limb to transverse the bar (black arrows in panel c). Typically the subject would successfully use the affected hind limb (black arrow in panels d and h) and would not attempt to use the front limb (white arrow in panels d and f) to transverse the horizontal bar. By 2 years after surgery the subjects were able to walk upright across the bar. Dashed line (e and g) represents the expected value for a normal monkey. % successful was determined as successful latches (hand or foot)/total attempts to latch onto the horizontal bar. The ipsilateral side of the body acts as an internal control for normal motor movements and did not show any paretic movements. Years 1, 2, and 3 refer to the age of the infant-lesioned subjects at testing which also corresponds to the years after lesion. For the adult-lesioned subjects, year 4 refers to the number of years after initial surgery. Adapted from [14].

had already begun much before surgery but was masked by the dysfunctional hemisphere. The release from the “negative” influence exerted by the diseased hemisphere would give the appearance of a rapid reorganization of the remaining hemisphere [10]. Data from our laboratory

as well as those from human studies suggest a critical role for subcortical and brainstem structures as well as corticospinal tracts in the neuroanatomical reorganization which result in the remarkable behavioral recovery following hemispherectomy [12, 40–43].

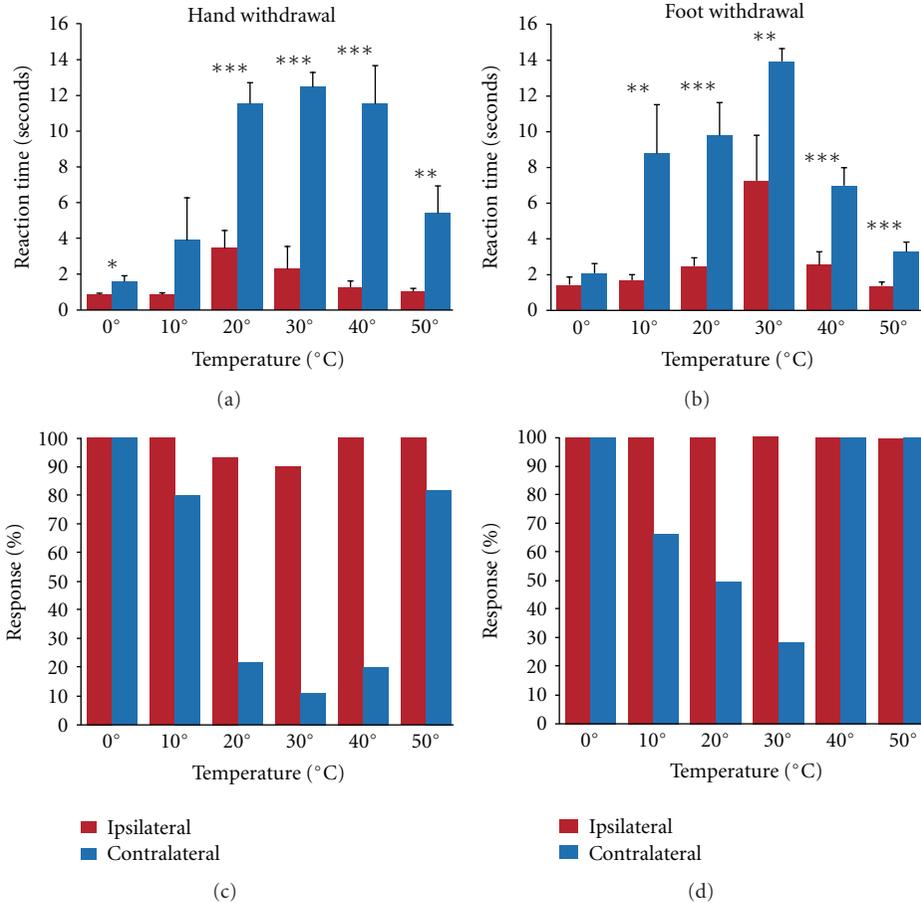


FIGURE 5: Thermal sensitivity: thermal sensitivity was tested in the infant-lesioned subjects at 3 years of age. The ipsilateral side of the body acts as a control for normal response to thermal stimuli and, as stated earlier, did not show any paretic movements. Withdrawal times were significantly longer for the contralateral (paretic side) than for the ipsilateral (nonparetic) limb (a and b). The contralateral lower limb tended to have higher response rates than upper limb to thermal sensitivity (c and d). Withdrawal response rates did not differ between upper and lower ipsilateral limbs. $*P < 0.05$, $**P < 0.01$, $***P < 0.001$.

3.1. Summary and Clinical Parallels

3.1.1. Vision. Clinical studies involving individuals that underwent hemispherectomy conclude that there is marked improvement of life post-surgery, mainly due to the cessation of seizure activity [5, 6, 44, 45]. Recovery largely depends on the system in question. We report here that infant-lesioned monkeys had residual vision up to 45° in the blind hemifield compared to adult-lesioned subjects, where a behavioral response could not be elicited in the blind hemifield. We have previously reported that these subjects demonstrate a pervasive ipsilateral turning preference that may be indicative of a visuospatial impairment related to hemianopia. A preference for processing visual information from the ipsilateral hemifield (i.e., the unaffected field of vision) may be responsible for ipsiversive turning [14]. These residual visual capabilities parallel clinical findings for individuals who underwent hemispherectomy for treatment of intractable epilepsy. Indeed, in human hemispherectomy patients, nonreflexive visual responses can be elicited in the blind hemifield [46]. Responses to visual stimuli presented

in the hemianopic side have been classified according to the patient's implicit (Type I blindsight) or explicit (Type II blindsight) acknowledgement of the presence of the stimulus [7]. The ability of infant-lesioned subjects in the current study to actively orientate towards food rewards in the blind hemifield is reminiscent of Type II blindsight. The lack of visual orientation in the blind hemifield of the adult-lesioned subjects suggests that like recovery in sensorimotor abilities, the younger cortical lesioned subjects tend to recover function more completely than their adult-lesioned counterparts [14]. Results from lesions restricted to the striate cortex of cats and monkeys [47] support a relationship between the degree of visual recovery and the age at which the lesion was performed [48, 49].

In clinics, it is generally accepted that early-lesioned patients have a larger chance of residual vision [50]. A recent clinical study suggests that children who have unilateral or bilateral loss of their occipital lobe(s) are capable of retaining normal vision in both visual fields when tested with a forced-choice preferential-looking perimetric method. A child who underwent a complete hemispherectomy at the age of 59

months could consciously detect light throughout the visual field. This is in sharp contrast with a visual perimetry of only 30° in a patient who underwent hemispherectomy at 13 years of age, suggesting that visual recovery is dependent upon age of lesion. In both patients, the luminance of the visual target had to be at least 45 cd/m^2 to allow detection in the affected visual hemifield, compared to 5 cd/m^2 in the unaffected hemifield [36]. Moreover, a patient born with developmental anomaly of both occipital lobes demonstrated a large reduction of the bilateral scotomas leading to an expansion of the total visual field [19]. Residual vision varies among patients and may not be entirely dependent on the age of surgery since residual vision has also been reported in patients who underwent hemispherectomy during late adolescence and early adulthood [7, 51, 52].

In patients and monkeys with massive unilateral lesions of the primary visual cortex, residual vision in the blind hemifield (Type I and Type II blindsight) has been ascribed to ipsilateral extrastriate cortical areas that receive inputs via the colliculopulvinar pathway [53, 54]. In the case of anatomical hemispherectomy where all visual cortical regions of one hemisphere have been removed, residual vision is dependent upon the contribution of the remaining hemisphere as demonstrated by brain-imaging methods. Thus, it was shown that visual stimulation of the blind hemifield in hemispherectomy patients activates visual cortical areas in the remaining hemisphere [55, 56]. A more recent, DTI study by Leh and colleagues [52] highlighted the pathway by which the visual information presented to the blind field reaches the contralateral visual cortex. Indeed, these authors traced a retinofugal projection to the SC ipsilateral to the removed hemisphere that reaches the contralateral SC, then the pulvinar and the extrastriate cortices [7, 52]. This indicates that the SC ipsilateral to the removed hemisphere survives the hemispherectomy and can be activated by visual stimuli presented in the hemianopic field.

Anatomical evidence, from our laboratory on the same hemispherectomized monkeys used in our behavioral experiments, suggests that retinal and subcortical visual structures survive the lesion but to varying degrees [12]. Although there is a massive transneuronal degeneration of retinal ganglion cells in the fovea, the peripheral retina remains unaffected [57]. The main thalamic target of the retina, the dorsolateral geniculate nucleus (dLGN), ipsilateral to the removed cortex undergoes a major loss of neurons and an intense gliosis [58]. Notwithstanding a large volume reduction, the dLGN still receives projections from each retina ending in the appropriate layers [15]. The paucity of surviving neurons within the magno- and parvocellular layers does not make the dLGN a likely candidate for sustaining residual vision. On the other hand, the SC retains functional capabilities as revealed by cytochrome oxidase activity and receives normal retinal inputs. Unlike the dLGN, the ipsilateral colliculus undergoes only moderate neuronal reduction following hemispherectomy and remains mostly intact [59]. Moreover, the ipsilateral substantia nigra also remains intact with no obvious volume or neuronal loss [60]. The substantia nigra plays an important role in saccadic eye movements with the lateral part committing the majority of its projections to

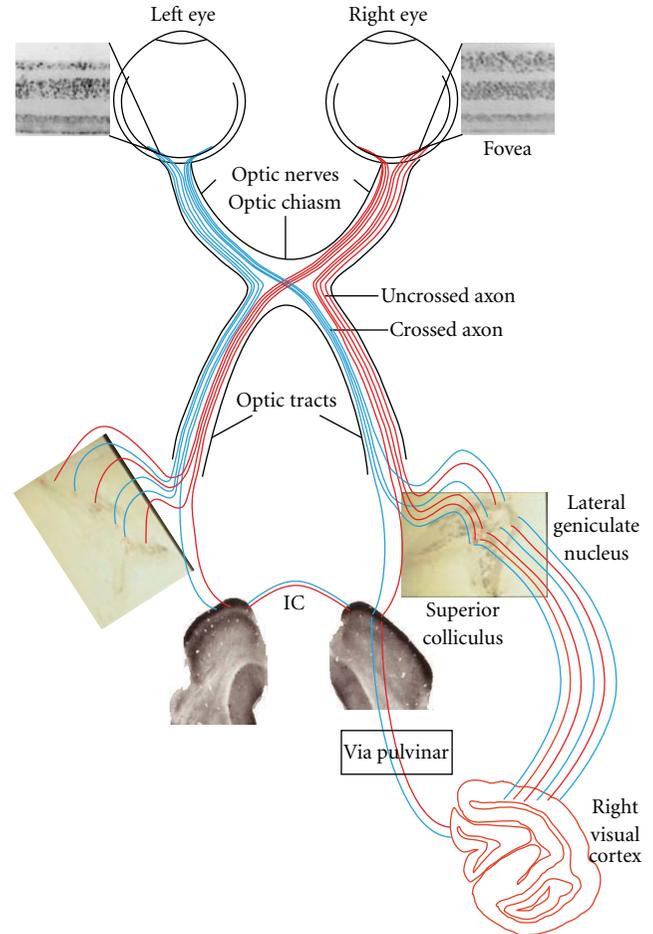


FIGURE 6: Suggested anatomical pathway for visual field recovery following early hemispherectomy (based on a series of anatomical studies performed on the same monkeys). The deafferented left temporal hemiretina (T) still contains ganglion cells [57] that send their axons to the remaining left dLGN as a dead end [12]. Retinofugal projections to the left superior colliculus (SC) are still robust and maintain a normal metabolic rate as measured by cytochrome oxidase immunocytochemistry [59]. The information reaching the left SC is transferred to the right SC via the intertectal commissure (IC), the right Pulvinar (P), and the occipital cortex of the remaining right hemisphere. The deafferented right nasal hemiretina (N) sends crossed projection through the optic chiasm (OC) to the appropriate layers of the left dLGN and to the left SC [15, 58, 59, 62]. The left Pulvinar is severely atrophied and receives no retinal projections. Projections from the afferented portions of the retinae (left nasal and right temporal) reach their subcortical targets in a normal fashion en route to the occipital cortex of the right hemisphere.

the nigrotectal pathway [61]. As suggested by tractography studies in human hemispherectomy patients, it seems, therefore, that the collicular system and the lateral substantia nigra play an important role in residual visual capabilities. Figure 6 summarizes the neural substrates implicated in residual vision in the blind hemifield of hemispherectomized monkeys.

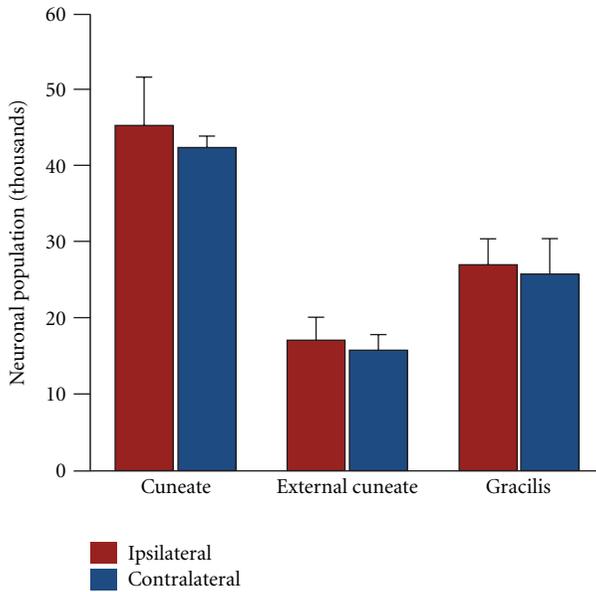


FIGURE 7: Design-based stereology neuronal counts in the dorsal column nuclei. There were no neuronal population differences between ipsi- and contralateral subdivisions of the dorsal column nuclei.

3.1.2. Motor and Somatosensory Functions. Our results on the behavioral recovery of sensory-motor behaviors in the infant hemispherectomized monkey parallel to those reported in clinical cases [5, 30, 63] as well as those reported in neonatal feline models of hemispherectomy [64–66]. The infant-lesioned monkeys tend to regain function in the lower limb within a year after surgery, but the upper limb appears to remain hemiparetic, mirroring clinical data where subjects may regain muscle strength in the leg but continue to show weakness in the contralateral arm and hand with an intact tactile detection [5, 30, 35]. The ineptitude of our subjects to effectively transverse the horizontal bar reflects an inability to integrate complex visuospatial, somatosensory, and motor information. A diminished tactile sensation became especially apparent with the overreaching and gliding of the arm and leg until the subject was able to latch onto the bar.

All the preserved functions, motor as well as somatosensory, following hemispherectomy in both humans and nonhuman primates have been attributed either to an extensive anatomical reorganization or to the use of compensatory mechanisms involving either the remaining cortex or subcortical residual structures. There is evidence that ipsilateral projections may play a role in the retention of function following hemispherectomy. Patients are able to perceive tactile stimulation applied to their paretic leg, and fMRI studies have shown that this type of stimulation leads to activation of the intact primary and secondary somatosensory cortices, suggesting ipsilateral pathways from the periphery [34, 38, 39, 67]. In one particular patient, thermal sensitivity was impaired on the paretic side of the body, but the patient was able to discriminate temperature changes on the skin. This patient experienced a pricking-burning

sensation of the skin (allodynia) that was exacerbated by cold. fMRI data indicated that the allodynia experienced on the paretic limbs was processed in the remaining cortex in areas normally associated with pain processing [67]. The remaining cortex has also been shown to undergo significant reshaping in the motor and sensorimotor cortical representations [37, 68–71]. There is evidence from fMRI studies that physical training with the paretic lower limb results in cortical activation of the remaining primary sensorimotor, supplementary motor, cingulate, and secondary somatosensory cortices, suggesting an experiential or active-dependent recovery [72].

The ability of the remaining hemisphere to assume functional control over the ipsilateral body may be due to a reorganization of the brainstem tracts such as the corticospinal tract and medial lemniscus [41–43, 72]. In a child with intractable epilepsy in the left hemisphere, a presurgical fMRI study showed activation of the right, but not of the left primary motor cortex following tactile and motor stimulation of the right hand. These results suggest that reorganization occurred prior to surgery and that the corticospinal fibers originating from the nonaffected hemisphere mediated the reorganization [41]. In normally developing children, ipsilateral corticospinal connections remain intact until around 10 years of age [73] providing an anatomical substrate for reorganization following early-life dysfunction. Diffusion tensor imaging (DTI) data further suggest that the medial lemniscus may play a vital role in sensory recovery as it retains symmetry following early-life hemispherectomy [43]. Using design-based stereology, our group has reported relatively preserved contralateral dorsal column nuclei (gracilis, cuneatus, and external cuneatus) following hemispherectomy in infant primates (Figure 7) that could, in part, mediate the behavioral recovery [40].

The differential effect of hemispherectomy on the upper-versus lower limbs may also be mediated by networks of interneurons within the spinal cord [5, 30, 42, 74]. In human and nonhuman primates, lower-limb locomotion is thought to be under the control of neuronal circuits of the central pattern generator, within the spinal cord, whereas the upper limbs are under the control of corticospinal pathways [5, 74]. It has also been suggested that early cortical lesions lead to a reinforcement of the ipsilateral corticospinal tracts, whereas for cortical lesions sustained late in life recovery may be mediated by the cortico reticulospinal pathway [42].

3.2. Conclusion. The data presented here suggest that significant functional recovery occurs after the removal of one hemisphere in monkeys with no preexisting neurological dysfunctions. The nonhuman primate model presented here offers a unique opportunity for studying the behavioral and functional neuroanatomical reorganization that underlies developmental plasticity. All the preserved visual, motor, and sensory functions following hemispherectomy in both humans and nonhuman primates have been attributed to an extensive anatomical reorganization or to the use of compensatory mechanisms involving either the remaining contralateral cortex or subcortical residual structures [12]. The anatomical state of the visual system of infant-lesioned

monkeys adds support to the implication of the collicular system in mediating the sparing of vision in the contralateral hemifield observed in hemispherectomized humans [7, 50]. Anatomical and imaging studies further suggest that recovery of the motor and somatosensory (both tactile and thermal) functions may be subserved by existing, nondegenerating, ipsilateral projections of the medial lemniscus, corticospinal tract, and dorsal column nuclei as well as preserved superior colliculi and substantia nigra [60]. The extent and manner to which subcortical areas, the brainstem, and the spinal cord participate in functional reorganization following early-life hemispherectomy remains unresolved. The nonhuman primate model presented here has provided a significant contribution to our understanding of the behavioral and functional neuroanatomical reorganization that underlies developmental plasticity.

Acknowledgments

The authors would like to thank the staff at the Behavioral Sciences Foundation of St. Kitts (West Indies) for their constant support and excellent technical assistance. This research, over the years, was supported by the National Science and Engineering Research Council of Canada (NSERC) to Maurice Ptito.

References

- [1] P. J. E. Wilson, "Cerebral hemispherectomy for infantile hemiplegia: a REPORT of 50 cases," *Brain*, vol. 93, no. 1, pp. 147–180, 1970.
- [2] L. A. French, D. R. Johnson, I. A. Brown, and F. B. van Bergen, "Cerebral hemispherectomy for control of intractable convulsive seizures," *Journal of Neurosurgery*, vol. 12, no. 2, pp. 154–164, 1955.
- [3] R. W. Krynauw, "Infantile hemiplegia treated by removal of one cerebral hemisphere," *South African Medical Journal*, vol. 24, no. 27, pp. 539–546, 1950.
- [4] H. H. White, "Cerebral hemispherectomy in the treatment of infantile hemiplegia; review of the literature and report of two cases," *Confinia Neurologica*, vol. 21, pp. 1–50, 1961.
- [5] R. van Empelen, A. Jennekens-Schinkel, E. Buskens, P. J. M. Helders, and O. van Nieuwenhuizen, "Functional consequences of hemispherectomy," *Brain*, vol. 127, no. 9, pp. 2071–2079, 2004.
- [6] A. M. Devlin, J. H. Cross, W. Harkness et al., "Clinical outcomes of hemispherectomy for epilepsy in childhood and adolescence," *Brain*, vol. 126, no. 3, pp. 556–566, 2003.
- [7] A. Ptito and S. E. Leh, "Neural substrates of blindsight after hemispherectomy," *Neuroscientist*, vol. 13, no. 5, pp. 506–518, 2007.
- [8] L. A. French and D. R. Johnson, "Observations on the motor system following cerebral hemispherectomy," *Neurology*, vol. 5, no. 1, pp. 11–14, 1955.
- [9] J. Gagnier, *La sensibilité thermique dans les cas d'hémisphérectomie humaine*, Maitrise Inedit, University of Montreal, Montreal, Canada, 1992.
- [10] F. Chiricozzi, D. Chieffo, D. Battaglia et al., "Developmental plasticity after right hemispherectomy in an epileptic adolescent with early brain injury," *Child's Nervous System*, vol. 21, no. 11, pp. 960–969, 2005.
- [11] R. M. Govindan, H. T. Chugani, A. F. Luat, and S. Sood, "Presurgical prediction of motor functional loss using tractography," *Pediatric Neurology*, vol. 43, no. 1, pp. 70–72, 2010.
- [12] D. Boire, H. Théoret, and M. Ptito, "Visual pathways following cerebral hemispherectomy," *Progress in Brain Research*, vol. 134, pp. 379–397, 2001.
- [13] M. Ptito, M. Herbin, D. Boire, and A. Ptito, "Neural bases of residual vision in hemispherectomized monkeys," *Progress in Brain Research*, vol. 112, pp. 385–404, 1996.
- [14] M. W. Burke, S. Zangenehpour, and M. Ptito, "Partial recovery of hemiparesis following hemispherectomy in infant monkeys," *Neuroscience Letters*, vol. 469, no. 2, pp. 243–247, 2010.
- [15] D. Boire, H. Theoret, M. Herbin, C. Casanova, and M. Ptito, "Retinogeniculate projections following early cerebral hemispherectomy in the vervet monkey," *Experimental Brain Research*, vol. 135, no. 3, pp. 373–381, 2000.
- [16] M. Ptito and S. Desgent, Eds., *Sensory Input-Based Adaptation and Brain Architecture*, Cambridge University Press, Cambridge, UK, 2006.
- [17] B. R. Payne and S. G. Lomber, "Plasticity of the visual cortex after injury: what's different about the young brain?" *Neuroscientist*, vol. 8, no. 2, pp. 174–185, 2002.
- [18] D. O. Frost, D. Boire, G. Gingras, and M. Ptito, "Surgically created neural pathways mediate visual pattern discrimination," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 20, pp. 11068–11073, 2000.
- [19] M. Ptito, M. Dalby, and A. Gjedde, "Visual field recovery in a patient with bilateral occipital lobe damage," *Acta Neurologica Scandinavica*, vol. 99, no. 4, pp. 252–254, 1999.
- [20] C. K. Kong, L. Y. Wong, and M. K. Yuen, "Visual field plasticity in a female with right occipital cortical dysplasia," *Pediatric Neurology*, vol. 23, no. 3, pp. 256–260, 2000.
- [21] S. O. Dumoulin, J. D. Jirsch, and A. Bernasconi, "Functional organization of human visual cortex in occipital polymicrogyria," *Human Brain Mapping*, vol. 28, no. 12, pp. 1302–1312, 2007.
- [22] M. Ptito, P. Johannsen, J. Faubert, and A. Gjedde, "Activation of human extrageniculostriate pathways after damage to area V1," *NeuroImage*, vol. 9, no. 1, pp. 97–107, 1999.
- [23] E. Kasten, D. A. Poppel, and B. A. Sabel, "Computer-based training of stimulus detection improves color and simple pattern recognition in the defective field of hemianopic subjects," *Journal of Cognitive Neuroscience*, vol. 12, no. 6, pp. 1001–1012, 2000.
- [24] B. A. Sabel and E. Kasten, "Restoration of vision by training of residual functions," *Current Opinion in Ophthalmology*, vol. 11, no. 6, pp. 430–436, 2000.
- [25] L. Bouwmeester, J. Heutink, and C. Lucas, "The effect of visual training for patients with visual field defects due to brain damage: a systematic review," *Journal of Neurology, Neurosurgery and Psychiatry*, vol. 78, no. 6, pp. 555–564, 2007.
- [26] F. Schmielau and E. K. Wong Jr., "Recovery of visual fields in brain-lesioned patients by reaction perimetry treatment," *Journal of NeuroEngineering and Rehabilitation*, vol. 4, article no. 31, 2007.
- [27] S. Chokron, C. Perez, M. Obadia, I. Gaudry, L. Laloum, and O. Gout, "From blindsight to sight: cognitive rehabilitation of visual field defects," *Restorative Neurology and Neuroscience*, vol. 26, no. 4–5, pp. 305–320, 2008.
- [28] S. M. Sherman, "Visual fields of cats with cortical and tectal lesions," *Science*, vol. 185, no. 4148, pp. 355–357, 1974.
- [29] S. G. Lomber, B. R. Payne, C. C. Hilgetag, and J. R. Rushmore, "Restoration of visual orienting into a cortically

- blind hemifield by reversible deactivation of posterior parietal cortex or the superior colliculus," *Experimental Brain Research*, vol. 142, no. 4, pp. 463–474, 2002.
- [30] J. A. Vilensky, S. Gilman, E. A. Dunn, and W. J. Wilson, "Utilization of the Denny-Brown collection: differential recovery of forelimb and hind limb stepping after extensive unilateral cerebral lesions," *Behavioural Brain Research*, vol. 82, no. 2, pp. 223–233, 1997.
- [31] M. A. Kennard, "Reactions of monkeys of various ages to partial and complete decortication," *Journal of Neuropathology and Experimental Neurology*, vol. 3, no. 21, pp. 289–310, 1944.
- [32] N. Honda, T. Matuoka, Y. Sawada et al., "Reorganization of sensorimotor function after functional hemispherectomy studied using near-infrared spectroscopy," *Pediatric Neurosurgery*, vol. 46, no. 4, pp. 313–317, 2011.
- [33] R. G. Bittar, J. V. Rosenfeld, G. L. Klug, I. J. Hopkins, and A. Simon Harvey, "Resective surgery in infants and young children with intractable epilepsy," *Journal of Clinical Neuroscience*, vol. 9, no. 2, pp. 142–146, 2002.
- [34] R. G. Bittar, A. Olivier, A. F. Sadikot, F. Andermann, and D. C. Reutens, "Cortical motor and somatosensory representation: effect of cerebral lesions," *Journal of Neurosurgery*, vol. 92, no. 2, pp. 242–248, 2000.
- [35] H. Backlund, C. Morin, A. Ptito, M. C. Bushnell, and H. Olausson, "Tactile functions after cerebral hemispherectomy," *Neuropsychologia*, vol. 43, no. 3, pp. 332–339, 2005.
- [36] R. Werth, "Visual functions without the occipital lobe or after cerebral hemispherectomy in infancy," *European Journal of Neuroscience*, vol. 24, no. 10, pp. 2932–2944, 2006.
- [37] V. Holloway, D. G. Gadian, F. Vargha-Khadem, D. A. Porter, S. G. Boyd, and A. Connelly, "The reorganization of sensorimotor function in children after hemispherectomy. A functional MRI and somatosensory evoked potential study," *Brain*, vol. 123, no. 12, pp. 2432–2444, 2000.
- [38] H. Olausson, B. Ha, G. H. Duncan et al., "Cortical activation by tactile and painful stimuli in hemispherectomized patients," *Brain*, vol. 124, no. 5, pp. 916–927, 2001.
- [39] R. G. Bittar, A. Ptito, and D. C. Reutens, "Somatosensory representation in patients who have undergone hemispherectomy: a functional magnetic resonance imaging study," *Journal of Neurosurgery*, vol. 92, no. 1, pp. 45–51, 2000.
- [40] P. Lauzon, D. Boire, and M. Ptito, *Stereological Evaluation of Dorsal Column Nuclei in Normal and Early Hemispherectomized Monkeys*, Society for Neuroscience, Miami, Fla, USA, 1999.
- [41] G. J. M. Rutten, N. F. Ramsey, P. C. van Rijen, H. Franssen, and C. W. M. van Veelen, "Interhemispheric reorganization of motor hand function to the primary motor cortex predicted with functional magnetic resonance imaging and transcranial magnetic stimulation," *Journal of Child Neurology*, vol. 17, no. 4, pp. 292–297, 2002.
- [42] R. Benecke, B. U. Meyer, and H. J. Freund, "Reorganisation of descending motor pathways in patients after hemispherectomy and severe hemispheric lesions demonstrated by magnetic brain stimulation," *Experimental Brain Research*, vol. 83, no. 2, pp. 419–426, 1991.
- [43] J. T. Choi, E. P. G. Vining, S. Mori, and A. J. Bastian, "Sensorimotor function and sensorimotor tracts after hemispherectomy," *Neuropsychologia*, vol. 48, no. 5, pp. 1192–1199, 2010.
- [44] T. Maehara, H. Shimizu, K. Kawai et al., "Postoperative development of children after hemispherotomy," *Brain and Development*, vol. 24, no. 3, pp. 155–160, 2002.
- [45] M. Ptito, A. Fortin, and A. Ptito, "Living with half a brain: the effects of hemispherectomy in humans," *Revue de Neuropsychologie*, vol. 12, no. 1, pp. 185–210, 2002.
- [46] P. Stoerig, "Cueless blindsight," *Frontiers in Human Neuroscience*, vol. 3, article 74, 2010.
- [47] T. Moore, H. R. Rodman, A. B. Repp, C. G. Gross, and R. S. Mezzrich, "Greater residual vision in monkeys after striate cortex damage in infancy," *Journal of Neurophysiology*, vol. 76, no. 6, pp. 3928–3933, 1996.
- [48] J. R. Villablanca and D. A. Hovda, "Developmental neuroplasticity in a model of cerebral hemispherectomy and stroke," *Neuroscience*, vol. 95, no. 3, pp. 625–637, 1999.
- [49] B. R. Payne, S. G. Lomber, and C. D. Gelston, "Graded sparing of visually-guided orienting following primary visual cortex ablations within the first postnatal month," *Behavioural Brain Research*, vol. 117, no. 1–2, pp. 1–11, 2000.
- [50] A. Ptito, A. Fortin, and M. Ptito, "'Seeing' in the blind hemifield following hemispherectomy," *Progress in Brain Research*, vol. 134, pp. 367–378, 2001.
- [51] C. M. Wessinger, R. Fendrich, A. Ptito, J. G. Villemure, and M. S. Gazzaniga, "Residual vision with awareness in the field contralateral to a partial or complete functional hemispherectomy," *Neuropsychologia*, vol. 34, no. 11, pp. 1129–1137, 1996.
- [52] S. E. Leh, H. Johansen-Berg, and A. Ptito, "Unconscious vision: new insights into the neuronal correlate of blindsight using diffusion tractography," *Brain*, vol. 129, no. 7, pp. 1822–1832, 2006.
- [53] A. Cowey, "Visual system: how does blindsight arise?" *Current Biology*, vol. 20, no. 17, pp. R702–R704, 2010.
- [54] A. Cowey, "The blindsight saga," *Experimental Brain Research*, vol. 200, no. 1, pp. 3–24, 2010.
- [55] R. G. Bittar, A. Olivier, A. F. Sadikot et al., "Localization of somatosensory function by using positron emission tomography scanning: a comparison with intraoperative cortical stimulation," *Journal of Neurosurgery*, vol. 90, no. 3, pp. 478–483, 1999.
- [56] E. Marx, T. Stephan, S. Bense, T. A. Yousry, M. Dieterich, and T. Brandt, "Motion perception in the ipsilateral visual field of a hemispherectomized patient," *Journal of Neurology*, vol. 249, no. 9, pp. 1303–1306, 2002.
- [57] M. Herbin, D. Boire, H. Théoret, and M. Ptito, "Transneuronal degeneration of retinal ganglion cells in early hemispherectomized monkeys," *NeuroReport*, vol. 10, no. 7, pp. 1447–1452, 1999.
- [58] D. Boire, H. Théoret, and M. Ptito, "Stereological evaluation of neurons and glia in the monkey dorsal lateral geniculate nucleus following an early cerebral hemispherectomy," *Experimental Brain Research*, vol. 142, no. 2, pp. 208–220, 2002.
- [59] H. Théoret, D. Boire, M. Herbin, and M. Ptito, "Anatomical sparing in the superior colliculus of hemispherectomized monkeys," *Brain Research*, vol. 894, no. 2, pp. 274–280, 2001.
- [60] H. Théoret, D. Boire, M. Herbin, and M. Ptito, "Stereological evaluation of substantia nigra cell number in normal and hemispherectomized monkeys," *Brain Research*, vol. 835, no. 2, pp. 354–359, 1999.
- [61] R. M. Beckstead and A. Frankfurter, "The distribution and some morphological features of substantia nigra neurons that project to the thalamus, superior colliculus and pedunculo-pontine nucleus in the monkey," *Neuroscience*, vol. 7, no. 10, pp. 2377–2388, 1982.
- [62] H. Théoret, D. Boire, and M. Ptito, "Retinal projections to the pregeniculate nucleus in the hemispherectomized monkey," *Brain Research Bulletin*, vol. 53, no. 2, pp. 239–243, 2000.

- [63] F. Liégeois, A. T. Morgan, L. H. Stewart, J. Helen Cross, A. P. Vogel, and F. Vargha-Khadem, "Speech and oral motor profile after childhood hemispherectomy," *Brain and Language*, vol. 114, no. 2, pp. 126–134, 2010.
- [64] J. W. Burgess and J. R. Villablanca, "Recovery of function after neonatal or adult hemispherectomy in cats. II. Limb bias and development, paw usage, locomotion and rehabilitative effects of exercise," *Behavioural Brain Research*, vol. 20, no. 1, pp. 1–17, 1986.
- [65] J. W. Burgess, J. R. Villablanca, and M. S. Levine, "Recovery of functions after neonatal or adult hemispherectomy in cats. III. Complex functions: open field exploration, social interactions, maze and holeboard performances," *Behavioural Brain Research*, vol. 20, no. 2, pp. 217–230, 1986.
- [66] J. R. Villablanca, J. W. Burgess, and C. E. Olmstead, "Recovery of function after neonatal or adult hemispherectomy in cats: I. Time course, movement, posture and sensorimotor tests," *Behavioural Brain Research*, vol. 19, no. 3, pp. 205–226, 1986.
- [67] H. Olausson, S. Marchand, R. G. Bittar, J. Bernier, A. Ptito, and M. C. Bushnell, "Central pain in a hemispherectomized patient," *European Journal of Pain*, vol. 5, no. 2, pp. 209–217, 2001.
- [68] C. J. Graveline, D. J. Mikulis, A. P. Crawley, and P. A. Hwang, "Regionalized sensorimotor plasticity after hemispherectomy fMRI evaluation," *Pediatric Neurology*, vol. 19, no. 5, pp. 337–342, 1998.
- [69] J. Rath, R. Schmidhammer, T. Steinkellner, N. Klinger, A. Geissler, and R. Beisteiner, "Evaluation of functional cortex for the diseased hand in a patient after hemispherectomy," *Archives of Neurology*, vol. 65, no. 12, pp. 1664–1665, 2008.
- [70] F. Pilato, M. Dileone, F. Capone et al., "Unaffected motor cortex remodeling after hemispherectomy in an epileptic cerebral palsy patient: a TMS and fMRI study," *Epilepsy Research*, vol. 85, no. 2-3, pp. 243–251, 2009.
- [71] G. Leonhardt, U. Bingel, G. Spiekermann, M. Kurthen, S. Müller, and A. Hufnagel, "Cortical activation in patients with functional hemispherectomy," *Journal of Neurology*, vol. 248, no. 10, pp. 881–888, 2001.
- [72] S. de Bode, G. W. Mathern, S. Bookheimer, and B. Dobkin, "Locomotor training remodels fMRI sensorimotor cortical activations in children after cerebral hemispherectomy," *Neurorehabilitation and Neural Repair*, vol. 21, no. 6, pp. 497–508, 2007.
- [73] K. Müller, F. Kass-Iliyya, and M. Reitz, "Ontogeny of ipsilateral corticospinal projections: a developmental study with transcranial magnetic stimulation," *Annals of Neurology*, vol. 42, no. 5, pp. 705–711, 1997.
- [74] V. Dietz, "Spinal cord pattern generators for locomotion," *Clinical Neurophysiology*, vol. 114, no. 8, pp. 1379–1389, 2003.

Review Article

GABA through the Ages: Regulation of Cortical Function and Plasticity by Inhibitory Interneurons

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Received 13 January 2012; Revised 30 March 2012; Accepted 30 March 2012

Academic Editor: Maurice Ptito

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Inhibitory interneurons comprise only about 20% of cortical neurons and thus constitute a clear minority compared to the vast number of excitatory projection neurons. They are, however, an influential minority with important roles in cortical maturation, function, and plasticity. In this paper, we will highlight the functional importance of cortical inhibition throughout brain development, starting with the embryonal formation of the cortex, proceeding by the regulation of sensory cortical plasticity in adulthood, and finishing with the GABA involvement in sensory information processing in old age.

1. Introduction

The functioning of the cerebral cortex depends critically on the precise balance between excitatory and inhibitory neurotransmitter systems. Excitation is mediated via glutamate by pyramidal neurons, the projection neurons of the cortex, and by a special class of local neurons in cortical layer IV, the spiny stellate cells. Inhibition is mediated via γ -aminobutyric acid (GABA) by cortical interneurons, which regulate the degree of glutamatergic excitation, filtering the input and regulate the output of projection neurons. GABAergic interneurons, the “nonpyramidal cells” of the cerebral cortex, take many different forms of dendritic and axonal arborization, which have been used for their morphological classification ever since their first description by Ramon y Cajal [1–5]. Moreover, interneurons also differ in their firing patterns, the neuropeptides they express, their calcium-binding protein content, and other molecular markers such as ion channels, receptors, and transporters. Based on the combination of structural, functional, and biochemical criteria, interneurons have been subdivided into many different subclasses and it is still a matter of hot debate among the experts of how many interneuron subtypes exist in the cortices of different species [6–8].

At the circuit level, interneurons control the flow of information and synchronization in the cerebral cortex.

There are about five times more glutamatergic neurons than GABAergic neurons in the neocortex; this ratio is consistently observed across many mammalian species. This then suggests that the numerical balance of excitatory and inhibitory neurons may be important for normal brain function and behavior. Even though GABAergic interneurons comprise only a small fraction of the cells in the neocortex, disturbances in their development, and hence the delicate balance between excitation and inhibition, can lead to neurological or neuropsychiatric diseases such as epilepsy, autism, and schizophrenia (reviewed in [9–13]). These disorders often emerge during childhood and adolescence. However, as we will describe later in this paper, alterations in GABAergic interneuron can also occur in the adult and aging brain, with important repercussions for cortical function and plasticity.

2. Setting the Balance: Interneuron Migration into the Cortical Plate

Cortical projection neurons, the excitatory pyramidal cells, arise in the ventricular zone (VZ) of the dorsal telencephalon and then migrate radially to form the laminated neocortex [14]. In contrast, GABAergic interneurons originate from the VZ of the ventral telencephalon from three regions: different

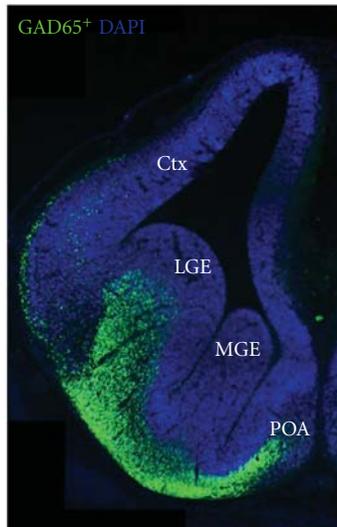


FIGURE 1: GABAergic interneurons are born in the basal telencephalon. Coronal cryosection of an E14 GAD65 EGFP mouse embryo. The MGE and the POA give rise to tangentially migrating cortical interneurons (green). GABAergic LGE-derived neurons migrate to the olfactory bulb, the striatum, and the lateral cortex. Ctx: cortex, MGE: medial ganglionic eminence, LGE: lateral ganglionic eminence, POA: preoptic area.

interneuron subtypes are generated in the medial ganglionic eminence (MGE), the caudal ganglionic eminence (CGE), and the preoptic area (POA) (see Figure 1). For example, parvalbumine positive and somatostatin-positive interneurons arise mainly from the MGE, while most calretinin-positive interneurons are born in the CGE and the POA gives birth to vasoactive intestinal peptide and neuropeptide Y-positive interneurons. Interneurons originating from these regions then migrate tangentially in separate streams over long distances towards their cortical destinations [15–22].

The cellular and molecular mechanisms that regulate and guide interneuron migration out of the ganglionic eminences and POA into the neocortex are beginning to be described. Different groups of signaling molecules, including semaphorins and slits, act as repulsive cues for migrating interneurons [23, 24]. On the other hand, two isoforms of neuregulin act as short- and long-term attractants that demarcate the migratory route of cortical interneurons [25]. Another group of signaling molecules that is expressed widely in the basal telencephalon during interneuron migration is the ephrins and their receptors, the Eph receptor tyrosine kinases. As will be described later, recent findings from our lab provided direct evidence for distinct roles of Eph/ephrin interactions in the guidance of cortical interneuron migration.

The mammalian Eph/ephrin system consists of a family of receptor tyrosine kinases subdivided into 9 EphAs and 5 EphBs. A-type receptors bind to all A-type ephrins (ephrinA1–5), which are tethered to the cell membrane by a GPI anchor. B-type receptors bind to all B-type ephrins (ephrinB1–3), which have a transmembrane domain that is followed by a short cytoplasmic region. An exception is EphA4, which can bind to both A-type and B-type ligands

[26–28]. A distinctive feature of this signaling system is that an Eph receptor can also act as a ligand in the same manner that an ephrin ligand can act as a receptor. Ephrin binding induces Eph forward signaling; however, ephrins can also signal into the cell, which is called reverse signaling (for review, see [29–31]). Using a library of riboprobes for all members of the Eph/ephrin gene family, we systematically mapped with *in situ* hybridizations the complete set of these wiring molecules at different developmental stages. Our results revealed that many members of the Eph/ephrin system can be detected in the developing telencephalon and that they exhibit highly dynamic expression patterns [32, 33]. Based on the spatial and temporal expression patterns we could make some prediction about the potential roles of these wiring molecules in regulating the tangential migration of cortical interneurons. These hypotheses have then been tested with different bioassays *in vitro* and in diverse gene-targeted mouse lines directly *in vivo*.

For example, we could demonstrate that ephrin-A3, which is expressed in the striatum, prevents migrating cortical interneurons from invading this inappropriate region [32]. We could also show that ephrin-A5 is expressed in the VZ of the ganglionic eminences, the dorsal boundary of the migratory route of MGE-derived interneurons, and that this molecule serves as an inhibitory border to channel these neurons into the subventricular zone [34]. Thus, as illustrated in Figure 2, the deep corridor of migrating cortical interneurons is at least in part defined by the concerted action of two different ephrin-A ligands, with ephrin-A5 flanking the dorsal portion and ephrin-A3 the ventral portion of this migratory stream.

These repulsive effects are mediated by the EphA4 receptor, which is expressed by cortical interneurons [32, 34]. As already mentioned above, particular interneuron subtypes are generated in a temporally regulated manner in the MGE, CGE, and POA of the basal telencephalon. We could reveal that POA- and MGE-derived cortical interneurons migrate within spatially segregated corridors. Ephrin-B3, expressed in POA-derived interneurons traversing the superficial route, acts as a repellent signal for deeply migrating interneurons born in the MGE, which is mediated by EphA4 forward signaling. In contrast, EphA4 induces repulsive ephrin-B3 reverse signaling in interneurons generated in the POA, restricting this population to the superficial path (Figure 3). Perturbation of this bidirectional ephrin-B3/EphA4 signaling *in vitro* and in ephrin-B3/EphA4 double mutants *in vivo* leads to a partial intermingling of cells in these segregated migratory pathways and—as shown in Figure 4—to a delayed migration of calbindin-positive interneurons to the cortex. Thus cell contact-mediated bidirectional ephrin-B3/EphA4 signaling mediates the sorting of MGE- and POA-derived interneurons in the deep and superficial migratory stream [33].

We could also demonstrate that EphA4-induced reverse signaling has a motogenic effect of MGE-derived interneurons. In these experiments we first used different *in vitro* assays for cell migration and found that recombinant EphA4 stimulates the migratory speed of cortical interneurons.

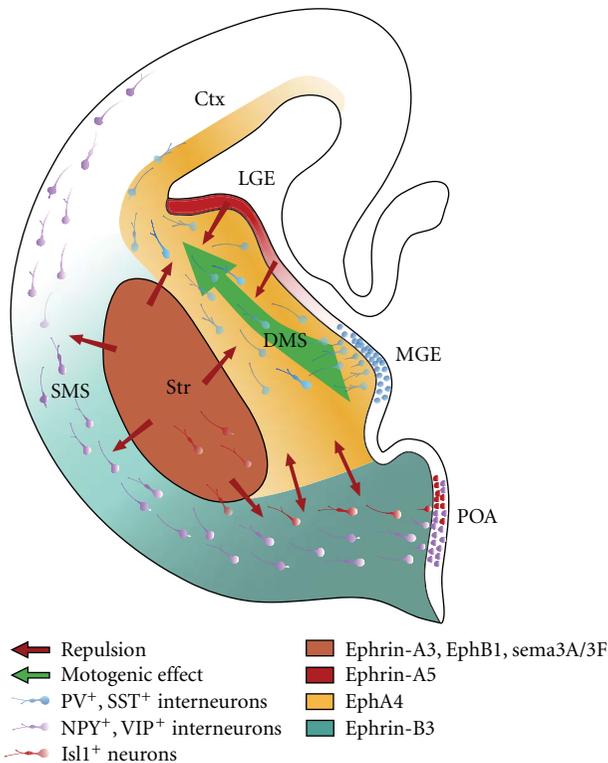


FIGURE 2: Different Eph/ephrin members act in concert to channel migrating MGE-derived neurons towards the cortex. Schematic drawing of a coronal brain slice from the right hemisphere; medial is right; dorsal is top. The MGE gives rise to parvalbumin-(PV-) and somatostatin-(SST-) positive interneurons. They are driven by EphA4 reverse signalling and guided by ephrin-A5, ephrin-A3, and Sema3A/3F forward signalling. The POA gives rise to neuropeptide Y (NPY), vasoactive intestinal protein (VIP) as well as islet 1 (Isl1) positive interneurons that are guided by EphB1/ephrin-B3 signalling preventing the POA-derived cortical interneurons from entering the striatum (Str). The same Eph/ephrin signalling allows the Isl1⁺ neurons to migrate towards the Str. The MGE gives rise to the deep migratory stream (DMS) and the POA provides neurons for the superficial migratory stream (SMS). Those streams are divided by a bidirectional EphA4 and ephrin-B3 signalling.

Thus, in addition to its established role in providing cell-contact mediated repulsion, EphA4 can also tune the molecular machinery for neuronal migration. The ephrin ligands mediating EphA4 reverse signaling and the signal transduction cascade involved in this process are currently under investigation. However, in order to study the function of EphA4 on interneuron migration *in vivo*, we already examined cortical interneurons in an EphA4 knockout mouse line. We found that there was a delayed relocation of calbindin-positive interneurons into the cortex [35].

3. Disrupting Interneuron Migration by Disrupted-in-Schizophrenia 1

Disrupted-in-Schizophrenia 1 (DISC1) is a prominent susceptibility gene for major psychiatric disorders [36, 37]. The biological functions attributed to the DISC1 protein

are complex and highly diverse. For example, previous work suggested that DISC1 plays an important role during neuronal proliferation, differentiation, neurite outgrowth, and synapse formation (reviewed in [38, 39]). There are also some studies that report that DISC1 is a necessary component for the correct positioning of radially migrating cortical pyramidal neurons [40, 41]. This prompted us to study the potential role of DISC1 for interneuron migration.

For this we first performed RT-PCR, *in situ* hybridization and immunostainings to verify that DISC1 is expressed in the MGE at the appropriate developmental stages. We also examined the subcellular distribution of DISC1. As illustrated in Figure 5, DISC1 is expressed in the tips of the leading processes. In addition, we also found DISC1 immunoreactivity at the rear of the nucleus, opposite to the leading process. A closer inspection revealed that DISC1 colocalizes with LIS1, previously described as a centrosomal protein [42] (Figure 6). Thus DISC1 is found in important strategic positions to control the molecular machinery involved in interneuron migration, for example, by interacting with cytoskeletal proteins tubulin and actin, motor proteins of the dynein and kinesin family, and regulatory proteins [40, 43].

To examine the functional role of DISC1 during interneuron migration, we performed *in utero* and *ex utero* electroporation to suppress DISC1 in the MGE *in vivo* and *in vitro*. Our results indicate that, after DISC1 knockdown, the proportion of tangentially migrating MGE neurons that reached their cortical target was reduced by 15%. In addition, there were profound alterations in the morphology of DISC1-deficient neurons, which exhibited longer and less branched leading processes than control cells [44].

These results indicate that DISC1 has an impact on the migratory behaviour of interneurons during early development that might lead to deficits in the number and/or composition of GABAergic neurons in the cortex. As mentioned in Section 1, dysfunctions of local GABAergic circuits have been often associated with the pathophysiology of schizophrenia [12]. Thus our findings support the notion that schizophrenia is a neurodevelopmental disease that may result from defects in interneuron integration [45].

4. Inhibitory Regulation of Sensory Cortical Plasticity

Vulnerability for psychiatric diseases is not uniform throughout life, but is increased during certain stages of pre- and postnatal development [46, 47]. The factors that make neuronal subsystems especially open for environmental influences during well-defined, so-called critical periods have been the subject of much research in the neurosciences. As a typical example, the binocular visual cortex exhibits a well-studied critical period during which, under undisturbed conditions, the orientation preferences for stimuli seen through one versus the other eye are harmonised in visual cortical neurons [48]. It has long been known that temporary closure (i.e., monocular deprivation) of one eye during this

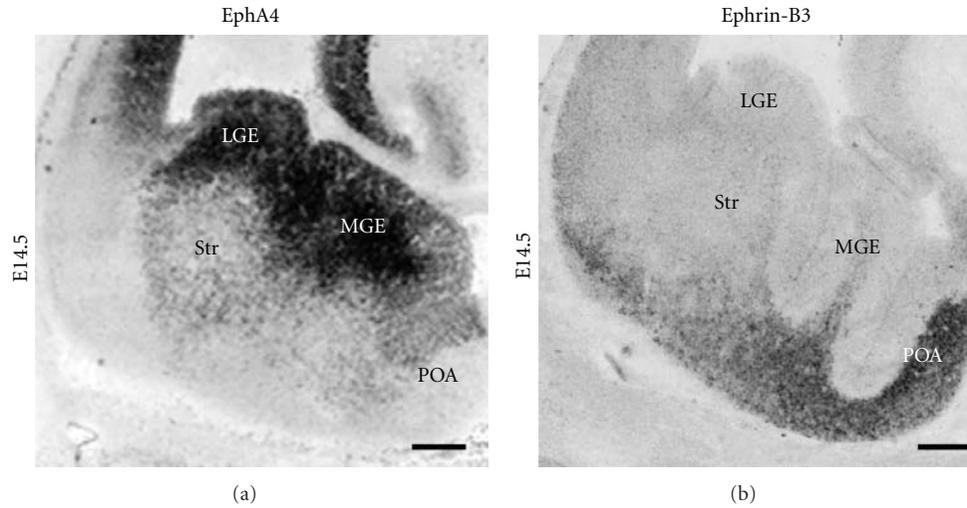


FIGURE 3: Complementary EphA4 and ephrin-B3 *in situ* hybridization expression patterns. On embryonic day E14.5 EphA4 and ephrin-B3 are complementary expressed in the basal telencephalon of mouse brains. Based on these expression patterns several predictions about the functional role of these wiring molecules on interneuron migration could be made (see text for details). Scale bar: 100 μm .

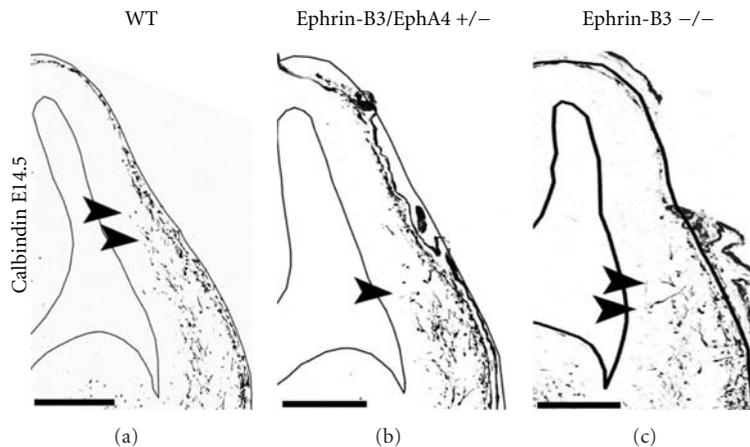


FIGURE 4: Alterations in the Eph/ephrin system cause a delayed migration of cortical interneurons *in vivo*. Analysing the calbindin-(CB-) positive interneurons in the cortex of E14.5 embryos revealed a delayed migration of interneurons in hetero- and homozygotic ephrin-B3/EphA4 knockout mice compared to the wild-type (WT) littermates. Arrow heads indicate the front of migrating interneurons. Scale bar: 100 μm .

critical period will shift the excitability of cortical neurons towards the open eye [49, 50]. This critical period for ocular dominance plasticity starts a few days after eye opening in mice, has a maximum at about postnatal day 28, and ends at around postnatal day 32, when short periods of deprivation have no detectable effect in the cortex [50]. Longer periods of deprivation, however, are still able to induce ocular dominance plasticity until postnatal day 100 (P100), but no longer (Figure 7, [51]).

The mechanisms regulating this period of enhanced plasticity have been the subject of intense research for many years. It has become obvious that the critical period is initiated by a shift in the cortical balance of excitation and inhibition [52, 53] (see [54] for review), a shift that is effected by the maturation of fast-spiking GABAergic interneurons, the so-called basket cells which are characterised by their expression of parvalbumin [55]. Thus, the start of the

critical period is delayed in knockout mice lacking the GABA synthesising enzyme GAD65 and is triggered in these mice as soon as inhibition at GABA_A receptors is increased by the intracerebral infusion of diazepam [52]. Enhanced inhibition in early childhood prepones the start of the critical period [53, 56, 57]. On the other hand, several interventions have been shown in recent years to reinstate critical period-like plasticity in adult animals by reducing intracortical inhibition: treatment with the antidepressant fluoxetine allows for ocular dominance plasticity in adult rats [58]; the effect is accompanied by reduced cortical GABA levels and prevented by diazepam infusion. Additionally, fluoxetine treatment promotes the recovery from amblyopia of adult rats [58]. The same holds true for environmental enrichment, and again diazepam infusion averts the effect [59]. Indeed, directly attenuating GABA release by a GAD inhibitor reestablishes CP-like plasticity in adult rats [60].

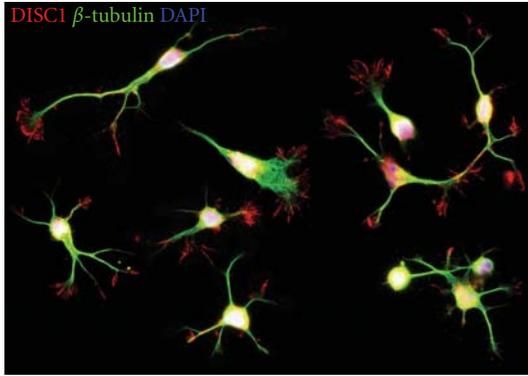


FIGURE 5: DISC1 immunocytochemistry on MGE-derived interneurons. Photomicrograph of MGE-derived neurons immunostained with DISC1 antibodies (red) and β -Tubulin antibodies (green) show a strong DISC1 signal in the processes of the interneurons.

Thus, it seems that GABAergic neurons single-handedly regulate visual cortical plasticity, and this impression is even enhanced by the recent observation that intracortical transplantation of embryonal cells from the medial ganglionic eminence (MGE), which are destined to become GABAergic cortical interneurons, induces a period of enhanced plasticity in mice beyond the critical period [61]. Interestingly, this effect is only present during a narrow time frame when the transplanted cells have a certain cellular age (33 to 35 day) corresponding to the age they would have had during the natural critical period in normal development.

Surprising as this active time window may be, it is in line with a lot of research on the activity-dependent maturation of parvalbumin-containing interneurons in the visual cortex. Before and during the critical period for ocular dominance plasticity, the strength of cortical inhibition triples. This increase is prevented by dark rearing which also delays the critical period [62]. Further research specified that the number of perisomatic boutons around pyramidal cells in the visual cortex increases until postnatal day 28, which marks the peak of the critical period; again, visual deprivation prevented the increase [63]. These results imply that the maturation of basket cells is necessary for the start of the critical period for ocular dominance plasticity, and they show that this maturation is activity dependent. As mentioned above, transsynaptic transfer of the homeoprotein Otx2 from the retina, which is triggered by light perception, has been shown to induce the maturation of parvalbumin-containing interneurons and the start of the critical period [55]. A high concentration of polysialic acid (PSA), which traps Otx2 [64], has been shown to prevent the start of the critical period, such that premature removal of PSA leads to an earlier maturation of parvalbumin-expressing cells and a preponed critical period [65]. Recent work has elucidated to a large degree the cell-autonomous mechanisms that promote the developmental synapse formation in basket cells: a knockdown of the GABA-synthesizing enzyme GAD67 leads to deficits in axon branching and perisomatic synapse formation, whereas overexpression of GAD67 speeds up these processes [66], (see [67] for review).

Thus, inhibitory innervation patterns are regulated by the cell's own activity, which in turn depends on GABA synthesis. An interesting recent twist in this story is the finding that the complete blockade of GABA synthesis in single cells does not, as one would expect, shrink axonal arbors drastically, but contrariwise increases their density and complexity [68]. The authors of that study suggest a model according to which basket cells make tentative contacts with many potential postsynaptic targets, which are pruned or stabilized by synaptic activity. While little activity is sufficient to remove, but not to stabilize connections, complete blockade allows for neither and therefore keeps axonal complexity and synapse number high [68]. In summary, visual stimulation induces the maturation of parvalbumin-containing basket cells, which is internally regulated by GABAergic activity, and shifts the excitation-inhibition balance of visual cortical neurons such that critical period plasticity becomes possible.

While it seems established that GABAergic inhibition controls the level of cortical plasticity, it is much less clear in how far GABAergic neurons are involved in the expression of plastic changes. Even after the drastic reduction of visual cortical inhibition just about the level that would evoke seizures, the ocular dominance shift evoked by monocular deprivation can be observed, and complete silencing of intracortical connections by muscimol confirms that the effective changes are expressed at thalamocortical synapses [69]. Indeed, ocular dominance plasticity is dependent on Hebbian plasticity at NMDA receptors [70–72]. Do inhibitory interneurons, then, participate in synaptic plasticity at all?

Several studies have tried to answer this question. Using calcium imaging, one recent study showed that GABAergic neurons are more binocular, that is, less dominated by one eye, in normal mice, but show a similar shift toward the open eye after monocular deprivation during the critical period [73]. If monocular deprivation was performed after the critical period, the ocular dominance shift of GABAergic neurons was even stronger than that of excitatory neurons. Another study, however, provided somewhat conflicting results: *In vivo* intracellular recording from pyramidal cells and fast-spiking interneurons showed that while excitatory cells have a normal bias towards the contralateral eye which they rapidly lose after a short monocular deprivation, fast-spiking interneurons were unbiased at the outset, showed a paradoxical shift towards the closed eye after short deprivation, and only shifted on to an ipsilateral bias after longer deprivation periods [74]. The discrepancies may arise from differences in anaesthesia and method or simply from the fact that the latter study focused on fast-spiking, parvalbumin-containing interneurons which play a central role in regulating the critical period for ocular dominance plasticity, whereas the Kameyama study did not distinguish among GABAergic neuronal subtypes. Both studies agree, however, that intracortical inhibition changes in response to monocular deprivation, and in adult plasticity, this change may even have a stronger influence on network function than the relatively small change in excitatory transmission [73].

In another primary sensory area, the somatosensory cortex, the role of GABAergic interneurons in the response to sensory deprivation has been firmly established in recent

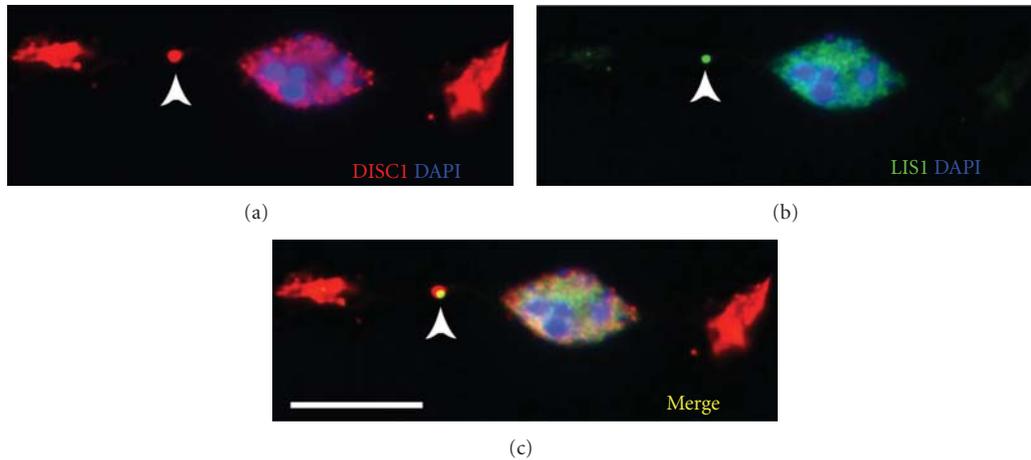


FIGURE 6: DISC1 and LIS1 immunocytochemistry on MGE-derived cells. Photomicrograph of an MGE-derived cell that was coimmunolabeled with a DISC1 antibody (a) and a LIS1 antibody (b). (c) represents the merged picture of (a) and (b). Note the precise overlap of DISC1 and LIS1 at the centrosome (yellow in c) pinpointed by the arrow heads. Scale bar: 10 μ m.

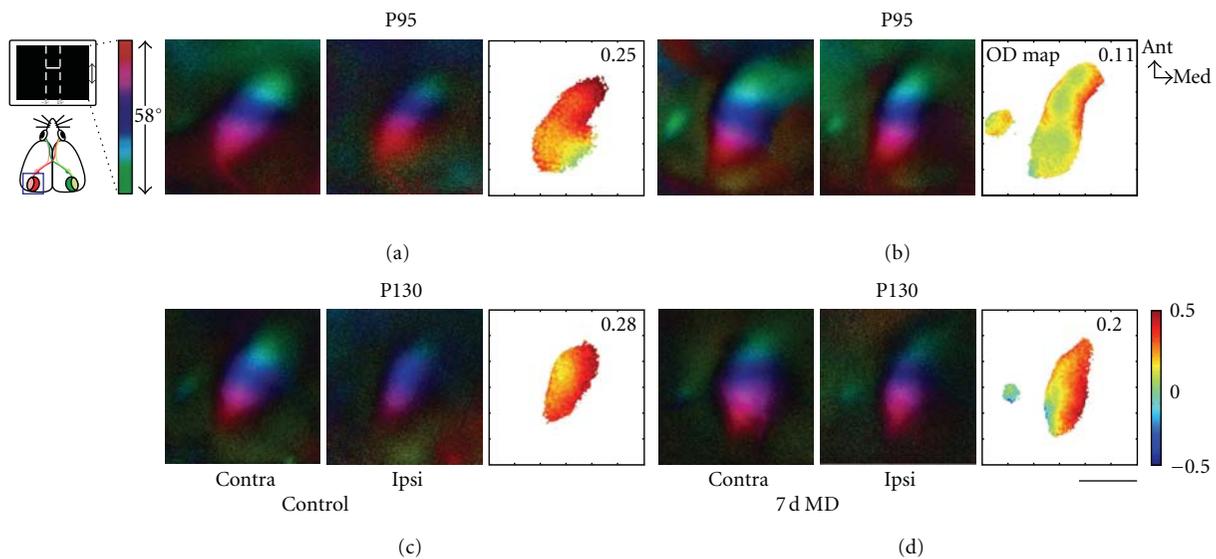


FIGURE 7: Adult ocular dominance plasticity ceases at postnatal day (P) 100. Retinotopic maps of the binocular visual cortex are shown. Elevation is colour coded according to the scheme on the left. Polar maps obtained by stimulation of the contralateral (contra) or ipsilateral (ipsi) eye are illustrated for four representative mice. Ocular dominance (OD) in the visual cortex is shown in the OD maps on the right, colour-coded according to the scheme on the right. In nondeprived control mice both before (a) and after (c) P100, the contralateral eye activates the cortex more strongly than the ipsilateral eye, which is reflected in warm-coloured OD maps. Seven days of monocular deprivation shift OD towards the ipsilateral eye in P95 animals ((b), colder colours of OD map), but have no such effect in the P130 animal (d). Scale: 1 mm.

years (see [75] for review). If a single row of whiskers was removed in mice starting on postnatal day 7, the number of parvalbumin-positive interneurons was significantly reduced in the cortical barrels representing that row, whereas it was increased in adjacent barrels [76]. This loss of parvalbumin expression went along with a lower number of perisomatic synaptic varicosities and weaker inhibitory transmission, an effect that required experience-dependent release of BDNF [77]. Further research confirmed the reduction in parvalbumin expression and showed that, upon whisker trimming, fast-spiking interneurons in the barrel cortex, but not other kinds of nonpyramidal cells, become less excitable, while their excitatory thalamocortical input is reduced [78].

Interestingly, these findings are somewhat at variance with earlier research demonstrating that in rats in which a row of whiskers was plucked between postnatal days 1 and 60, the number of GABAergic synapses on dendritic spines, but not on somata, was strongly reduced in the deprived barrels [79] (see [80] for review). A potential approach to reconcile these conflicting results might be that whisker trimming at a very early age delays the maturation of cortical inhibition in a similar way as dark rearing does in the visual cortex. Here, this kind of deprivation also results in reduced parvalbumin expression [81]. As parvalbumin-positive interneurons mature quickly before the onset of the critical period for ocular dominance plasticity in the visual

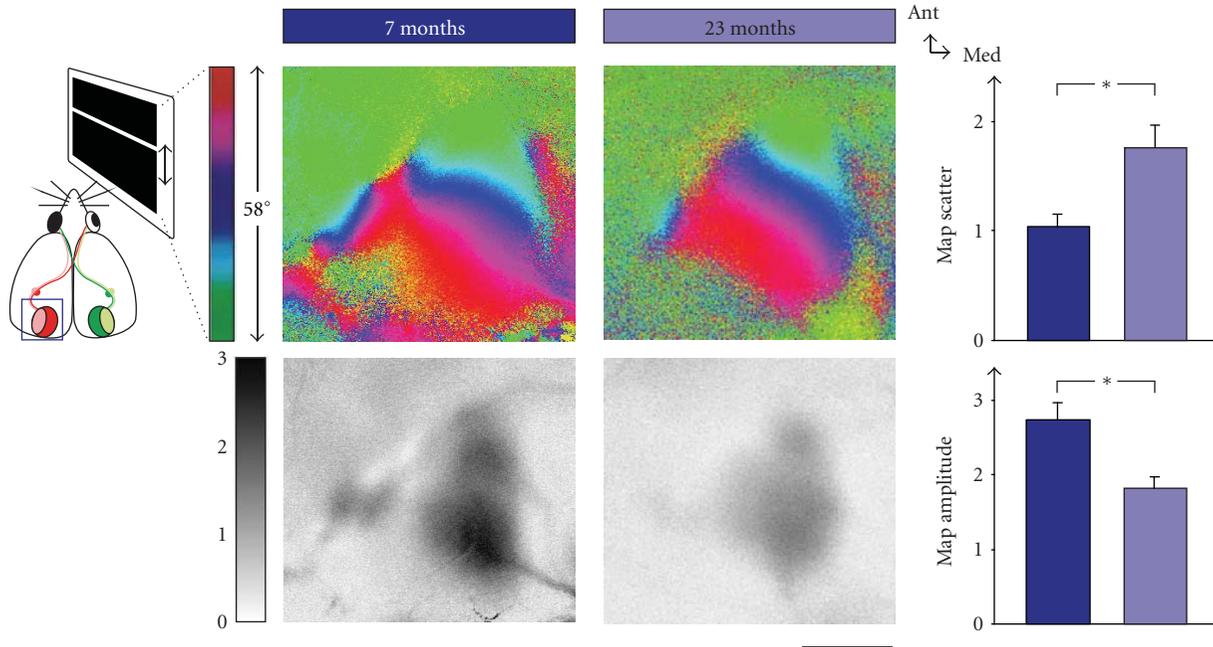


FIGURE 8: Visual cortical maps deteriorate in old age. Retinotopic phase maps (colour-coded according to scheme on the top left) and corresponding activity maps (coded according to grey scale on the left) are shown for mice of seven and 23 months. Map scatter increases in old mice, while the amplitude decreases. Scale bar is 1 mm and applies to all panels showing maps.

cortex [63, 82], so they do in the somatosensory cortex between postnatal days 10 and 30 [83]. It is conceivable that only a longer period of deprivation (two months in [79]) allows for the full adaptation to changed input.

Although such issues still need to be clarified, it appears that the mechanisms regulating sensory plasticity are similar in different cortical areas (see [84] for review), involving the expression of parvalbumin and the formation of perisomatic synapses by basket interneurons in both the somatosensory and the visual cortex. In the auditory cortex, too, similar processes seem to control critical period plasticity [85, 86]. This uniformity of function across the neocortex holds a promise for our ability to understand cortical plasticity, but a lot of work still remains to be done to understand the cellular and network mechanisms by which plasticity is enabled.

5. Age-Related Decline of Inhibition and Signal Processing

Perceptual sensitivity declines in old age. In aged humans, visual acuity and contrast sensitivity decline in an accelerated fashion [87–89]. In order to test in how far mice might serve as an animal model of age-related vision loss in humans, we have just shown that, in pigmented mice, too, visual acuity and contrast sensitivity deteriorate, starting at approx. 18 months of age, and the progressive loss parallels that which is seen in humans [90]. While age-related degradations in the sensory organs certainly impair access to the environment, there has been a long-standing notion that central nervous changes may also contribute to the decline in visual or somatosensory function [91, 92]. Indeed, we could show in

the same study that visual cortical activity, as measured by optical imaging of intrinsic signals, and cortex-dependent behavioural plasticity were strongly reduced in old mice (Figure 8, [90]).

A precise connection between cellular changes and visual function loss in old animals has been achieved in macaques. In these animals, orientation tuning of visual cortical neurons is reduced to the point of being scarcely detectable [93, 94]. Electrophoretic application of GABA or the GABA agonist muscimol to the recorded neurons, however, restored orientation selectivity similar to that seen in young animals, whereas the GABA receptor antagonist bicuculline abolished orientation tuning in visual cortical neurons of young monkeys [94]. A similar degradation of visual cortical function, that is, decreased orientation sensitivity, higher spontaneous activity, and lower signal-to-noise ratio, was observed in aged cats and, partly, in rats [95, 96]. Looking for the reason of this decrease in intracortical inhibition, Hua and colleagues [97] found the density and proportion of GABA-immunopositive neurons in the cat visual cortex to be decreased by about half in old compared to young animals, whereas there was no change in the number of excitatory neurons.

Does this loss of inhibitory interneurons affect all GABAergic subtypes equally? In humans and dogs it appears that basket cells characterised by parvalbumin are remarkably resilient to old age [98, 99]. This fits to the most recent finding that the activity of parvalbumin-containing interneurons has almost no effect on the orientation tuning of visual cortical pyramidal cells [100], whereas a genetically induced loss of dendrite-targeting interneurons leads to impaired orientation tuning in the mouse visual

cortex [101]. The density of calbindin-immunoreactive interneurons, in contrast, is diminished in almost all of the aged human brain, but significantly so only in a few areas, among them the primary visual cortex [98]. The density of calretinin-positive neurons is also affected by age, but mostly in temporal areas. Somewhat conflicting results were found in the somatosensory and motor cortices of rats, where the density of both parvalbumin- and somatostatin-positive interneurons was found to be decreased in old animals [102]. It is as yet unclear whether the discrepancy concerning parvalbumin-positive cells is due to differences in brain region, strain, or species. It appears certain, however, that some kinds of GABAergic interneurons are lost during ageing and that this loss may be the reason for the functional degradation found in old animals. This knowledge may provide a basis for future therapeutic interventions.

6. Conclusions

Despite being only a minority of all cortical neurons, inhibitory interneurons have a key role in modulating cortical function and plasticity, and even subtle impairments to the integrity of these cells can lead to severe neuronal and psychiatric disturbances. It is therefore crucial to understand the development of GABAergic interneurons, their integration into cortical circuits, and the factors necessary for their preservation. In consequence, it might become possible to treat neuronal disorders at their basic circuit level.

Acknowledgment

The authors thank Falk Nitzsche for providing Figures 5 and 6.

References

- [1] S. R. Y. Cajal, "Structure and connections of neurons," *Bulletin of the Los Angeles Neurological Society*, vol. 17, no. 1-2, pp. 5–46, 1952.
- [2] E. G. Jones, "Varieties and distribution of non pyramidal cells in the somatic sensory cortex of the squirrel monkey," *Journal of Comparative Neurology*, vol. 160, no. 2, pp. 205–267, 1975.
- [3] J. DeFelipe, "Cortical interneurons: from Cajal to 2001," *Progress in Brain Research*, vol. 136, pp. 215–238, 2002.
- [4] Y. Kawaguchi and S. Kondo, "Parvalbumin, somatostatin and cholecystokinin as chemical markers for specific GABAergic interneuron types in the rat frontal cortex," *Brain Cell Biology*, vol. 31, no. 3-5, pp. 277–287, 2002.
- [5] R. Batista-Brito and G. Fishell, "Chapter 3 the developmental integration of cortical interneurons into a functional network," *Current Topics in Developmental Biology*, vol. 87, pp. 81–118, 2009.
- [6] H. Markram, M. Toledo-Rodriguez, Y. Wang, A. Gupta, G. Silberberg, and C. Wu, "Interneurons of the neocortical inhibitory system," *Nature Reviews Neuroscience*, vol. 5, no. 10, pp. 793–807, 2004.
- [7] G. A. Ascoli, L. Alonso-Nanclares, S. A. Anderson et al., "Petilla terminology: nomenclature of features of GABAergic interneurons of the cerebral cortex," *Nature Reviews Neuroscience*, vol. 9, no. 7, pp. 557–568, 2008.
- [8] R. Batista-Brito, R. MacHold, C. Klein, and G. Fishell, "Gene expression in cortical interneuron precursors is prescient of their mature function," *Cerebral Cortex*, vol. 18, no. 10, pp. 2306–2317, 2008.
- [9] G. Sanacora, G. F. Mason, and J. H. Krystal, "Impairment of GABAergic transmission in depression: new insights from neuroimaging studies," *Critical Reviews in Neurobiology*, vol. 14, no. 1, pp. 23–45, 2000.
- [10] J. M. Rennie and G. B. Boylan, "Neonatal seizures and their treatment," *Current Opinion in Neurology*, vol. 16, no. 2, pp. 177–181, 2003.
- [11] P. Levitt, "Developmental neurobiology and clinical disorders: lost in translation?" *Neuron*, vol. 46, no. 3, pp. 407–412, 2005.
- [12] D. A. Lewis, T. Hashimoto, and D. W. Volk, "Cortical inhibitory neurons and schizophrenia," *Nature Reviews Neuroscience*, vol. 6, no. 4, pp. 312–324, 2005.
- [13] P. Levitt, K. L. Eagleson, and E. M. Powell, "Regulation of neocortical interneuron development and the implications for neurodevelopmental disorders," *Trends in Neurosciences*, vol. 27, no. 7, pp. 400–406, 2004.
- [14] P. Rakic, "Radial versus tangential migration of neuronal clones in the developing cerebral cortex," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 92, no. 25, pp. 11323–11327, 1995.
- [15] A. A. Lavdas, M. Grigoriou, V. Pachnis, and J. G. Parnavelas, "The medial ganglionic eminence gives rise to a population of early neurons in the developing cerebral cortex," *Journal of Neuroscience*, vol. 19, no. 18, pp. 7881–7888, 1999.
- [16] J. G. Parnavelas, S. A. Anderson, A. A. Lavdas, M. Grigoriou, V. Pachnis, and J. L. R. Rubenstein, "The contribution of the ganglionic eminence to the neuronal cell types of the cerebral cortex," *Novartis Foundation Symposium*, vol. 228, pp. 129–147, 2000.
- [17] S. A. Anderson, O. Marín, C. Horn, K. Jennings, and J. L. R. Rubenstein, "Distinct cortical migrations from the medial and lateral ganglionic eminences," *Development*, vol. 128, no. 3, pp. 353–363, 2001.
- [18] O. Marín and J. L. R. Rubenstein, "A long, remarkable journey: tangential migration in the telencephalon," *Nature Reviews Neuroscience*, vol. 2, no. 11, pp. 780–790, 2001.
- [19] S. Nery, H. Wichterle, and G. Fishell, "Sonic hedgehog contributes to oligodendrocyte specification in the mammalian forebrain," *Development*, vol. 128, no. 4, pp. 527–540, 2001.
- [20] H. Wichterle, D. H. Turnbull, S. Nery, G. Fishell, and A. Alvarez-Buylla, "In utero fate mapping reveals distinct migratory pathways and fates of neurons born in the mammalian basal forebrain," *Development*, vol. 128, no. 19, pp. 3759–3771, 2001.
- [21] L. R. Hernández-Miranda, J. G. Parnavelas, and F. Chiara, "Molecules and mechanisms involved in the generation and migration of cortical interneurons," *ASN Neuro*, vol. 2, no. 2, pp. 75–86, 2010.
- [22] E. Rossignol, "Genetics and function of neocortical GABAergic interneurons in neurodevelopmental disorders," *Neural Plasticity*, vol. 2011, Article ID 649325, 2011.
- [23] O. Marín, A. Yaron, A. Bagri, M. Tessier-Lavigne, and J. L. R. Rubenstein, "Sorting of striatal and cortical interneurons regulated by semaphorin-neuropilin interactions," *Science*, vol. 293, no. 5531, pp. 872–875, 2001.
- [24] W. Andrews, M. Barber, L. R. Hernandez-Miranda et al., "The role of Slit-Robo signaling in the generation, migration and morphological differentiation of cortical interneurons," *Developmental Biology*, vol. 313, no. 2, pp. 648–658, 2008.

- [25] N. Flames, J. E. Long, A. N. Garratt et al., "Short- and long-range attraction of cortical GABAergic interneurons by neuregulin-1," *Neuron*, vol. 44, no. 2, pp. 251–561, 2004.
- [26] J. G. Flanagan and P. Vanderhaeghen, "The ephrins and Eph receptors in neural development," *Annual Review of Neuroscience*, vol. 21, pp. 309–345, 1998.
- [27] J. P. Himanen, M. J. Chumley, M. Lackmann et al., "Repelling class discrimination: Ephrin-A5 binds to and activates EphB2 receptor signaling," *Nature Neuroscience*, vol. 7, no. 5, pp. 501–509, 2004.
- [28] A. Martínez and E. Soriano, "Functions of ephrin/Eph interactions in the development of the nervous system: emphasis on the hippocampal system," *Brain Research Reviews*, vol. 49, no. 2, pp. 211–226, 2005.
- [29] A. Davy and P. Soriano, "Ephrin signaling in vivo: look both ways," *Developmental Dynamics*, vol. 232, no. 1, pp. 1–10, 2005.
- [30] R. Klein, "Bidirectional modulation of synaptic functions by Eph/ephrin signaling," *Nature Neuroscience*, vol. 12, no. 1, pp. 15–20, 2009.
- [31] J. Egea and R. Klein, "Bidirectional Eph-ephrin signaling during axon guidance," *Trends in Cell Biology*, vol. 17, no. 5, pp. 230–238, 2007.
- [32] J. Rudolph, G. Zimmer, A. Steinecke, S. Barchmann, and J. Bolz, "Ephrins guide migrating cortical interneurons in the basal telencephalon," *Cell Adhesion and Migration*, vol. 4, no. 3, pp. 400–408, 2010.
- [33] G. Zimmer, J. Rudolph, J. Landmann et al., "Bidirectional EphrinB3/EphA4 signaling mediates the segregation of medial ganglionic eminence- and preoptic area-derived interneurons in the deep and superficial migratory stream," *Journal of Neuroscience*, vol. 31, no. 50, pp. 18364–18380, 2011.
- [34] G. Zimmer, P. Garcez, J. Rudolph et al., "Ephrin-A5 acts as a repulsive cue for migrating cortical interneurons," *European Journal of Neuroscience*, vol. 28, no. 1, pp. 62–73, 2008.
- [35] A. Steinecke, J. Rudolph, G. Zimmer, S. Barchmann, and J. Bolz, "A dual role of EphA4 signaling in the basal telencephalon during interneuron migration," *Society for Neuroscience, Abstract*, 2010, Neuroscience Meeting Planner.
- [36] D. H. Blackwood, P. M. Visscher, and W. J. Muir, "Genetic studies of bipolar affective disorder in large families," *The British Journal of Psychiatry*, vol. 41, pp. s134–s136, 2001.
- [37] J. E. Chubb, N. J. Bradshaw, D. C. Soares, D. J. Porteous, and J. K. Millar, "The DISC locus in psychiatric illness," *Molecular Psychiatry*, vol. 13, no. 1, pp. 36–64, 2008.
- [38] N. J. Brandon and A. Sawa, "Linking neurodevelopmental and synaptic theories of mental illness through DISC1," *Nature Reviews Neuroscience*, vol. 12, no. 12, pp. X707–X722, 2011.
- [39] N. J. Brandon, J. K. Millar, C. Korth, H. Sive, K. K. Singh, and A. Sawa, "Understanding the role of DISC1 in psychiatric disease and during normal development," *Journal of Neuroscience*, vol. 29, no. 41, pp. 12768–12775, 2009.
- [40] A. Kamiya, K. I. Kubo, T. Tomoda et al., "A schizophrenia-associated mutation of DISC1 perturbs cerebral cortex development," *Nature Cell Biology*, vol. 7, no. 12, pp. 1067–1078, 2005.
- [41] T. L. Young-Pearse, S. Suth, E. S. Luth, A. Sawa, and D. J. Selkoe, "Biochemical and functional interaction of disrupted-in-schizophrenia 1 and amyloid precursor protein regulates neuronal migration during mammalian cortical development," *Journal of Neuroscience*, vol. 30, no. 31, pp. 10431–10440, 2010.
- [42] N. J. Brandon, E. J. Handford, I. Schurov et al., "Disrupted in Schizophrenia 1 and Nudel form a neurodevelopmentally regulated protein complex: implications for schizophrenia and other major neurological disorders," *Molecular and Cellular Neuroscience*, vol. 25, no. 1, pp. 42–55, 2004.
- [43] Q. Wang and N. J. Brandon, "Regulation of the cytoskeleton by Disrupted-in-Schizophrenia 1 (DISC1)," *Molecular and Cellular Neuroscience*, vol. 48, no. 4, pp. 359–364, 2011.
- [44] A. Steinecke, C. Gampe, C. Valkova, C. Kaether, and J. Bolz, "Disrupted-in-schizophrenia 1 (DISC1) is necessary for the correct migration of cortical interneurons," *Journal of Neuroscience*, vol. 32, no. 2, pp. 738–745, 2012.
- [45] D. A. Lewis and P. Levitt, "Schizophrenia as a disorder of neurodevelopment," *Annual Review of Neuroscience*, vol. 25, pp. 409–432, 2002.
- [46] P. Kalus, D. Senitz, and H. Beckmann, "Disturbances of corticogenesis in schizophrenia: morphological findings provide new evidence for the maldevelopmental hypothesis," *Neuropsychobiology*, vol. 40, no. 1, pp. 1–13, 1999.
- [47] F. Bagorda, G. Teuchert-Noodt, and K. Lehmann, "Isolation rearing or methamphetamine traumatization induce a "dysconnection" of prefrontal efferents in gerbils: implications for schizophrenia," *Journal of Neural Transmission*, vol. 113, no. 3, pp. 365–379, 2006.
- [48] B. S. Wang, R. Sarnaik, and J. Cang, "Critical period plasticity matches binocular orientation preference in the visual cortex," *Neuron*, vol. 65, no. 2, pp. 246–256, 2010.
- [49] D. H. Hubel and T. N. Wiesel, "The period of susceptibility to the physiological effects of unilateral eye closure in kittens," *Journal of Physiology*, vol. 206, no. 2, pp. 419–436, 1970.
- [50] J. A. Gordon and M. P. Stryker, "Experience-dependent plasticity of binocular responses in the primary visual cortex of the mouse," *Journal of Neuroscience*, vol. 16, no. 10, pp. 3274–3286, 1996.
- [51] K. Lehmann and S. Löwel, "Age-dependent ocular dominance plasticity in adult mice," *PLoS ONE*, vol. 3, no. 9, Article ID e3120, 2008.
- [52] T. K. Hensch, M. Fagiolini, N. Mataga, M. P. Stryker, S. Baekkeskov, and S. F. Kash, "Local GABA circuit control of experience-dependent plasticity in developing visual cortex," *Science*, vol. 282, no. 5393, pp. 1504–1508, 1998.
- [53] M. Fagiolini and T. K. Hensch, "Inhibitory threshold for critical-period activation in primary visual cortex," *Nature*, vol. 404, no. 6774, pp. 183–186, 2000.
- [54] T. K. Hensch, "Critical period regulation," *Annual Review of Neuroscience*, vol. 27, pp. 549–579, 2004.
- [55] S. Sugiyama, A. A. Di Nardo, S. Aizawa et al., "Experience-dependent transfer of Otx2 homeoprotein into the visual cortex activates postnatal plasticity," *Cell*, vol. 134, no. 3, pp. 508–520, 2008.
- [56] J. L. Hanover, Z. J. Huang, S. Tonegawa, and M. P. Stryker, "Brain-derived neurotrophic factor overexpression induces precocious critical period in mouse visual cortex," *The Journal of Neuroscience*, vol. 19, no. 22, p. RC40, 1999.
- [57] Z. J. Huang, A. Kirkwood, T. Pizzorusso et al., "BDNF regulates the maturation of inhibition and the critical period of plasticity in mouse visual cortex," *Cell*, vol. 98, no. 6, pp. 739–755, 1999.
- [58] J. F. M. Vetencourt, A. Sale, A. Viegi et al., "The antidepressant fluoxetine restores plasticity in the adult visual cortex," *Science*, vol. 320, no. 5874, pp. 385–388, 2008.
- [59] A. Sale, J. F. Maya Vetencourt, P. Medini et al., "Environmental enrichment in adulthood promotes amblyopia

- recovery through a reduction of intracortical inhibition," *Nature Neuroscience*, vol. 10, no. 6, pp. 679–681, 2007.
- [60] A. Harauzov, M. Spolidoro, G. DiCristo et al., "Reducing intracortical inhibition in the adult visual cortex promotes ocular dominance plasticity," *Journal of Neuroscience*, vol. 30, no. 1, pp. 361–371, 2010.
- [61] D. G. Southwell, R. C. Froemke, A. Alvarez-Buylla, M. P. Stryker, and S. P. Gandhi, "Cortical plasticity induced by inhibitory neuron transplantation," *Science*, vol. 327, no. 5969, pp. 1145–1148, 2010.
- [62] B. Morales, S. Y. Choi, and A. Kirkwood, "Dark rearing alters the development of GABAergic transmission in visual cortex," *Journal of Neuroscience*, vol. 22, no. 18, pp. 8084–8090, 2002.
- [63] B. Chattopadhyaya, G. Di Cristo, H. Higashiyama et al., "Experience and activity-dependent maturation of perisomatic GABAergic innervation in primary visual cortex during a postnatal critical period," *Journal of Neuroscience*, vol. 24, no. 43, pp. 9598–9611, 2004.
- [64] A. H. Joliot, A. Triller, M. Volovitch, C. Pernelle, and A. Prochiantz, " α -2,8-polysialic acid is the neuronal surface receptor of antennapedia homeobox peptide," *New Biologist*, vol. 3, no. 11, pp. 1121–1134, 1991.
- [65] G. Di Cristo, B. Chattopadhyaya, S. J. Kuhlman et al., "Activity-dependent PSA expression regulates inhibitory maturation and onset of critical period plasticity," *Nature Neuroscience*, vol. 10, no. 12, pp. 1569–1577, 2007.
- [66] B. Chattopadhyaya, G. Di Cristo, C. Z. Wu et al., "GAD67-mediated GABA synthesis and signaling regulate inhibitory synaptic innervation in the visual cortex," *Neuron*, vol. 54, no. 6, pp. 889–903, 2007.
- [67] Z. J. Huang, G. Di Cristo, and F. Ango, "Development of GABA innervation in the cerebral and cerebellar cortices," *Nature Reviews Neuroscience*, vol. 8, no. 9, pp. 673–686, 2007.
- [68] X. Wu, Y. Fu, G. Knott, J. Lu, G. D. Cristo, and Z. Josh Huang, "GABA signaling promotes synapse elimination and axon pruning in developing cortical inhibitory interneurons," *Journal of Neuroscience*, vol. 32, no. 1, pp. 331–343, 2012.
- [69] L. A. Khibnik, K. K. A. Cho, and M. F. Bear, "Relative contribution of feedforward excitatory connections to expression of ocular dominance plasticity in layer 4 of visual cortex," *Neuron*, vol. 66, no. 4, pp. 493–500, 2010.
- [70] A. Kleinschmidt, M. F. Bear, and W. Singer, "Blockade of 'NMDA' receptors disrupts experience dependent plasticity of kitten striate cortex," *Science*, vol. 238, no. 4825, pp. 355–358, 1987.
- [71] N. B. Sawtell, M. Y. Frenkel, B. D. Philpot, K. Nakazawa, S. Tonegawa, and M. F. Bear, "NMDA receptor-dependent ocular dominance plasticity in adult visual cortex," *Neuron*, vol. 38, no. 6, pp. 977–985, 2003.
- [72] M. Sato and M. P. Stryker, "Distinctive features of adult ocular dominance plasticity," *Journal of Neuroscience*, vol. 28, no. 41, pp. 10278–10286, 2008.
- [73] K. Kameyama, K. Sohya, T. Ebina, A. Fukuda, Y. Yanagawa, and T. Tsumoto, "Difference in binocularity and ocular dominance plasticity between GABAergic and excitatory cortical neurons," *Journal of Neuroscience*, vol. 30, no. 4, pp. 1551–1559, 2010.
- [74] Y. Yazaki-Sugiyama, S. Kang, H. Cteau, T. Fukai, and T. K. Hensch, "Bidirectional plasticity in fast-spiking GABA circuits by visual experience," *Nature*, vol. 462, no. 7270, pp. 218–221, 2009.
- [75] Q. Q. Sun, "The missing piece in the "use it or lose it" puzzle: is inhibition regulated by activity or does it act on its own accord?" *Reviews in the Neurosciences*, vol. 18, no. 3-4, pp. 295–310, 2007.
- [76] Y. Jiao, C. Zhang, Y. Yanagawa, and Q. Q. Sun, "Major effects of sensory experiences on the neocortical inhibitory circuits," *Journal of Neuroscience*, vol. 26, no. 34, pp. 8691–8701, 2006.
- [77] Y. Jiao, Z. Zhang, C. Zhang et al., "A key mechanism underlying sensory experience-dependent maturation of neocortical GABAergic circuits in vivo," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 29, pp. 12131–12136, 2011.
- [78] Q. Q. Sun, "Experience-dependent intrinsic plasticity in interneurons of barrel cortex layer IV," *Journal of Neurophysiology*, vol. 102, no. 5, pp. 2955–2973, 2009.
- [79] K. D. Micheva and C. Beaulieu, "An anatomical substrate for experience-dependent plasticity of the rat barrel field cortex," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 92, no. 25, pp. 11834–11838, 1995.
- [80] K. D. Micheva and C. Beaulieu, "Development and plasticity of the inhibitory neocortical circuitry with an emphasis on the rodent barrel field cortex: a review," *Canadian Journal of Physiology and Pharmacology*, vol. 75, no. 5, pp. 470–478, 1997.
- [81] T. Pizzorusso, P. Medini, N. Berardi, S. Chierzi, J. W. Fawcett, and L. Maffei, "Reactivation of ocular dominance plasticity in the adult visual cortex," *Science*, vol. 298, no. 5596, pp. 1248–1251, 2002.
- [82] M. S. Lazarus and Z. Josh Huang, "Distinct maturation profiles of perisomatic and dendritic targeting GABAergic interneurons in the mouse primary visual cortex during the critical period of ocular dominance plasticity," *Journal of Neurophysiology*, vol. 106, no. 2, pp. 775–787, 2011.
- [83] K. D. Micheva and C. Beaulieu, "Quantitative aspects of synaptogenesis in the rat barrel field cortex with special reference to GABA circuitry," *Journal of Comparative Neurology*, vol. 373, no. 3, pp. 340–354, 1996.
- [84] K. Fox and R. O. L. Wong, "A comparison of experience-dependent plasticity in the visual and somatosensory systems," *Neuron*, vol. 48, no. 3, pp. 465–477, 2005.
- [85] E. De Villers-Sidani, K. L. Simpson, Y. F. Lu, R. C. S. Lin, and M. M. Merzenich, "Manipulating critical period closure across different sectors of the primary auditory cortex," *Nature Neuroscience*, vol. 11, no. 8, pp. 957–965, 2008.
- [86] T. R. Barkat, D. B. Polley, and T. K. Hensch, "A critical period for auditory thalamocortical connectivity," *Nature Neuroscience*, vol. 14, pp. 1189–1194, 2011.
- [87] L. Frisen and M. Frisen, "How good is normal visual acuity? A study of letter acuity thresholds as a function of age," *Albrecht von Graefes Archiv fur Klinische und Experimentelle Ophthalmologie*, vol. 215, no. 3, pp. 149–157, 1980.
- [88] D. B. Elliott, K. C. H. Yang, and D. Whitaker, "Visual acuity changes throughout adulthood in normal, healthy eyes: seeing beyond 6/6," *Optometry and Vision Science*, vol. 72, no. 3, pp. 186–191, 1995.
- [89] T. Ferrer-Blasco, J. M. González-Méijome, and R. Montés-Micó, "Age-related changes in the human visual system and prevalence of refractive conditions in patients attending an eye clinic," *Journal of Cataract and Refractive Surgery*, vol. 34, no. 3, pp. 424–432, 2008.
- [90] K. Lehmann, K.-F. Schmidt, and S. Löwel, "Vision and visual plasticity in ageing mice," *Restorative Neurology and Neuroscience*, vol. 30, no. 2, pp. 161–178, 2012.

- [91] P. D. Spear, "Neural bases of visual deficits during aging," *Vision Research*, vol. 33, no. 18, pp. 2589–2609, 1993.
- [92] B. Godde, T. Berkefeld, M. David-Jürgens, and H. R. Dinse, "Age-related changes in primary somatosensory cortex of rats: evidence for parallel degenerative and plastic-adaptive processes," *Neuroscience and Biobehavioral Reviews*, vol. 26, no. 7, pp. 743–752, 2002.
- [93] M. T. Schmolesky, Y. Wang, M. Pu, and A. G. Leventhal, "Degradation of stimulus selectivity of visual cortical cells in senescent rhesus monkeys," *Nature Neuroscience*, vol. 3, no. 4, pp. 384–390, 2000.
- [94] A. G. Leventhal, Y. Wang, M. Pu, Y. Zhou, and Y. Ma, "GABA and its agonists improved visual cortical function in senescent monkeys," *Science*, vol. 300, no. 5620, pp. 812–815, 2003.
- [95] T. Hua, X. Li, L. He, Y. Zhou, Y. Wang, and A. G. Leventhal, "Functional degradation of visual cortical cells in old cats," *Neurobiology of Aging*, vol. 27, no. 1, pp. 155–162, 2006.
- [96] H. Wang, X. Xie, X. Li, B. Chen, and Y. Zhou, "Functional degradation of visual cortical cells in aged rats," *Brain Research*, vol. 1122, no. 1, pp. 93–98, 2006.
- [97] T. Hua, C. Kao, Q. Sun, X. Li, and Y. Zhou, "Decreased proportion of GABA neurons accompanies age-related degradation of neuronal function in cat striate cortex," *Brain Research Bulletin*, vol. 75, no. 1, pp. 119–125, 2008.
- [98] J. Bu, V. Sathyendra, N. Nagykeri, and C. Geula, "Age-related changes in calbindin-D28k, calretinin, and parvalbumin-immunoreactive neurons in the human cerebral cortex," *Experimental Neurology*, vol. 182, no. 1, pp. 220–231, 2003.
- [99] M. Pugliese, J. L. Carrasco, M. C. Geloso, J. Mascort, F. Michetti, and N. Mahy, " γ -aminobutyric acidergic interneuron vulnerability to aging in canine prefrontal cortex," *Journal of Neuroscience Research*, vol. 77, no. 6, pp. 913–920, 2004.
- [100] B. V. Atallah, W. Bruns, M. Carandini, and M. Scanziani, "Parvalbumin-expressing interneurons linearly transform cortical responses to visual stimuli," *Neuron*, vol. 73, no. 1, pp. 159–170, 2012.
- [101] R. Mao, J. Schummers, U. Knoblich et al., "Influence of a subtype of inhibitory interneuron on stimulus-specific responses in visual cortex," *Cerebral Cortex*, vol. 22, no. 3, pp. 493–508, 2012.
- [102] R. Miettinen, J. Sirvio, P. Riekkinen, M. P. Laakso, M. Riekkinen, and P. Riekkinen, "Neocortical, hippocampal and septal parvalbumin- and somatostatin-containing neurons in young and aged rats: correlation with passive avoidance and water maze performance," *Neuroscience*, vol. 53, no. 2, pp. 367–378, 1993.

Research Article

Development of Brainstem-Evoked Responses in Congenital Auditory Deprivation

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Received 16 January 2012; Accepted 17 April 2012

Academic Editor: Steve Lomber

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To compare the development of the auditory system in hearing and completely acoustically deprived animals, naive congenitally deaf white cats (CDCs) and hearing controls (HCs) were investigated at different developmental stages from birth till adulthood. The CDCs had no hearing experience before the acute experiment. In both groups of animals, responses to cochlear implant stimulation were acutely assessed. Electrically evoked auditory brainstem responses (E-ABRs) were recorded with monopolar stimulation at different current levels. CDCs demonstrated extensive development of E-ABRs, from first signs of responses at postnatal (p.n.) day 3 through appearance of all waves of brainstem response at day 8 p.n. to mature responses around day 90 p.n.. Wave I of E-ABRs could not be distinguished from the artifact in majority of CDCs, whereas in HCs, it was clearly separated from the stimulus artifact. Waves II, III, and IV demonstrated higher thresholds in CDCs, whereas this difference was not found for wave V. Amplitudes of wave III were significantly higher in HCs, whereas wave V amplitudes were significantly higher in CDCs. No differences in latencies were observed between the animal groups. These data demonstrate significant postnatal subcortical development in absence of hearing, and also divergent effects of deafness on early waves II–IV and wave V of the E-ABR.

1. Introduction

The auditory system demonstrates extensive developmental changes during postnatal life, both in humans as well as in altricial animals (review in [1]). Which developmental effects are caused by experience and which are preprogrammed by genetic makeup (independent of hearing experience; comp. [2, 3]) is not always straightforward.

An interesting model in resolving this question is the congenitally deaf cat (CDCs, [4]). CDCs show a cochlear degeneration before hearing onset that prevents hearing experience. However, the spiral ganglion cells are well preserved [5]. This is a substantial advantage in comparison to pharmacological deafening, where the numbers of surviving spiral ganglion cells decrease within short time down to less than 50%, in some down to 10% of the counts in hearing counterparts [6]. Loss of spiral ganglion cells may lead to a

down-regulation in supply of trophic factors to the cochlear nucleus (denervation effects). Loss of spiral ganglion cells is also a confounding factor for developmental studies, further aggravated by the interindividual variability in the ganglion cell loss. Electrical stimulation of the auditory nerve may thus yield a different extent of activation in the central auditory system in deafened animals. In contrast, in CDCs it is possible to test the functionality of the auditory system by electrical stimulation of the auditory nerve in a more reproducible way.

Extensive changes in the auditory cortex have been demonstrated in adult CDCs [7] as well as in neonatally-deafened cats [8]. Many of these changes are the consequence of an altered postnatal developmental sequence [9]. Consequently, auditory experience has a shaping influence on the functional maturation of the auditory system [10]. In addition, effects of auditory deprivation have been uncovered

in the synapses and function of the brainstem [11] and midbrain [12–15]. It remains unclear how these changes contribute to the cortical-sensitive period for therapy of deafness with cochlear implants [16, 17].

The present study was designed to investigate the consequences of inborn deafness on the brainstem function. In auditorily naive animals of different ages, brainstem-evoked signals were measured in response to electrical stimulation through a cochlear implant. The responses were compared to electrically evoked brainstem responses in hearing (acutely deafened) controls. Electrically-evoked auditory brainstem responses (E-ABRs) are also used clinically in human cochlear implanted subjects to objectively assess the auditory function [18–20]. The present results can, therefore, be directly compared to the measurements in cochlear-implanted subjects. Additionally, brainstem-evoked responses include components from different structures in the brainstem [21] and can be used to compare effects of deprivation on several auditory structures at the same time.

The present data reveal that auditory deprivation does affect the brainstem and that these functional effects can be detected using E-ABRs. The data demonstrate that a significant portion of the developmental process is, however, preserved in complete deafness. Consequently, this part of development is dependent on genetic makeup. Finally, the present data show that different portions of the afferent auditory pathway are differentially sensitive to deprivation.

2. Materials and Methods

2.1. Animals. For the present study, 35 cats were investigated. Their age was in the range from birth (P0) to adult (>6 months), 17 animals were congenitally deaf and 18 were normal hearing controls. All congenitally deaf cats had no hearing experience before the final (acute) experiment. Two animals from the deaf white cat colony were investigated at 0 p.n. (right after birth), one deaf animal at day 3 p.n. (P3) and one deaf animal at P8, remaining animals were older than 21 days. Congenital deafness was, however, verified in the 4th week p.n. by the absence of acoustically evoked brainstem responses (see [5]). In younger animals, the same test was performed at the beginning of the experiment, whereas in animals < P10 the test cannot yield certainty on the potential hearing status later in life, since hearing thresholds drop below 100 dB SPL only after P10 in hearing cats [22].

2.2. Experimental Procedure. All animals were anaesthetized by subcutaneous application of ketamine-hydrochloride (Ketavet, Parker-Davis; 15 mg/kg b.w.) and xylazine-hydrochloride (Rompun, Bayer; 0.6 mg/kg b.w.). The pinna was removed at both sides, the tympanic membranes and the bullae were exposed. The animals were placed in a soundproof chamber and fixated in a stereotactic frame. For acoustic stimulation, an inversely driven calibrated Bruel and Kjaer condenser microphone was placed in front of the tympanic membrane. For assessing hearing thresholds, condensation clicks (50 μ s) were used. Stimulation was performed at intensities in the range of 5–120 dB SPL,

presented at a rate of 13 stimuli per second. For recording of auditory brainstem responses (ABRs), a small trephination was drilled at the vertex of the skull and a recording electrode was placed epidurally. The indifferent electrode was inserted in the muscles under the bulla at the site of stimulation. The recorded signal was amplified by 80 dB (Tektronix V122 and 5A22N), filtered (0.01 kHz–10 kHz, 6 dB/octave, Tektronix 5A22N), and averaged by a computer (100 repetitions). Animals classified as hearing had a click-evoked ABR threshold <40 dB SPL.

After determining the hearing thresholds, both bullae and round windows were opened. In hearing controls the hair cells were destroyed using a slow instillation (within 5 minutes) of 0.3 mL neomycine sulfate (25 mg/mL) into the scala tympani. This procedure was performed to avoid electrical stimulation of hair cells (electrophonic effects). After further 5 minutes, the neomycine was carefully washed out by Ringer's solution and deafness was verified by the absence of auditory-evoked brainstem responses (condensation click, 120 dB SPL). The deafening procedure was performed on both ears. Thereafter, a custom-made cochlear implant with five intrasclerular gold contacts with a spacing of 1 mm [9] was placed in the scala tympani. The insertion depth was ~6 mm. Electrical stimulation with biphasic, charge-balanced pulses (200 μ s/phase, monopolar stimulation with the apicalmost electrode of the implant and an indifferent electrode placed in the muscles at the neck) was performed using an optically isolated current source. Presentation rate was 13 stimuli per second in all animals. Electrically evoked brainstem responses were recorded at varying intensities. Stimulus levels were computed from peak-to-peak amplitudes of the pulses. The highest current level was reached when facial nerve stimulation appeared. Recordings with facial nerve stimulation resulting in muscle contractions were contaminated with muscle activity and discarded from further processing.

2.3. Statistics. Individual waves of the electrically evoked brainstem responses (E-ABR) were designated *I-V* based on morphology and latency (Figure 1). Thresholds for each wave were determined as the current level at which the E-ABR curve showed a peak that appeared reproducibly at higher stimulation intensities at a similar latency. The amplitude of each wave was determined as peak-to-peak value between the maximum of each wave and the neighboring minimum of the E-ABR curve. The latency was determined as peak latency. Statistical comparisons of amplitudes and latencies were performed at 4 dB above thresholds.

Normality was tested using the Pearson-Stephens test ($\alpha = 10\%$), similarity of the distribution using the *f*-test ($\alpha = 10\%$) and final comparisons using *t*-tests (two-tailed tests $\alpha = 5\%$).

3. Results

In all investigated animals, the middle ear and the bulla were free of signs of infection. Deafness of all animals classified as

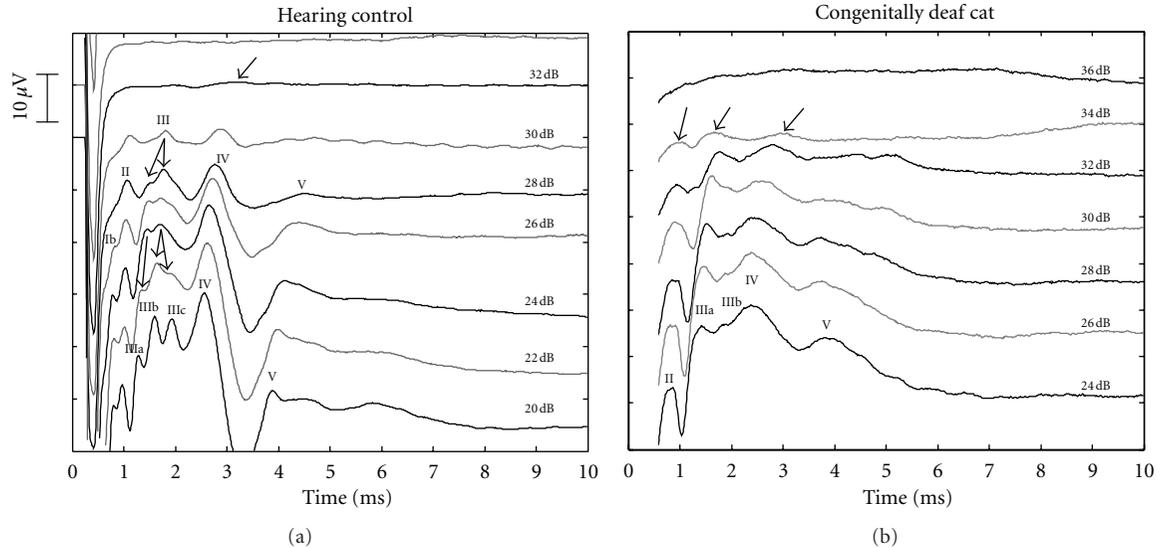


FIGURE 1: Examples of E-ABRs recorded from an adult hearing control (a) and an adult CDC (b). Arrows point to E-ABR components at threshold intensity. Current levels are given in dB attenuation (re 3 mApp). In general, similar morphology was observed, with some less-well differentiated waves in CDCs. Stimulus artifact (starting at 0 ms) was removed.

deaf in the hearing screening (at 4 weeks p.n.) was confirmed in the acute experiment.

Electrically evoked brainstem evoked responses (E-ABRs) in adult animals revealed a morphology characteristic of acoustically evoked brainstem response (Figure 1). In most animals, 4 waves could be consistently differentiated. Additional waves appeared at high stimulus levels. To avoid misinterpretation with cortical local field potentials, the waves were numbered by Arabic numerals. Morphology of the E-ABRs was similar between deaf and hearing animals. In deaf animals, the peak of wave I was frequently hidden in the artifact of the stimulus, whereas in hearing controls it was discernible in most animals (Figure 1(a)). The waves IV and V appeared less differentiated in deaf animals (Figure 1(b)), and the variability in the morphology of E-ABRs was higher in CDCs than in controls. Wave III has split into several subcomponents at high current levels in both groups of animals. Quantitative assessment was always performed at the earliest subcomponent of wave III (wave IIIa).

3.1. Age Group P0–P8. None of these four animals revealed any ABRs up to click levels of 120 dB SPL, even though the ear canals have been surgically removed, the tympanic membrane was exposed and the stimulation was performed using a closed system.

Individual fissures of the skull were not yet ossified in this group of animals. The external meati were closed. At P3 and P8, however, they already started to open from the round window side. Consequently, at P3 and P8, close to the tympanic membrane, the walls of the meatus did not adhere to each other anymore and the closed portion was few millimeters distant from the tympanic membrane. The middle ear was not pneumatized in this age group. The bulla was filled with a viscous whitish tissue that did not adhere to

the bulla walls nor to the round window (Figure 2). It could be easily removed to allow access to the round window for cochlear implantation. The round window membrane was not as clear as in adults. In the animal investigated at P8, the fluid was more translucent than in younger animals. At P8, the round window niche was free of the viscous fluid filling the remaining part of the bulla, that is, there was a small pneumatized space in the niche of the round window. In all older animals, the middle ear and bulla were fully pneumatized and all membranes were clear (Figure 2).

Stimulation with a cochlear implant allowed a controlled and comparable testing of animals despite a nonfunctional cochlea. Using monopolar configuration, the maximal portion of the auditory nerve was stimulated and positional effects were minimized.

Substantial maturation of E-ABRs was observed within the first days of life (Figure 3). At P0, E-ABR was not discernible in both investigated kitten. It was present but very small at P3 (Figure 3). Its amplitudes increased and latencies systematically decreased within the following weeks (Figure 3). Due to the atypical E-ABR morphology in very young animals, the wave assignment was not unequivocal at P3.

3.2. Comparison of Developing Animals. The range of suprathreshold intensities that were possible to test differed in individual animals due to muscular artifacts. These were generated by facial nerve stimulation at high current levels. Consequently, the dynamic range of E-ABRs that was possible to evaluate varied between 4 and >10 dB above threshold. To include all investigated animals, individual E-ABRs waves were compared at 4 dB above thresholds. Only positive peaks of the E-ABR components were processed.

With the exception of an increase in amplitudes between day 3 and 35 p.n. (Figure 3), the amplitudes showed

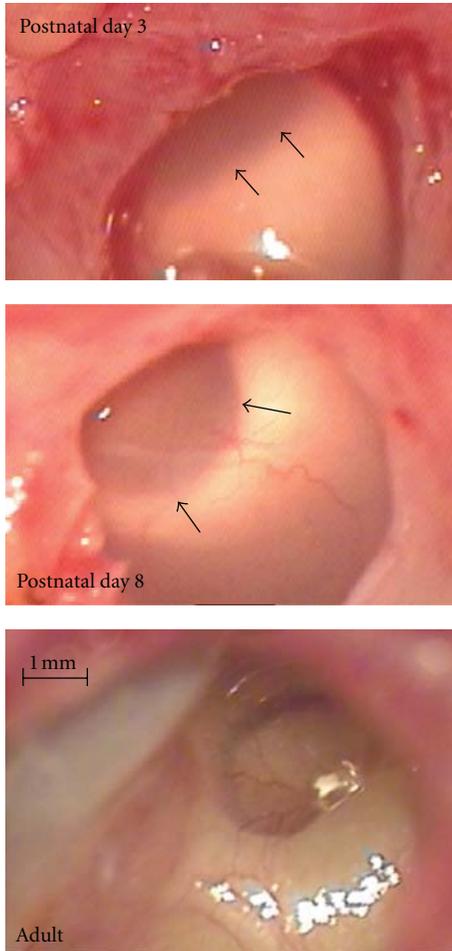


FIGURE 2: Photographs of the bulla in animals of the youngest group, compared to an adult animal. At postnatal days 0 and 3 (top), the bulla was filled with milky viscous tissue that covered the round window (its rims show through, marked by arrows). At postnatal day 8 (middle), the tissue was more translucent. In all older animals investigated, the bulla was fully pneumatized and did not show any further developmental changes (bottom).

a high degree of variability and no clear developmental trend. When analyzing the developmental pattern of latencies from postnatal day 8 on (at which for the first time wave components could unequivocally be compared in wavewise manner to adult animals), a decrease in latencies could be observed, most prominent within the first 3 months (Figure 4). For the purpose of comparing the developmental pattern, the data were pooled into two groups: from 0–3 months (comp. [23]) and above 4 months. When comparing such young and older animals within the hearing group, the latencies were significantly different for most waves of the E-ABR (wave I $P = 0.0489$; wave II $P = 0.00115$; wave III $P = 0.030$; wave IV $P = 0.012$; wave V $P = 0.021$; two-tailed t -test), demonstrating a developmental change. In deaf animals, the developmental change was similar, whereas for wave II, the differences did not reach the level of statistical significance (wave II $P = 0.084$; wave III $P = 0.044$; wave IV $P = 0.047$; wave V $P = 0.036$). In HCs and CDCs, the

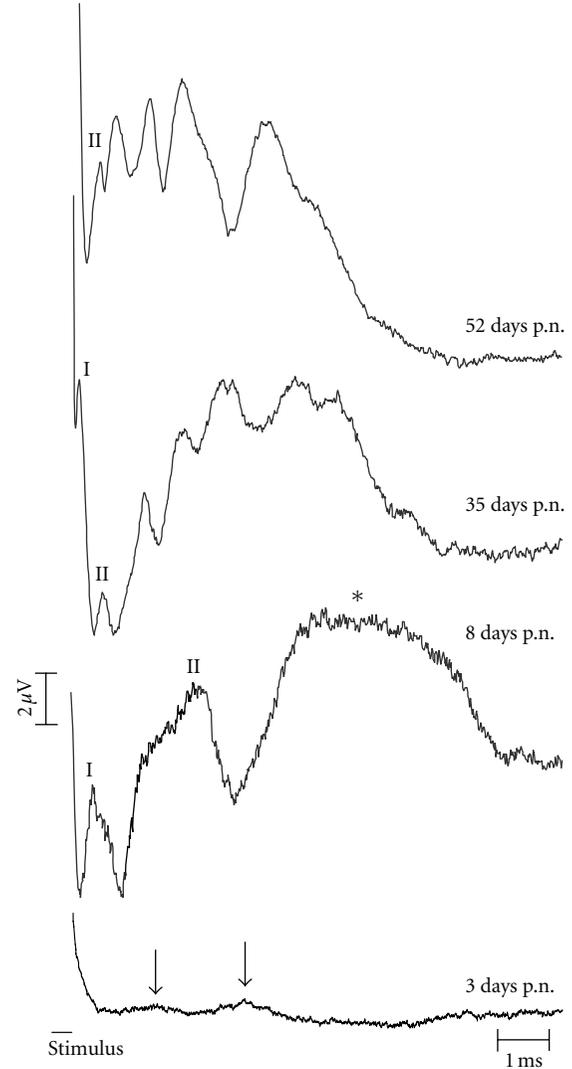


FIGURE 3: Development of E-ABRs in the first 52 days postnatally. All animals are from the white cat colony, but only after hearing onset (after day 10 p.n.) CDCs can be differentiated from white cats with residual hearing. Stimulus: biphasic pulse, $200 \mu\text{s}/\text{phase}$ at 10 dB above threshold. Stimulus duration is shown as a black bar, the artifact is removed from the recordings. At day 3 p.n., even at this high stimulation intensity the amplitudes of the waves were very small and only two waves could be differentiated from noise (marked by arrows). At day 8 p.n., all waves were observed (at the high intensity shown, waves III–V fused into a single large wave marked by the asterisk).

timecourse of development of brainstem evoked response latencies was similar (Figure 4).

3.3. Detailed Comparison of Matured Animals (>3 Months).

As no pronounced age-related changes were observed in waves II, III, and IV after the 3rd month (Figure 4), animals older than 3 months were pooled together and hearing controls were statistically compared to CDCs (8 congenitally deaf and 9 hearing controls).

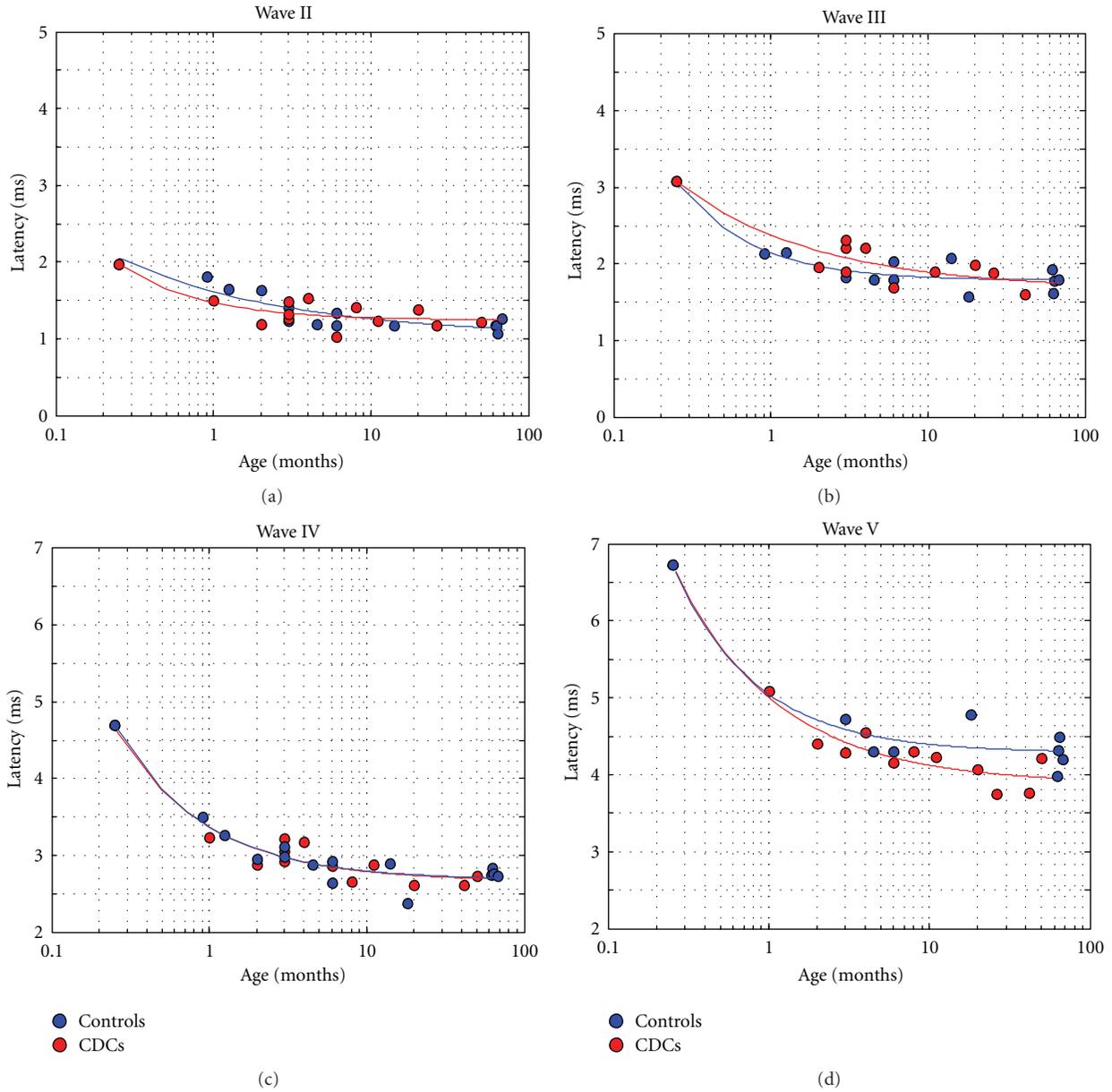


FIGURE 4: Development of wave latencies in hearing controls and CDCs, shown are peak latencies from all animals. Not in every animal all waves could be identified. The animal recorded at day 8 p.n. (before hearing onset) was considered a common starting point for the development in both groups. Systematic differences between animal groups were not observed, only in wave V had a tendency toward shorter latencies in CDCs.

3.4. *Thresholds.* Lowest E-ABR thresholds did not significantly differ between hearing and deaf animals if all waves were included ($186 \pm 52.6 \mu\text{A}$ in controls versus $180.8 \pm 61.8 \mu\text{A}$ in deaf). Already at threshold intensity, E-ABRs showed most frequently waves III/IV and V. However, in some animals, also (sometime only) earlier waves (typically II) or later waves (typically IV) appeared at threshold. To compare the E-ABRs in a wave-specific manner, the thresholds were also compared on the basis of “wave thresholds” (Figure 5). For wave V, the thresholds did not

differ between deaf and hearing animals (345 ± 201 versus $209 \pm 94 \mu\text{A}$, $P = 0.13$; Figure 5). On the other hand, the thresholds of waves II, III, and IV were significantly higher in CDCs (II: $202 \pm 26 \mu\text{A}$ in controls versus $223 \pm 97 \mu\text{A}$ in deaf, $P = 0.03$, two-tailed t -test; III: $225 \pm 15 \mu\text{A}$ in controls versus $275 \pm 110 \mu\text{A}$ in deaf, $P = 0.05$, two-tailed t -test; IV: $210 \pm 26 \mu\text{A}$ versus $251 \pm 94 \mu\text{A}$, $P = 0.03$, two-tailed t -test). This result indicates that potentially divergent deprivation effects take place in the generators of the early waves II–IV and the wave V. Additionally, deaf cats

consistently demonstrated higher interindividual variance in thresholds.

3.5. Amplitudes. Wave amplitudes were typically below $20\ \mu\text{V}$ and showed some interindividual variability (Figure 6). When comparing deaf and hearing animals at 4 dB above threshold, significant differences were observed for wave III ($4.85 \pm 2.7\ \mu\text{V}$ in controls versus $1.99 \pm 1.43\ \mu\text{V}$ in deaf; $P = 0.03$, two-tailed t -test) and wave V (1.43 ± 1.18 in controls versus 4.84 ± 2.82 in deaf; $P = 0.04$, two-tailed t -test). Also in this respect, a discrepant effect of deprivation was observed on wave III (smaller in CDCs) and wave V (larger in CDCs).

3.6. Latencies. Wave I was difficult to evaluate in deaf cats, as the recordings did not include this peak in sufficient number of animals. This was attributed to a shorter latency of this wave in deaf animals (by which the wave and the stimulus artifact coincided). Latencies of waves II–V at 4 dB above threshold were not significantly different between deaf and control animals in this age group (Figure 7). Also all interpeak latencies were not significantly different ($P = 0.9$ for interpeak intervals II–III, $P = 0.86$ for interpeak III–IV, $P = 0.17$ for interpeak IV–V, $P = 0.14$ for interpeak II–V).

4. Discussion

For the first time the present manuscript describes development of brainstem-evoked responses in inborn deafness. The developmental pattern of brainstem evoked responses with well-controlled auditory stimulation (through a cochlear implant) was compared between hearing controls and congenitally deaf cats. Using electrical stimulation it has been possible to investigate brainstem-evoked response development prior to hearing onset. At postnatal day 3, E-ABRs could be evoked, corresponding to “acoustic” studies with first responses at postnatal day 2–3 using an unphysiologically high sound pressure level of 140 dB [24].

The study demonstrates a developmental change in the brainstem evoked responses in both hearing and deaf animals. Thus, many functional developmental steps in the brainstem are set-in by genetic programs and do not require a functional cochlea. This stands in contrast to the findings in the auditory cortex, where the developmental pattern has been found extensively modified by deafness [7].

However, several effects of deprivation were observed in the brainstem responses. Qualitatively, the E-ABR appeared more smeared and were more variable in CDCs and indicated a desynchronization of the underlying neuronal activity in CDCs. Quantitatively, there was an increase in thresholds of early E-ABR waves and divergent effects of deafness on waves II, III, and V with respect to amplitudes and thresholds. Wave V has generators in the midbrain, whereas earlier waves are generated in the brainstem [21, 25, 26]. These results speak for a different response to deprivation in more peripheral and more central parts of the auditory pathway. Potentially, the wave V effects (higher

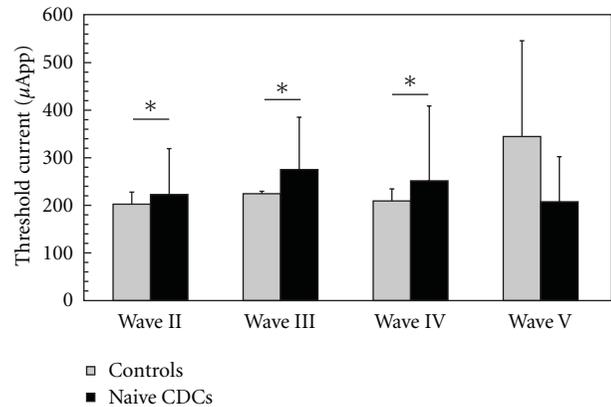


FIGURE 5: Comparisons of wave thresholds in matured animals (older than 3 months). Significantly larger thresholds were observed in CDCs for waves II, III, and IV. The trend in wave V is in opposite direction (two-tailed t -test at $\alpha = 5\%$).

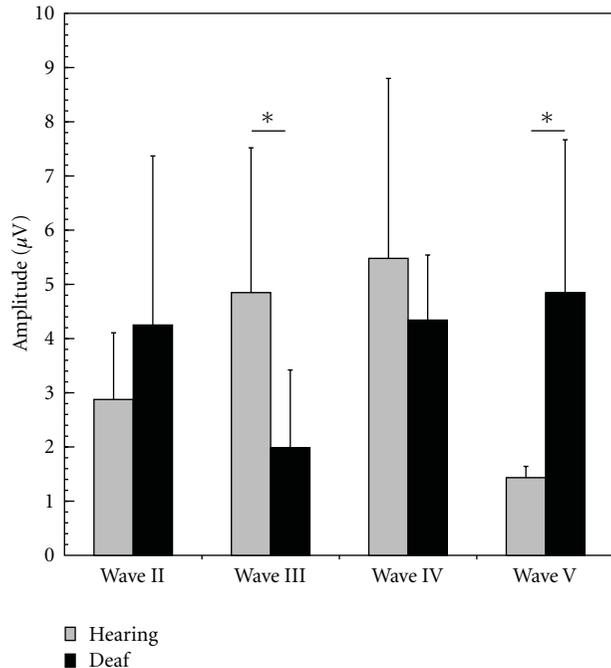


FIGURE 6: Wave mean peak amplitudes in matured animals (older than 3 months). In CDCs, wave III had lower and wave V higher amplitude (two-tailed t -test at $\alpha = 5\%$).

amplitude and lower threshold in deaf animals) correspond to compensation of the missing input by downregulated inhibition and upregulated excitatory transmission, as described in the inferior colliculus [27] and auditory cortex of deaf or deafened animals [9, 28].

Considering that brainstem-evoked responses are not capable reflecting all details of the differences in synaptic and neuronal function between HCs and CDCs, the effects of deafness on waves II–IV are well in agreement with studies demonstrating synaptic reorganization in the cochlear nucleus following congenital deafness [11, 29–35], as well as

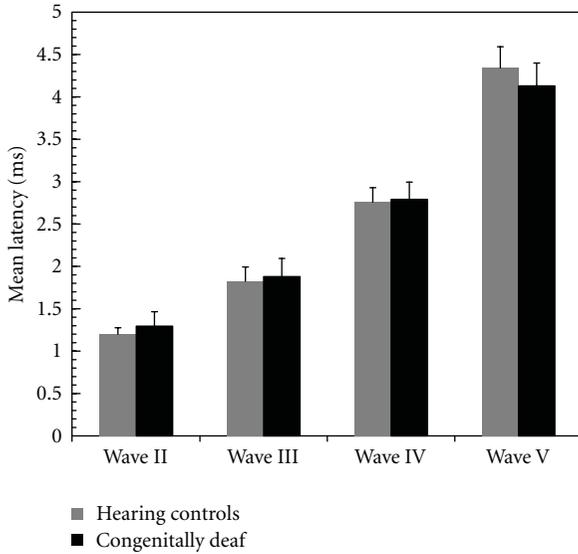


FIGURE 7: Mean peak latencies of all waves in matured animals (older than 3 months). No statistically significant differences were observed between CDCs and hearing controls.

in the more central parts of the auditory system (reviewed in [7]).

4.1. Methodological Discussion. Several methodological factors could have influenced the present results. First of all, amplitudes of brainstem responses are known to have a higher variability than latencies (reviewed in [36]). This is the consequence of the fast temporal order of the components, causing them to partly overlap. In acoustic stimulation, some authors filtered out the low-frequency component to facilitate the quantification [37]. We decided against this procedure due to the presence of the electrical stimulus artifact that complicated the offline filtering. Nonetheless, statistically significant results were obtained also for amplitudes. Therefore, we considered our amplitude quantification as sufficiently robust.

The threshold of E-ABRs depends on the position of the cochlear implant within the scala tympani. Positioning the stimulation electrodes closer to the modiolus can decrease E-ABR thresholds by 6 dB [38]. The position of the electrode within the scala tympani cannot be controlled by the surgeon and could act as a confounding factor. Nonetheless, a systematic bias between the investigated age groups is highly unlikely, given that the lowest E-ABR threshold was the same in both groups. The effect of electrode position was further minimized by using a monopolar stimulation configuration.

The number of surviving spiral ganglion cells may significantly influence threshold currents [39]. Nonetheless, in contrast to neonatally deafened animals, the congenitally deaf cats do not suffer from a pronounced degeneration of the spiral ganglion cells in the basalmost halfturn of the cochlea and the ages investigated here [5, 6]. It is the basalmost halfturn of the cat cochlea where a cochlear implant can be inserted [40]. Therefore, a systematic effect

of the spiral ganglion cell loss on the present results is not probable.

Finally, the repetition rate could have influenced the results, especially in the youngest animals. To avoid confounding the results by different repetition rates in different age groups, we decided to select a constant low repetition rate (13 Hz), whereas the brainstem response in adults can be reliably recorded up to the repetition rates of 70 Hz [41]. Even in animals at P3, we could record a brainstem evoked response. In consequence, the selected rate appears to be a good compromise. Cortical responses at P8 have latencies of ~ 50 ms [7], thus this repetition rate (with interstimulus intervals of ~ 77 ms) should allow a sufficient activation of the entire afferent auditory system. Nonetheless, the repetition rate needs to be taken into consideration as a factor lowering the amplitudes of E-ABRs in youngest animals (day 0, 3 and 8 p.n.).

4.2. E-ABR Generators. Electrically evoked brainstem responses are similar to acoustically evoked brainstem responses [42–45]. Consequently, one can assume the same generators of both types of signals. Here, we used a terminology of brainstem response waves that rests on the terminology used with acoustic stimulation of the cat auditory system. We assume that the waves found in electrically evoked brainstem responses correspond to the ones in acoustically evoked brainstem responses, as the morphology and the latency range (considering the absence of acoustic transduction in the inner ear) correspond well. Interestingly, in hearing controls wave III reproducibly broke up into 3 waves with increasing electrical intensity. Similarly, with high-intensity acoustic stimulation, wave III sometimes splits into 2 components [37]. This finding supports the theory that wave III is caused by activation of up to 3 different structures in the brainstem [21], and that acoustical and electrical stimulation result in ABRs with similar generators and morphology of responses. Possibly, the “hypersynchronization” of the electrically evoked activity compared to acoustic stimulation [46] additionally allows to better reveal these tree generators. Also, in CDCs, a similar effect on wave III has been observed, although wave III broke up only into 2 well-differentiated components. The observation of fewer subcomponents of wave III in CDCs further supports a desynchronization of the underlying neuronal activity in deafness. All shown quantitative comparisons were consequently performed on the component of wave III that appeared with shortest latency (wave IIIa).

4.3. Development of Auditory Pathway. Development of brainstem responses to electrical stimulation has not been systematically investigated yet. On the other hand, development of brainstem-evoked responses to acoustical stimulation has been investigated in great detail in hearing cats (comp. [23, 24, 47, 48]). When latencies are plotted as a function of age at same sensation levels in these acoustically stimulated animals, the data correspond well to the present study: also there the main development took

place in the first 90 days [23]. In hearing, acoustically stimulated animals, exponential decays similar to the present study have been observed (*ibid.*, comp. Figure 4). The electrically evoked responses in the present study had slightly shorter latencies when compared to the acoustical of the previous studies, explicable by by-passing the hair cell-primary afferent synapse with cochlear implant stimulation.

The present experiments provide further support for the ABR hearing screening procedure performed on the deaf white cats [5]: none of the animals classified as deaf showed any signs of hearing in the acute experiments. Additionally, even though electrically evoked responses could be demonstrated at P3, no signs of hearing were observed in the animals tested between P3 and P8, even after surgical opening of the closed outer ear canal. Additionally, performing the E-ABRs in the same animals, we confirmed that the central auditory system beyond the cochlea was functional. Consequently, absence of acoustical responses was solely due to lack of cochlear function. From P3 on, morphological degeneration of the organ of Corti in CDCs is quickly progressing, with loss of cochlear microphonics and hair cells [4, 49].

Electrically evoked brainstem responses can be recorded even before acoustically evoked responses with physiological sound pressure levels. This demonstrates that the afferent auditory pathway (at least in part) is already functional before significant hearing is possible. Additionally, it shows that the general pattern in the afferent auditory pathway develops in absence of auditory experience (see also [5, 45, 50]).

4.4. Wave I in CDCs. Wave I corresponds to compound action potentials of the auditory nerve [51]. In the present experiments, wave I was not consistently observed in CDCs. Thus, one can speculate that the action potentials in the auditory nerve were generated less synchronously in CDCs, possibly at a more central site when compared to controls. The reason for such a shift in action potential generation site could be a change in the physiological properties of primary afferents in deaf cats, possibly by demyelination of the most peripheral portions of the primary afferents [52, 53]. As the highest probability for action potential generation is at the nodes of Ranvier, and the demyelination is connected with changed distribution of fast voltage-sensitive sodium channels along the primary afferents, the demyelination could lead to a shift of the action potential generation site to more central nodes of Ranvier ([52, 53]; compare also discussion in [40]). That would lead to shorter wave I latencies. However, the absence of wave I in the present experiments could have also been caused by a higher asynchrony of action potential generation in the auditory nerve by differentially shifting the action potential generation site along the auditory nerve in different fibers. The later waves did not show a systematic latency difference between controls and CDCs, favoring the asynchrony hypothesis or indicating that some additional deprivation-induced delay has to take place in CDCs between wave I and wave II.

4.5. Matured Animals. The present results on matured animals are in accordance with data on neonatally pharmacologically deafened adult cats. Pharmacologically deafened adult animals show smaller E-ABR amplitudes [31]: this was also the case in the present animals, although it was only statistically significant for wave III. A correlation between the amplitudes of E-ABRs and the number of spiral ganglion cells was demonstrated [51], although for later waves of the E-ABR, this correlation was weaker [42, 54, 55]. Nonetheless, significant correlation of wave IV amplitude with spiral ganglion cells has also been shown [56]. A more pronounced denervation effects on the development or degeneration of the brainstem in the neonatally deafened cats as a consequence of the loss of spiral ganglion cells early in postnatal life could be one reason why the difference in amplitudes was more pronounced in adult neonatally deafened cats when compared to CDCs from the present study.

Another mechanism for smaller amplitudes of the E-ABRs in deaf cats could be a loss of neurons in the cochlear nucleus of deprived animals. This was shown in rodents when the neuronal activity was completely silenced before hearing onset [57, 58]. To date, there is no systematic study on the functional properties of “deaf” auditory nerve fibers in CDCs. A report on six animals from a deaf white cat colony with some residual hearing showed severely reduced spontaneous activity in auditory nerve fibers (assessed with intracellular recordings) of animals with high hearing thresholds [59]. However, despite a reduction in the total nuclear volume within the auditory brainstem [29–31], no evidence for a decrease in neuronal counts has been reported, neither for CDCs nor for neonatally deafened cats [5, 31].

Numerous studies described morphological changes in the synapses of the brainstem in CDCs [11, 32–35]. It is probable that the decrease in amplitude of wave III is due to weakening of synaptic activity and desynchronization rather than anatomical loss of central auditory neurons.

Delayed or incomplete myelination in CDCs may be a further reason for smaller E-ABR amplitudes in CDCs. However, the absence of differences in interpeak latencies between CDCs and HCs argue against a delayed transmission along the auditory brainstem and thus also against this interpretation. Further studies are required to eventually identify the mechanism behind smaller E-ABR amplitudes in CDCs.

There was an increase in individual wave II–IV thresholds in CDCs. This was also observed in adult neonatally deafened animals [31]. Increase in thresholds of individual E-ABR waves can be explained by weakening of synaptic efficacies along the auditory pathway [11, 32, 34, 35, 60] or by desynchronization of their activity. Deafness-induced increase in jitter of action potentials in inferior colliculus has been shown in neonatally deafened animals [15]. Nonetheless, the individual contribution of these mechanisms cannot be assessed solely from brainstem-evoked responses.

4.6. Human ABRs. Development of human brainstem-evoked responses has been also described in great detail in hearing children (review in [36]). Brainstem-evoked

responses represent a valuable tool for exploring the function of the afferent auditory pathway in deafness, with remarkably corresponding data between cochlear-implanted humans and animals. Brainstem responses of cats and humans, after appropriate normalization, show similar developmental rates for the early waves, whereas humans show a slower development for the late waves (wave V) [61]. As in the present study, also in brainstem-evoked responses of humans, a similar development has been observed in hearing children and in deaf, cochlear-implanted children [62]. In consequence, also in deaf children the brainstem-evoked responses develop comparably to hearing peers and do not require cochlear input.

Correspondingly, no sensitive periods in the development of brainstem response of humans have been observed with chronic electrical stimulation (cochlear implant use, [62, 63]). These are most likely determined by the parts of the auditory system beyond the brainstem and midbrain ([62]; comp. also [64]). The most probable candidate, strongly interacting with all levels of the auditory system, is the auditory cortex, where developmental sensitive periods have been observed both in CDCs as well as in cochlear-implanted humans (reviewed in [7]). In combination, this evidence indicates that sensitive periods for cochlear implantation [63, 65] are determined by the auditory cortex.

5. Conclusions

Electrically evoked brainstem responses show postnatal development even in complete absence of hearing experience. Deprivation-induced effects include reductions of wave III amplitude, increase of wave V amplitude, and increases of wave thresholds. No effects of deafness on E-ABR latencies were found. The results indicate desynchronization and/or weakening of synaptic activity in auditory brainstem and some additional compensatory “hypersensitivity” in the midbrain of deaf animals.

Acknowledgment

The work was supported by Deutsche Forschungsgemeinschaft (DFG Kr 3370).

References

- [1] A. Kral, “Unimodal and cross-modal plasticity in the “deaf” auditory cortex,” *International Journal of Audiology*, vol. 46, no. 9, pp. 479–493, 2007.
- [2] C. Kapfer, A. H. Seidl, H. Schweizer, and B. Grothe, “Experience-dependent refinement of inhibitory inputs to auditory coincidence-detector neurons,” *Nature Neuroscience*, vol. 5, no. 3, pp. 247–253, 2002.
- [3] A. Brand, O. Behrend, T. Marquardt, T. McAlpine, and B. Grothe, “Precise inhibition is essential for microsecond interaural time difference coding,” *Nature*, vol. 417, no. 6888, pp. 543–547, 2002.
- [4] I. W. S. Mair, “Hereditary deafness in the white cat,” *Acta Otolaryngologica. Supplementum*, vol. 314, pp. 1–48, 1973.
- [5] S. Heid, R. Hartmann, and R. Klinke, “A model for prelingual deafness, the congenitally deaf white cat—population statistics and degenerative changes,” *Hearing Research*, vol. 115, no. 1–2, pp. 101–112, 1998.
- [6] P. A. Leake, G. T. Hradek, and R. L. Snyder, “Chronic electrical stimulation by a cochlear implant promotes survival of spiral ganglion neurons after neonatal deafness,” *Journal of Comparative Neurology*, vol. 412, no. 4, pp. 543–562, 1999.
- [7] A. Kral and A. Sharma, “Developmental neuroplasticity after cochlear implantation,” *Trends in Neurosciences*, vol. 35, no. 2, pp. 111–122, 2011.
- [8] J. B. Fallon, D. R. F. Irvine, and R. K. Shepherd, “Cochlear implant use following neonatal deafness influences the cochleotopic organization of the primary auditory cortex in cats,” *Journal of Comparative Neurology*, vol. 512, no. 1, pp. 101–114, 2009.
- [9] A. Kral, J. Tillein, S. Heid, R. Hartmann, and R. Klinke, “Postnatal cortical development in congenital auditory deprivation,” *Cerebral Cortex*, vol. 15, no. 5, pp. 552–562, 2005.
- [10] R. Klinke, A. Kral, S. Heid, J. Tillein, and R. Hartmann, “Recruitment of the auditory cortex in congenitally deaf cats by long-term cochlear electrostimulation,” *Science*, vol. 285, no. 5434, pp. 1729–1733, 1999.
- [11] D. K. Ryugo, E. A. Kretzmer, and J. K. Niparko, “Restoration of auditory nerve synapses in cats by cochlear implants,” *Science*, vol. 310, no. 5753, pp. 1490–1492, 2005.
- [12] R. L. Snyder, S. J. Rebscher, K. L. Cao, P. A. Leake, and K. Kelly, “Chronic intracochlear electrical stimulation in the neonatally deafened cat. I: expansion of central representation,” *Hearing Research*, vol. 50, no. 1–2, pp. 7–33, 1990.
- [13] R. L. Snyder, S. J. Rebscher, P. A. Leake, K. Kelly, and K. Cao, “Chronic intracochlear electrical stimulation in the neonatally deafened cat. II: temporal properties of neurons in the inferior colliculus,” *Hearing Research*, vol. 56, no. 1–2, pp. 246–264, 1991.
- [14] N. A. Hardie, A. Martsi-McClintock, L. M. Aitkin, and R. K. Shepherd, “Neonatal sensorineural hearing loss affects synaptic density in the auditory midbrain,” *NeuroReport*, vol. 9, no. 9, pp. 2019–2022, 1998.
- [15] R. K. Shepherd, J. H. Baxi, and N. A. Hardie, “Response of inferior colliculus neurons to electrical stimulation of the auditory nerve in neonatally deafened cats,” *Journal of Neurophysiology*, vol. 82, no. 3, pp. 1363–1380, 1999.
- [16] A. Kral, R. Hartmann, J. Tillein, S. Heid, and R. Klinke, “Hearing after congenital deafness: central auditory plasticity and sensory deprivation,” *Cerebral Cortex*, vol. 12, no. 8, pp. 797–807, 2002.
- [17] A. Sharma, M. F. Dorman, and A. Kral, “The influence of a sensitive period on central auditory development in children with unilateral and bilateral cochlear implants,” *Hearing Research*, vol. 203, no. 1–2, pp. 134–143, 2005.
- [18] S. Gallégo, B. Frachet, C. Micheyl, E. Truy, and L. Collet, “Cochlear implant performance and electrically-evoked auditory brain-stem response characteristics,” *Electroencephalography and Clinical Neurophysiology*, vol. 108, no. 6, pp. 521–525, 1998.
- [19] M. Sparreboom, A. J. Beynon, A. F. M. Snik, and E. A. Mylanus, “Electrically evoked auditory brainstem responses in children with sequential bilateral cochlear implants,” *Otology and Neurotology*, vol. 31, no. 7, pp. 1055–1061, 2010.
- [20] K. A. Gordon, J. Valero, S. F. Jewell, J. Ahn, and B. C. Papsin, “Auditory development in the absence of hearing in infancy,” *NeuroReport*, vol. 21, no. 3, pp. 163–167, 2010.

- [21] J. R. Melcher and N. Y. Kiang, "Generators of the brainstem auditory evoked potential in cat III: identified cell populations," *Hearing Research*, vol. 93, no. 1-2, pp. 52-71, 1996.
- [22] Brugge, "Development of the Lower Auditory Brainstem of the Cat," in *Development of Auditory and Vestibular Systems 2*, Romand, Ed., pp. 273-296, Elsevier, New York, NY, USA, 1992.
- [23] E. J. Walsh, J. McGee, and E. Javel, "Development of auditory-evoked potentials in the cat. II. Wave latencies," *Journal of the Acoustical Society of America*, vol. 79, no. 3, pp. 725-744, 1986.
- [24] E. J. Walsh, J. McGee, and E. Javel, "Development of auditory-evoked potentials in the cat. I. Onset of response and development of sensitivity," *Journal of the Acoustical Society of America*, vol. 79, no. 3, pp. 712-724, 1986.
- [25] D. M. Caird and R. Klinke, "The effect of inferior colliculus lesions on auditory evoked potentials," *Electroencephalography and Clinical Neurophysiology*, vol. 68, no. 3, pp. 237-240, 1987.
- [26] D. Caird, D. Sontheimer, and R. Klinke, "Intra- and extracranially recorded auditory evoked potentials in the cat. I. Source location and binaural interaction," *Electroencephalography and Clinical Neurophysiology*, vol. 61, no. 1, pp. 50-60, 1985.
- [27] S. C. Bledsoe, S. Nagase, J. M. Miller, and R. A. Altschuler, "Deafness-induced plasticity in the mature central auditory system," *NeuroReport*, vol. 7, no. 1, pp. 225-229, 1995.
- [28] V. C. Kotak, S. Fujisawa, F. A. Lee, O. Karthikeyan, C. Aoki, and D. H. Sanes, "Hearing loss raises excitability in the auditory cortex," *Journal of Neuroscience*, vol. 25, no. 15, pp. 3908-3918, 2005.
- [29] M. Hultcrantz, R. Snyder, S. Rebscher, and P. Leake, "Effects of neonatal deafening and chronic intracochlear electrical stimulation on the cochlear nucleus in cats," *Hearing Research*, vol. 54, no. 2, pp. 272-280, 1991.
- [30] S. Heid, *Morphologische Befunde am peripheren und zentralen auditorischen System der kongenital gehörlosen weißen Katze*, Ph.D. Thesis, Frankfurt Am Main, 1998.
- [31] N. A. Hardie and R. K. Shepherd, "Sensorineural hearing loss during development: morphological and physiological response of the cochlea and auditory brainstem," *Hearing Research*, vol. 128, no. 1-2, pp. 147-165, 1999.
- [32] S. A. Larsen and T. M. Kirchhoff, "Anatomical evidence of synaptic plasticity in the cochlear nuclei of white-deaf cats," *Experimental Neurology*, vol. 115, no. 1, pp. 151-157, 1992.
- [33] E. E. Redd, H. B. Cahill, T. Pongstaporn, and D. K. Ryugo, "The effects of congenital deafness on auditory nerve synapses: type I and type II multipolar cells in the anteroventral cochlear nucleus of cats," *Journal of the Association for Research in Otolaryngology*, vol. 3, no. 4, pp. 403-417, 2002.
- [34] D. K. Ryugo, C. A. Baker, K. L. Montey et al., "Synaptic plasticity after chemical deafening and electrical stimulation of the auditory nerve in cats," *Journal of Comparative Neurology*, vol. 518, no. 7, pp. 1046-1063, 2010.
- [35] C. A. Baker, K. L. Montey, T. Pongstaporn, and D. K. Ryugo, "Postnatal development of the endbulb of held in congenitally deaf cats," *Frontiers in Neuroanatomy*, vol. 4, article 19, 2010.
- [36] A. Salamy, "Maturation of the auditory brainstem response from birth through early childhood," *Journal of Clinical Neurophysiology*, vol. 1, no. 3, pp. 293-329, 1984.
- [37] B. C. Fullerton and N. Y. Kiang, "The effect of brainstem lesions on brainstem auditory evoked potentials in the cat," *Hearing Research*, vol. 49, no. 1-3, pp. 363-390, 1990.
- [38] R. K. Shepherd, S. Hatsushika, and G. M. Clark, "Electrical stimulation of the auditory nerve: the effect of electrode position on neural excitation," *Hearing Research*, vol. 66, no. 1, pp. 108-120, 1993.
- [39] C. A. Miller, P. J. Abbas, and B. K. Robinson, "The use of long-duration current pulses to assess nerve survival," *Hearing Research*, vol. 78, no. 1, pp. 11-26, 1994.
- [40] A. Kral, R. Hartmann, D. Mortazavi, and R. Klinke, "Spatial resolution of cochlear implants: the electrical field and excitation of auditory afferents," *Hearing Research*, vol. 121, no. 1-2, pp. 11-28, 1998.
- [41] B. C. Fullerton, R. A. Levine, H. L. Hosford-Dunn, and N. Y. Kiang, "Comparison of cat and human brain-stem auditory evoked potentials," *Electroencephalography and Clinical Neurophysiology*, vol. 66, no. 6, pp. 547-570, 1987.
- [42] R. K. Shepherd, G. M. Clark, and R. C. Black, "Chronic electrical stimulation of the auditory nerve in cats. Physiological and histopathological results," *Acta Oto-Laryngologica*, vol. 96, no. 399, pp. 19-31, 1983.
- [43] C. A. Miller, P. J. Abbas, and C. J. Brown, "Electrically evoked auditory brainstem response to stimulation of different sites in the cochlea," *Hearing Research*, vol. 66, no. 2, pp. 130-142, 1993.
- [44] R. K. Shepherd and E. Javel, "Electrical stimulation of the auditory nerve. I. Correlation of physiological responses with cochlear status," *Hearing Research*, vol. 108, no. 1-2, pp. 112-144, 1997.
- [45] R. Hartmann, R. K. Shepherd, S. Heid, and R. Klinke, "Response of the primary auditory cortex to electrical stimulation of the auditory nerve in the congenitally deaf white cat," *Hearing Research*, vol. 112, no. 1-2, pp. 115-133, 1997.
- [46] R. Hartmann and A. Kral, "Central responses to electrical stimulation," in *Cochlear Implants. Auditory Prostheses and Electric Hearing*, F. G. Zeng, A. N. Popper, and R. R. Fay, Eds., pp. 213-285, Springer, New York, NY, USA, 2004.
- [47] S. Heid, T. K. Jahn-Siebert, R. Klinke, R. Hartmann, and G. Langner, "Afferent projection patterns in the auditory brainstem in normal and congenitally deaf white cats," *Hearing Research*, vol. 110, no. 1-2, pp. 191-199, 1997.
- [48] E. J. Walsh, J. McGee, and E. Javel, "Development of auditory-evoked potentials in the cat. III. Wave amplitudes," *Journal of the Acoustical Society of America*, vol. 79, no. 3, pp. 745-754, 1986.
- [49] I. W. Mair and H. H. Elverland, "Hereditary deafness in the cat. An electron microscopic study of the stria vascularis and Reissner's membrane," *Archives of Oto-Rhino-Laryngology*, vol. 217, no. 2, pp. 199-217, 1977.
- [50] S. Heid, T. K. Jahn-Siebert, R. Klinke, R. Hartmann, and G. Langner, "Afferent projection patterns in the auditory brainstem in normal and congenitally deaf white cats," *Hearing Research*, vol. 110, no. 1-2, pp. 191-199, 1997.
- [51] R. D. Hall, "Estimation of surviving spiral ganglion cells in the deaf rat using the electrically evoked auditory brainstem response," *Hearing Research*, vol. 45, no. 1-2, pp. 123-136, 1990.
- [52] H. Bostock, T. A. Sears, and R. M. Sherratt, "The spatial distribution of excitability and membrane current in normal and demyelinated mammalian nerve fibres," *Journal of Physiology*, vol. 341, pp. 41-58, 1983.
- [53] R. Zhou, P. J. Abbas, and J. G. Assouline, "Electrically evoked auditory brainstem response in peripherally myelin-deficient mice," *Hearing Research*, vol. 88, no. 1-2, pp. 98-106, 1995.
- [54] J. M. Miller, L. G. Duckert, M. A. Malone, and B. E. Pflugst, "Cochlear prostheses: stimulation-induced damage," *Annals of*

- Otology, Rhinology and Laryngology*, vol. 92, no. 6, part 1, pp. 599–609, 1983.
- [55] P. H. Stypulkowski, C. van den Honert, and S. D. Kvisstad, “Electrophysiologic evaluation of the cochlear implant patient,” *Otolaryngologic Clinics of North America*, vol. 19, no. 2, pp. 249–257, 1986.
- [56] L. Smith and F. B. Simmons, “Estimating eighth nerve survival by electrical stimulation,” *Annals of Otology, Rhinology and Laryngology*, vol. 92, no. 1, part 1, pp. 19–23, 1983.
- [57] T. S. Tierney, F. A. Russell, and D. R. Moore, “Susceptibility of developing cochlear nucleus neurons to deafferentation-induced death abruptly ends just before the onset of hearing,” *Journal of Comparative Neurology*, vol. 378, no. 2, pp. 295–306, 1997.
- [58] T. S. Tierney and D. R. Moore, “Naturally occurring neuron death during postnatal development of the gerbil ventral cochlear nucleus begins at the onset of hearing,” *Journal of Comparative Neurology*, vol. 387, no. 3, pp. 421–429, 1997.
- [59] D. K. Ryugo, B. T. Rosenbaum, P. J. Kim, J. K. Niparko, and A. A. Saada, “Single unit recordings in the auditory nerve of congenitally deaf white cats: morphological correlates in the cochlea and cochlear nucleus,” *Journal of Comparative Neurology*, vol. 397, no. 4, pp. 532–548, 1998.
- [60] A. A. Saada, J. K. Niparko, and D. K. Ryugo, “Morphological changes in the cochlear nucleus of congenitally deaf white cats,” *Brain Research*, vol. 736, no. 1-2, pp. 315–328, 1996.
- [61] E. J. Walsh, M. Gorga, and J. McGee, “Comparisons of the development of auditory brainstem response latencies between cats and humans,” *Hearing Research*, vol. 60, no. 1, pp. 53–63, 1992.
- [62] K. A. Gordon, B. C. Papsin, and R. V. Harrison, “An evoked potential study of the developmental time course of the auditory nerve and brainstem in children using cochlear implants,” *Audiology and Neurotology*, vol. 11, no. 1, pp. 7–23, 2006.
- [63] J. K. Niparko, E. A. Tobey, D. J. Thal et al., “Spoken language development in children following cochlear implantation,” *Journal of the American Medical Association*, vol. 303, no. 15, pp. 1498–1506, 2010.
- [64] C. M. Moore, M. Vollmer, P. A. Leake, R. L. Snyder, and S. J. Rebscher, “The effects of chronic intracochlear electrical stimulation on inferior colliculus spatial representation in adult deafened cats,” *Hearing Research*, vol. 164, no. 1-2, pp. 82–96, 2002.
- [65] A. Kral and G. M. O’Donoghue, “Profound deafness in childhood,” *The New England Journal of Medicine*, vol. 363, no. 15, pp. 1438–1450, 2010.

Review Article

The Corpus Callosum and the Visual Cortex: Plasticity Is a Game for Two

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Received 16 December 2011; Accepted 19 April 2012

Academic Editor: Steve Lomber

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Throughout life, experience shapes and selects the most appropriate brain functional connectivity to adapt to a changing environment. An ideal system to study experience-dependent plasticity is the visual cortex, because visual experience can be easily manipulated. In this paper, we focus on the role of interhemispheric, transcallosal projections in experience-dependent plasticity of the visual cortex. We review data showing that deprivation of sensory experience can modify the morphology of callosal fibres, thus altering the communication between the two hemispheres. More importantly, manipulation of callosal input activity during an early critical period alters developmental maturation of functional properties in visual cortex and modifies its ability to remodel in response to experience. We also discuss recent data in rat visual cortex, demonstrating that the corpus callosum plays a role in binocularity of cortical neurons and is involved in the plastic shift of eye preference that follows a period of monocular eyelid suture (monocular deprivation) in early age. Thus, experience can modify the fine connectivity of the corpus callosum, and callosal connections represent a major pathway through which experience can mediate functional maturation and plastic rearrangements in the visual cortex.

1. Introduction

From playing the piano to riding a bike, interhemispheric communication is a crucial tool that our brain uses to perform a variety of everyday actions, from very simple to complex behaviours. One of the most important pathways through which this communication is achieved is the Corpus Callosum, the major fiber bundle in the brain [1–3]. In humans it contains about 170–190 millions of fibers that interconnect homologous cortical areas in the two hemispheres, as estimated by a pioneer work on human brains by Tomasch in 1954 [4].

A number of experiments and observations suggest that the two hemispheres could inhibit each other via the callosal pathway, to achieve the segregation of lateralized functions such as language or face recognition [3, 5]. However, other experiments suggest that the callosal pathway provides an excitatory input to the opposite hemisphere that enables cortical integration [5, 6].

In vision, our main sensory system, the corpus callosum serves to bind together the separate representations of the two halves of the visual field [2, 7, 8]. Indeed, each hemisphere receives information from the opposite visual hemifield; thus, the visual world is represented discontinuously in cortical maps, being split between the two hemispheres along the central vertical meridian. One key role of the callosum is to combine these two partial cortical maps of the visual field into a single, coherent representation [9]. Recent electrophysiological data obtained in the ferret confirm that callosal connections integrate the visual field across the vertical midline in a stimulus-specific manner [10].

2. Anatomy of Callosal Projections in the Visual Cortex

Consistently with a role in perceptual binding, callosal connections are strongly concentrated in a zone at the border between areas 17 and 18, corresponding to the

representation of the vertical meridian in cats, macaques, and humans [9, 11, 12]. Recent studies have employed diffusion tensor imaging to show evidence for callosal connections in human V1 [13–15]. The callosum links cortical loci that are in retinotopic correspondence [16, 17]. Neuroanatomical tracing in cats shows that clusters of callosal boutons are preferentially distributed in regions representing also the same orientation and not only the same visuotopic location in the opposite hemisphere [18].

In cats, interhemispheric fibres originate from a narrow transition zone between area 17 and 18 [8] and, according to the general rule that retinotopic loci are callosally connected, callosal terminals in the opposite hemisphere are particularly concentrated in this area 17/area 18 border [19–21]. Tracing studies show that callosal axons display 2 or 3 clusters of synaptic boutons in layers 2–3 and the upper part of layer 5 [18].

In rodents, different from cats and primates, the entire extent of the primary visual cortex contains callosal cells [22]. However, their terminals are still particularly concentrated in a quite narrow stripe at the area 17/18 border [23, 24]. Recently, Mizuno et al. [25] have confirmed this findings in mice by labelling callosal axons via in utero electroporation of green fluorescent protein. These experiments have also shown that axonal arborisations of callosal cells are mainly located in layers 1–3 and layer 5 [25, 26].

Callosal cells do not constitute a homogenous population, since they have different morphochemical phenotypes [8, 27, 28]. In cats, the vast majority of callosal neurons are large pyramidal cells, and immunohistochemistry observations and studies using the selective uptake of radiolabeled transmitters have failed to identify GABA containing callosal neurons [29]. Nevertheless, experiments with retrograde transport of horseradish peroxidase injected into border region of the opposite hemisphere report the occasional observation of transcallosal nonpyramidal cells in cats [30]. Also, transiently during early rat development immunocytochemical staining reveals numerous GABA-positive fibres in the callosum, which largely disappear at later stages [31]. In keeping with these results, our group has shown that in juvenile rat visual cortex only 1% of callosally projecting neurons display GABA immunoreactivity [32].

In monkeys, chimpanzees, and humans, callosal axons of distinct size interconnect functionally different cortical areas [33, 34]. The axons originating from each cortical site cover a considerable range of conduction velocities, dispersing in time the action potentials transmitted to the other hemisphere. A wide range of temporal delays might expand the number of neuronal ensembles that transcallosal connectivity can activate [33].

3. Physiology of Callosal Connections in the Visual Cortex

Electrophysiological observations have shown that callosal inputs can provide both excitation and inhibition to the contralateral side [35, 36]. On the one hand, the removal of the callosal input to the opposite visual cortex (via

cooling or GABA injections in one hemisphere) results in a decrease of neuronal responsivity in a fraction of the recorded cells, suggesting a callosal excitatory contribution to these neurons. On the other hand, a subset of neurons show an increase in the response magnitude, compatible with the removal of a callosally driven inhibition [35, 36].

These physiological data showing transcallosal inhibitory and excitatory effects are corroborated by the intra- and extracellular results obtained in cats by different groups showing that callosal fibers mainly evoke a direct excitation of neurons in the opposite hemisphere but can also produce a disynaptic inhibitory postsynaptic potential via a local GABAergic cell [8, 37].

The type of information transmitted by the corpus callosum to the visual cortex has been studied more recently by electrophysiological, optical imaging and psychophysical approaches.

Recordings of local field potentials before, during, and after inactivation by cooling of the opposite hemisphere demonstrated that callosal input modifies visual responses in a complex and stimulus-dependent manner [10]. Specifically, callosal influences more frequently depress the responses elicited through the thalamocortical pathway (indicative of interhemispheric inhibition), but facilitatory events are also observed [10]. This callosal excitation is mainly between neurons tuned to the same orientation, consistent with anatomical evidences of direct monosynaptic connections linking neuronal clusters representing the same orientation in the two sides of the brain [18]. Conversely, transcallosal inhibition is both between iso-oriented and cross-oriented neurons. It is possible that this effect is mediated via local interneurons and spread of GABAergic inhibition across columns of different orientations [10].

The callosum also modulates visual response properties, like orientation and direction selectivity across the midline. In particular, in the cat, callosal connections contribute to the strength and specificity of the orientation and directional response in cortical neurons [38]. Cortical domains preferring cardinal contours seem to receive a strong interhemispheric input, that is lost after cooling of the contralateral hemisphere [38].

Another key function of the visual callosal connections is to create transhemispheric neuronal assemblies by synchronizing the activity of neurons in the two hemispheres. Indeed, section of the corpus callosum or inactivation of one side substantially impacts functional coupling of the two hemispheres [39–42].

Callosal connections also play a role in determining cortical binocularity and in other functions such as depth perception, horizontal disparity tuning, contrast sensitivity, and transfer of adaptation [38, 43–45]. Spatial and temporal characteristics of the visual information transmitted through the callosum are similar to those of a lowpass filter, indeed high spatial and temporal frequencies are attenuated, and callosal neurons have reduced sensitivity to low contrasts [43]. A recent study in human has confirmed the importance of callosal communication in processing high contrasts. Indeed, after rTMS silencing of the left visual cortex the

authors found a selective increase, in the opposite hemisphere, of field potentials evoked by high-contrast stimuli [46].

4. Development of the Callosum: Role of Spontaneous Activity and Visual Experience

The development of the corpus callosum is a slow process that spans many years in humans; the fibers appear at 10–11 weeks of gestation but the maturation continues until myelination is completed during puberty [47]. In cats, the callosum is fully developed between 1 and 3 months of age [48]. In rodents maturation of the callosum is complete just after eye opening (postnatal day 15) [25], but the process of myelination continues into adulthood [49].

Visual callosal axons are initially exuberant, but during development they undergo a phenomenon of partial elimination [50, 51]. During the first two postnatal months in cats, the callosal efferent zones become progressively restricted to their adult locations in visual cortex [51]. Also in the rat parietal cortex, the major factor in the progressive restriction of the callosal projection is the withdrawal or degeneration of axon collaterals, rather than the selective death of many of the cells that initially project to the opposite side [52]. Initial exuberance of neuronal connectivity followed by a later phase of axon pruning is a common theme in neural development [53].

Like any other brain structure, the callosal pathway can undergo plastic changes during its early formation and maturation. It has been shown that the development of visual callosal connections is strongly dependent upon neural activity even before eye opening [25, 56]. Neonatal enucleation experiments show that activity is required for the refining of callosal projections. Electron microscopy on sections from enucleated rats shows that eye presence is necessary for the development and/or maintenance of callosal terminals forming multiple synaptic contacts [57]. Eye presence is also important during a window of callosal plasticity (from postnatal day 4 to 6 in rats and mice) to specify callosal maps in a non-NMDA receptor-mediated process [17, 58]. NMDA receptors seem to be required mainly for the initial elaboration of callosal arbor development [17]. In mice, Mizuno et al. [25] explored the role of spontaneous activity in callosal development, showing that a decrease in firing activity of callosal neurons leads to an impaired growth of axon and their arbors. Conversely interfering with firing of callosal target neurons has only a limited effect on the pattern of callosal terminals [25]. During development there has been also demonstrated the presence in the rat cortex of a substantial, but transient population of functional GABAergic transcallosal neurons. These GABAergic neurons are detectable perinatally but do not seem to persist into adulthood and could work as pathfinding or differentiation cues [31]. There are also a few identified molecular determinants responsible for the callosal fate of excitatory projecting neurons in mouse cortex. In the absence of *Fezf2*, a zinc finger transcription factor, cortical neurons adopt the axonal targeting of callosal neurons and their typical strong spike

frequency adaptation in response to intracellular current injection. *Fezf2*^{-/-} neurons also acquire the expression of a known callosal marker, the chromatin remodeling protein *Satb2* [59–61].

Modulation of visual experience also affects the development of callosal connections. Rearing animals in complete darkness from birth exaggerates the partial elimination of callosal projections, with fewer terminating callosal axons at the area 17/18 border [62, 63]. Similarly, bilateral eyelid suture causes a clear reduction of callosal connections, with a 50% reduction of the total number of callosal neurons [64]. Conversely, monocular enucleation produces an abnormally wide distribution of callosal cells at the 17/18 border. This latter effect is similar to that described in cats reared with convergent or divergent strabismus, or monocular eyelid suture. All these manipulations produce a widespread distribution and exuberant number of callosal terminals [65–68].

There are also data showing that visual experience can influence the functional properties of callosal neurons in adult cats. For example, a study demonstrated that MD in adulthood is able to induce functional changes in visual callosal map, leading to an increase of receptive field size and to a loss of orientation selectivity [69].

5. Role of the Callosum in Developmental Maturation of the Visual Cortex

The experiments described so far demonstrate that spontaneous activity and sensory experience can modify the fine connectivity of the corpus callosum. The question arises whether there is a role for inter-hemispheric communication in visual cortical development. The visual cortex is immature at the time of eye opening and gradually develops its functional and structural properties during a critical period early in life [70]. During this time window, experience refines a number of visual properties. Among these, an important marker of maturation is the increase of visual acuity, that in rats reaches adult values around postnatal day 35. In parallel with the maturation of acuity, there is a progressive loss of the potential for plasticity in the cortex. This is usually demonstrated by a downregulation of the effect of a period of monocular eyelid suture (monocular deprivation, MD) on eye preference of cortical cells [70, 71]. Many studies have described the role of visual experience in visual cortex maturation [71–73]. Total lack of visual experience by dark rearing, for example, halts maturation of visual acuity and prolongs the period of sensitivity to MD [71, 73]. While the role of visual experience in cortical maturation is well established, our group has recently addressed the specific role of callosal connectivity in functional development of the visual cortex. Specifically, we produced a unilateral, prolonged silencing of activity in the developing rat primary visual cortex by taking advantage of the clostridial enzyme botulinum neurotoxin E (BoNT/E). BoNT/E is a metalloprotease that enters the cytosol of nerve terminals close to the site of delivery and specifically cleaves the synaptic protein SNAP-25 (synaptosomal-associated protein

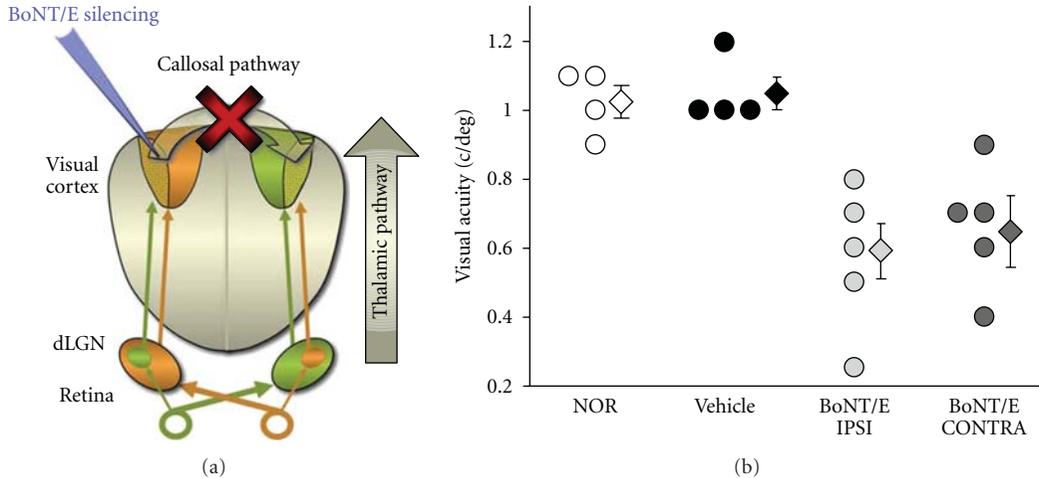


FIGURE 1: Silencing callosal input during the early critical period impairs maturation of visual acuity. (a) Schematics of the experimental protocol. BoNT/E was unilaterally injected into the visual cortex in P14 rat pups to cause a prolonged silencing of one hemisphere. The contralateral, uninjected side has a normal visual experience through the retinogeniculate pathway and only lacks callosal input activity. (b) Bilateral impairments in visual acuity at P35 after unilateral injection of BoNT/E at P14: summary of visual acuities in naïve rats (NOR), rats injected with vehicle (VEHICLE) and in the hemisphere ipsilateral (IPSI) and contralateral (CONTRA) to BoNT/E infusion. Each circle represents one animal. Mean visual acuity (diamonds) is significantly reduced in both hemispheres of BoNT/E rats in comparison with that in normal or vehicle-injected animals. Error bars indicate SE. Data are from [54].

of 25 kDa), causing a prolonged blockade of transmitter release [74–76].

We unilaterally injected BoNT/E into the visual cortex of rat pups at the time of eye opening [54]. BoNT/E injection resulted in a selective blockade of activity in the injected, but not contralateral, cortex that persisted at least 2 weeks, thus spanning most of the “critical period” for cortical development [71]. This experimental approach is ideal to dissect the role of the interhemispheric connections during cortical development, because the uninjected cortex experiences normal vision through the retinogeniculate pathway and only lacks callosal input (see scheme in Figure 1(a)). This transient unilateral silencing of intrinsic cortical activity prevented functional cortical maturation on both sides. The injected cortex displayed deficits in visual acuity and an extension of the critical period for ocular dominance plasticity. Remarkably, these same effects were detectable in the visual cortex of the opposite uninjected side (Figure 1(b)), pointing to a crucial role for interhemispheric connections in postnatal development. Thus, maturation of the blocked cortex was superimposable to that of the opposite side, which only lacks callosal input and maintains normal afferent activity through the direct retinogeniculate pathway (Figure 1(b)). The very similar developmental deficits observed ipsilateral and contralateral to the activity blockade indicate a fundamental role for callosal linkages in coordinating the process of cortical maturation [54]. This finding is consistent with the well-known role of the callosum in synchronizing activity in the two halves of the brain [39–42].

Explanations for these bilateral effects after unilateral silencing may implicate the lack of a sustaining callosal input to the opposite visual cortex during activity blockade in one

side [54, 77]. In teleological terms, parallel development of the two sides of the brain is needed to ensure a match in information processing between the cerebral hemispheres; the results of these experiments show that transcallosal pathways mediate this coordinated maturation.

To corroborate this idea, we took advantage of a mouse model with conditional deletion of the AP2 γ transcription factor. Deletion of AP2 γ during development results in a specific reduction of upper layer neurons in the occipital cortex, particularly callosally projecting neurons [78]. As a result, adult AP2 γ conditional knockout mice display reduced size of the corpus callosum. At the functional level, this phenotype was coupled to a profound reduction in visual acuity. As the reduced visual acuity was reminiscent of a physiologically more immature state of the visual cortex, we tested the hypothesis of whether this was also accompanied by maintenance of a higher degree of plasticity, as is the case at more immature stages. Indeed, ocular dominance plasticity triggered by a brief period of MD was retained in adult AP2 $\gamma^{-/-}$ mice. These data provide further support for the hypothesis that callosal projections act as an important determinant for the functional maturation of visual cortex [78].

Reports in the literature suggest that the role of the callosum in cortical maturation might be well conserved across species. Indeed, transection of the callosum in kittens during an early phase of postnatal development (but not at later stages) produces a reduction in behaviorally measured visual acuity, supporting a role for interhemispheric communication in cortical maturation [79]. Monkeys that received unilateral lesions of primary visual cortex in infancy display impairments of stimulus detection in the intact visual hemifield [80]. There may be a similar early sensitive

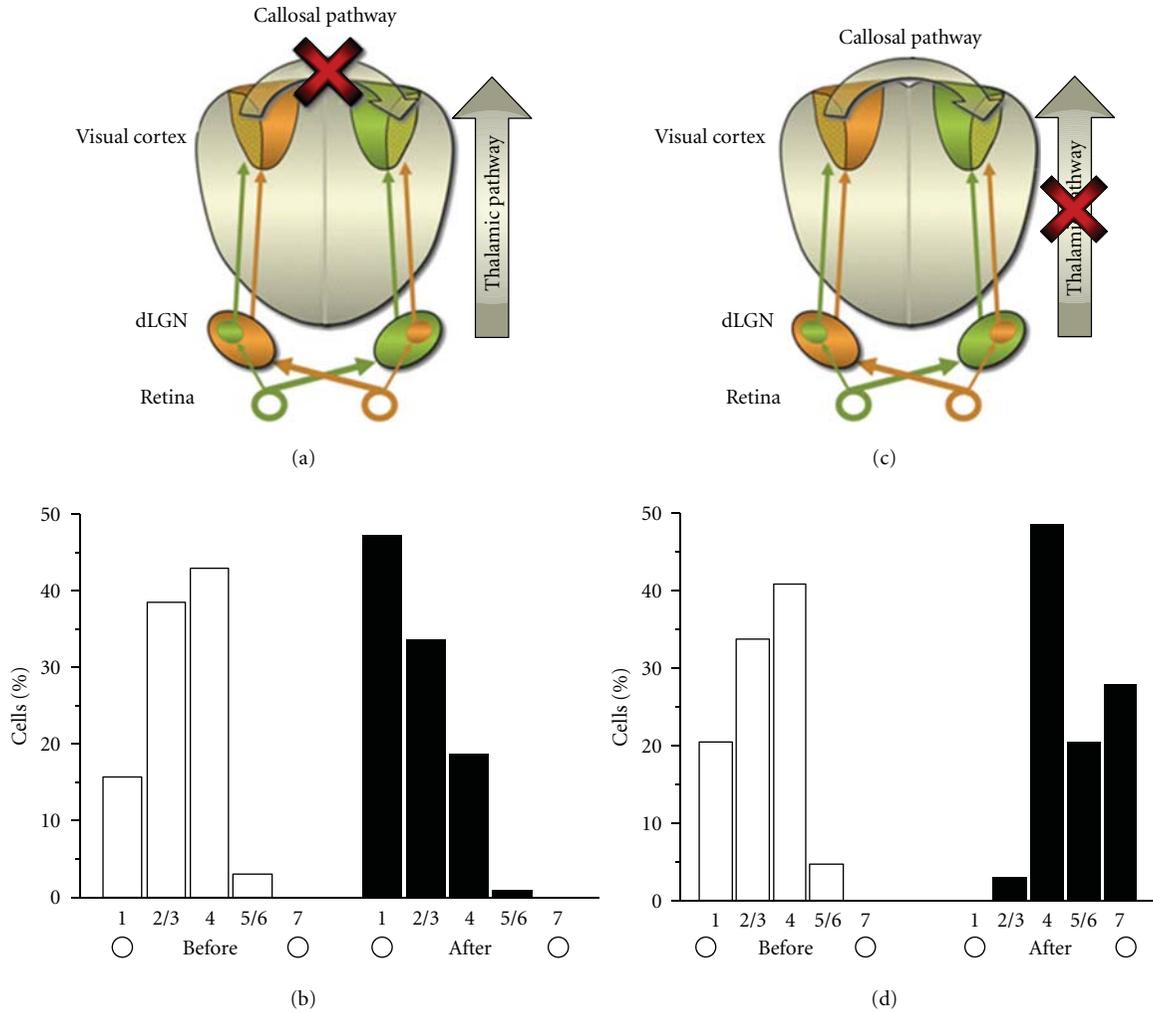


FIGURE 2: Relative contribution of callosal and thalamic pathways to cortical binocularity. (a, b) Effects of acute callosal silencing on OD in rat primary visual cortex. Binocularity was measured in one side before and after acute pharmacological inactivation of the opposite cortex (a). The results indicate a significant shift of cortical OD towards the contralateral eye, due to the reduction of responses driven by the ipsilateral eye (b). (c, d) Effects of acute geniculate silencing on OD in rat primary visual cortex. The OD histogram shifts towards the ipsilateral eye following acute geniculate inactivation (d). This effect is mainly due to a loss of contralateral eye-driven input. Data are from [32, 55].

period in humans, when callosal integrity appears to be particularly important for the development of visual acuity. Indeed, children born preterm show correlation between white matter microstructure and visual acuity [81]. In keeping with the idea of a “critical period” for the role of the callosum in acuity maturation, adult patients experiencing callosotomy have no impairments of visual acuity [82]. It is important to mention that functional maturation of other cortical properties during development appears to proceed independent of callosal influences. For example, development of orientation selectivity in ferret visual cortex is not affected by activity blockade in the contralateral hemisphere [83].

The development of visual acuity is dependent on proper callosal function only during an early critical period, but the importance of the callosal pathway in integrating cerebral processing is still apparent in adults. Patients with

unilateral occipital cortex injury show reduced spatial and temporal sensitivities in the sighted hemifield [84, 85]. Moreover, patients with hemianopia show impairments in figure detection tasks also in the intact hemi-field, suggesting that this deficit may be caused by loss of interhemispheric interactions [86].

6. The Role of the Corpus Callosum in Cortical Binocularity

The particularly high concentration of callosal terminals at the area 17/18 border, close to the vertical meridian, prompts for a role of the callosum in binocularity.

In cats, callosal neurons are highly binocular cells (i.e., they respond equally to a stimulus presented to the ipsilateral or the contralateral eye) [7, 87] and a number of experiments

have been performed to probe a role for the corpus callosum in eye preference. However, they have yielded contradictory results. Section of the callosum had no effect [88] or lead to a dramatic reduction in binocularity [19, 89–91] in cat visual cortex. The discrepancies in these results may be consequence of technical aspects, including age at which the callosal section is performed and time elapsed between surgery and recording.

In rodents, visual cortex responsiveness is biased towards the contralateral eye [71, 92–94], due to the high percentage (over 95%) of retinal fibres crossing at the chiasm [95], as compared to about 50% of fibers (from the nasal hemiretina) in cats, monkeys, and humans. In rats and mice, the contralateral bias is progressively reduced getting closer to the highly binocular V1/V2 border [92, 96] that maps the vertical meridian and where callosal projections are particularly dense [23, 25]. To clarify the role of the callosum in cortical binocularity in rats, we recorded single unit responses in layers 2-3 before and after acute blockade of callosal input. This was achieved via delivery of muscimol, a GABA_A receptor agonist, into the striate cortex contralateral to the recording site (Figure 2(a)). Extracellular recordings of spiking activity demonstrated an enhancement of the contralateral bias of single units following injection of muscimol into the opposite hemisphere [32]. The ocular dominance (OD) shift towards the contralateral eye was due to a reduction of the strength of responses evoked by the ipsilateral eye [32]. The effect was observed for cells with receptive fields close to the vertical meridian and disappeared at more medial locations in the cortex (i.e., within the core of V1, mapping more peripheral parts of the visual hemifield) [32]. This is consistent with callosal projections being particularly concentrated in the lateral aspect of primary visual cortex (vertical meridian) [23, 25]. Our results are consistent with previous experiments in albino rats that also show a dramatic shift of eye preference (due to loss of ipsilateral eye responses) after cooling of the opposite cortex [97].

We have also carried out a complementary experiment where visual responses were recorded before and after acute thalamic inactivation in the same animal. We measured OD before and after removal of the thalamic input with an acute silencing of the geniculate via tetrodotoxin (TTX) injection [55]. This protocol allows one to isolate visual responses driven exclusively by callosal afferents. After geniculate inactivation, OD shifted towards the ipsilateral eye (Figures 2(c) and 2(d)) and this was due to a robust loss of contralateral eye-driven responses, while ipsilateral eye responses were reduced to a less extent [55].

Altogether, these data identify two sources of binocularity in rat visual cortex: the retinogeniculate pathway carrying mainly contralateral eye input, and the callosal pathway mainly providing ipsilateral eye input to neurons in the opposite cortex [32, 55, 97]. It is important to mention that the role of callosal input in generating binocular responses might be a specific property of the rodent visual system, given that the proportion of retinogeniculate cortical projections for the ipsilateral eye is low in rodents (due to the massive fiber crossing at the chiasm; see the aforementioned part).

Thus, in the normal rat visual cortex, binocularity appears to depend on the function of callosal fibres. In particular, a substantial fraction of the ipsilateral eye drive on cortical responses arrives via callosal connections from the opposite hemisphere, where it is the dominant eye.

7. Role of the Callosum in Ocular Dominance Plasticity

Given the importance of callosal input for cortical development and in the generation of binocularity, our group has recently investigated a possible involvement of the callosum in the plastic shift of OD triggered by monocular deprivation. Experience is particularly influential during sensitive periods during development, when appropriate patterns of functional connectivity are selected from wide varieties of potential patterns [98]. During this period, brief modifications of visual experience can induce a profound rearrangement in visual cortical circuitry. Brief MD during the critical period unbalances the amount and pattern of visual information coming from the two eyes and causes an OD shift towards the open eye. In particular, the deprived eye loses its ability in driving cortical neurons (response depression), while neurons respond more vigorously to the open eye (response potentiation).

We deprived a group of rats at the peak of the critical period for seven days to induce the expected shift in OD. Then we measured the OD shift before and after acute silencing of the callosal pathway, via muscimol infusion of the cortex opposite to the recording hemisphere. This experimental protocol allows plasticity to proceed normally and probes the results of acute removal of callosal input. Surprisingly, muscimol injection opposite to the recording cortex restored binocularity after MD in juvenile rats [32]. This recovery of binocularity following callosal silencing was due to an increase in the strength of the deprived eye. Thus, acute removal of callosal influence following MD unmasks deprived eye inputs. These data indicate that callosal afferents act primarily to inhibit closed eye inputs under visual deprivation [32]. In keeping with this observation, continuous silencing of callosal input throughout the MD prevented the loss of responsiveness of the deprived eye, resulting in a dramatic reduction of the OD shift. Thus, transcallosal connections are crucially involved in the weakening of deprived eye responses during MD [32].

An enhanced intracortical inhibition has been previously shown to contribute to the reduced ability of deprived afferents to activate cortical neurons [99]. Our findings demonstrate that callosal inputs are a major source of inhibition following MD. It is likely that this inhibition is relayed by local interneurons, as about 99% of callosal cells are glutamatergic [32]. The specific type(s) of interneurons receiving callosal input are presently not known and are the subject of intense investigation.

It is interesting to mention that transcallosal inhibition appears to play an important role in plastic events occurring during several brain pathological conditions. For example, in neglect patients some of the behavioural symptoms are

attributable to a pathological state of increased inhibition exerted onto the damaged parietal cortex by the contralateral, intact hemisphere [100, 101]. In these patients, silencing the intact side with transcranial magnetic stimulation results in substantial, long-lasting amelioration of the behavioural performances [100, 101].

8. Conclusions

In this paper, we have described the contribution of transcallosal pathways to experience-dependent plasticity in the visual cortex. On one side, the fine connectivity of callosal fibers is affected by alterations of visual input (e.g., visual deprivation). On the other hand, the transcallosal route appears to play a critical role in ensuring a functional matching in the developmental maturation of the two cerebral hemispheres during an early critical period. Callosal fibers also contribute to normal binocularity and to the shift of OD occurring after monocular deprivation in rat visual cortex. Thus, the corpus callosum is a key player in the plastic phenomena that underlie adaptation of the juvenile brain to a changing environment.

References

- [1] G. Berlucchi, S. Aglioti, C. A. Marzi, and G. Tassinari, "Corpus callosum and simple visuomotor integration," *Neuropsychologia*, vol. 33, no. 8, pp. 923–936, 1995.
- [2] G. M. Innocenti, D. Aggoun-Zouaoui, and P. Lehmann, "Cellular aspects of callosal connections and their development," *Neuropsychologia*, vol. 33, no. 8, pp. 961–987, 1995.
- [3] M. S. Gazzaniga, "Cerebral specialization and interhemispheric communication. Does the corpus callosum enable the human condition?" *Brain*, vol. 123, no. 7, pp. 1293–1326, 2000.
- [4] J. Tomasch, "Size, distribution, and number of fibres in the human corpus callosum," *The Anatomical Record*, vol. 119, no. 1, pp. 119–135, 1954.
- [5] J. S. Bloom and G. W. Hynd, "The role of the corpus callosum in interhemispheric transfer of information: excitation or inhibition?" *Neuropsychology Review*, vol. 15, no. 2, pp. 59–71, 2005.
- [6] L. K. Paul, W. S. Brown, R. Adolphs et al., "Agenesis of the corpus callosum: genetic, developmental and functional aspects of connectivity," *Nature Reviews Neuroscience*, vol. 8, no. 4, pp. 287–299, 2007.
- [7] G. Berlucchi and G. Rizzolatti, "Binocularly driven neurons in visual cortex of split-chiasm cats," *Science*, vol. 159, no. 3812, pp. 308–310, 1968.
- [8] G. M. Innocenti, "The primary visual pathway through the corpus callosum: morphological and functional aspects in the cat," *Archives Italiennes de Biologie*, vol. 118, no. 2, pp. 124–188, 1980.
- [9] J. C. Houzel and C. Milleret, "Visual inter-hemispheric processing: constraints and potentialities set by axonal morphology," *Journal of Physiology Paris*, vol. 93, no. 4, pp. 271–284, 1999.
- [10] V. A. Makarov, K. E. Schmidt, N. P. Castellanos, L. Lopez-Aguado, and G. M. Innocenti, "Stimulus-dependent interaction between the visual areas 17 and 18 of the 2 hemispheres of the ferret (*Mustela putorius*)," *Cerebral Cortex*, vol. 18, no. 8, pp. 1951–1960, 2008.
- [11] D. C. Van Essen, W. T. Newsome, and J. L. Bixby, "The pattern of interhemispheric connections and its relationship to extrastriate visual areas in the macaque monkey," *Journal of Neuroscience*, vol. 2, no. 3, pp. 265–283, 1982.
- [12] S. Clarke and J. Miklosy, "Occipital cortex in man: organization of callosal connections, related myelo- and cytoarchitecture, and putative boundaries of functional visual areas," *Journal of Comparative Neurology*, vol. 298, no. 2, pp. 188–214, 1990.
- [13] R. F. Dougherty, M. Ben-Shachar, R. Bammer, A. A. Brewer, and B. A. Wandell, "Functional organization of human occipital-callosal fiber tracts," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 102, no. 20, pp. 7350–7355, 2005.
- [14] M. C. Putnam, M. S. Steven, K. W. Doron, A. C. Riggall, and M. S. Gazzaniga, "Cortical projection topography of the human splenium: hemispheric asymmetry and individual differences," *Journal of Cognitive Neuroscience*, vol. 22, no. 8, pp. 1662–1669, 2010.
- [15] M. Saenz and I. Fine, "Topographic organization of V1 projections through the corpus callosum in humans," *NeuroImage*, vol. 52, no. 4, pp. 1224–1229, 2010.
- [16] J. W. Lewis and J. F. Olavarria, "Two rules for callosal connectivity in striate cortex of the rat," *Journal of Comparative Neurology*, vol. 361, no. 1, pp. 119–137, 1995.
- [17] J. F. Olavarria, R. Laing, R. Hiroi, and J. Lasiene, "Topography and axon arbor architecture in the visual callosal pathway: effects of deafferentation and blockade of N-methyl-D-aspartate receptors," *Biological Research*, vol. 41, no. 4, pp. 413–424, 2008.
- [18] N. L. Rochefort, P. Buzás, N. Quenech'Du et al., "Functional selectivity of interhemispheric connections in cat visual cortex," *Cerebral Cortex*, vol. 19, no. 10, pp. 2451–2465, 2009.
- [19] C. Blakemore, Y. Diao, and M. Pu, "Possible functions of the interhemispheric connexions between visual cortical areas in the cat," *Journal of Physiology*, vol. 337, pp. 331–349, 1983.
- [20] B. R. Payne, "Function of the corpus callosum in the representation of the visual field in cat visual cortex," *Visual Neuroscience*, vol. 5, no. 2, pp. 205–211, 1990.
- [21] B. R. Payne and D. F. Siwek, "Visual-field map in the callosal recipient zone at the border between areas 17 and 18 in the cat," *Visual Neuroscience*, vol. 7, no. 3, pp. 221–236, 1991.
- [22] J. F. Olavarria and R. C. Van Sluyters, "Overall pattern of callosal connections in visual cortex of normal and enucleated cats," *Journal of Comparative Neurology*, vol. 363, no. 2, pp. 161–176, 1995.
- [23] C. G. Cusick and R. D. Lund, "The distribution of the callosal projection to the occipital visual cortex in rats and mice," *Brain Research*, vol. 214, no. 2, pp. 239–259, 1981.
- [24] S. Jacobson, "Distribution of commissural axon terminals in the rat neocortex," *Experimental Neurology*, vol. 28, no. 2, pp. 193–205, 1970.
- [25] H. Mizuno, T. Hirano, and Y. Tagawa, "Evidence for activity-dependent cortical wiring: formation of interhemispheric connections in neonatal mouse visual cortex requires projection neuron activity," *Journal of Neuroscience*, vol. 27, no. 25, pp. 6760–6770, 2007.
- [26] S. Jacobson and J. Q. Trojanowski, "The cells of origin of the corpus callosum in rat, cat and rhesus monkey," *Brain Research*, vol. 74, no. 1, pp. 149–155, 1974.
- [27] E. H. Buhl and W. Singer, "The callosal projection in cat visual cortex as revealed by a combination of retrograde tracing and intracellular injection," *Experimental Brain Research*, vol. 75, no. 3, pp. 470–476, 1989.

- [28] T. Voigt, S. LeVay, and M. A. Stamnes, "Morphological and immunocytochemical observations on the visual callosal projections in the cat," *Journal of Comparative Neurology*, vol. 272, no. 3, pp. 450–460, 1988.
- [29] A. J. Elberger, "Selective labeling of visual corpus callosum connections with aspartate in cat and rat," *Visual Neuroscience*, vol. 2, no. 1, pp. 81–85, 1989.
- [30] A. Peters, B. R. Payne, and K. Josephson, "Transcallosal non-pyramidal cell projections from visual cortex in the cat," *Journal of Comparative Neurology*, vol. 302, no. 1, pp. 124–142, 1990.
- [31] F. Kimura, "GABAergic transcallosal neurons in developing rat neocortex," *European Journal of Neuroscience*, vol. 9, no. 6, pp. 1137–1143, 1997.
- [32] L. Restani, C. Cerri, M. Pietrasanta, L. Gianfranceschi, L. Maffei, and M. Caleo, "Functional masking of deprived eye responses by callosal input during ocular dominance plasticity," *Neuron*, vol. 64, no. 5, pp. 707–718, 2009.
- [33] R. Caminiti, H. Ghaziri, R. Galuske, P. R. Hof, and G. M. Innocenti, "Evolution amplified processing with temporally dispersed slow neuronal connectivity in primates," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 46, pp. 19551–19556, 2009.
- [34] S. Tomasi, R. Caminiti, and G. M. Innocenti, "Areal differences in diameter and length of corticofugal projections," *Cerebral Cortex*, vol. 22, no. 6, pp. 1463–1472, 2012.
- [35] B. R. Payne, D. F. Siwek, and S. G. Lomber, "Complex transcallosal interactions in visual cortex," *Visual Neuroscience*, vol. 6, no. 3, pp. 283–289, 1991.
- [36] J. S. Sun, B. Li, M. H. Ma, and Y. C. Diao, "Transcallosal circuitry revealed by blocking and disinhibiting callosal input in the cat," *Visual Neuroscience*, vol. 11, no. 2, pp. 189–197, 1994.
- [37] K. Toyama, K. Matsunami, T. Ohno, and S. Tokashiki, "An intracellular study of neuronal organization in the visual cortex," *Experimental Brain Research*, vol. 21, no. 1, pp. 45–66, 1974.
- [38] K. E. Schmidt, S. G. Lomber, and G. M. Innocenti, "Specificity of neuronal responses in primary visual cortex is modulated by interhemispheric corticocortical input," *Cerebral Cortex*, vol. 20, no. 12, pp. 2776–2786, 2010.
- [39] M. H. J. Munk, L. G. Nowak, J. I. Nelson, and J. Bullier, "Structural basis of cortical synchronization II. Effects of cortical lesions," *Journal of Neurophysiology*, vol. 74, no. 6, pp. 2401–2414, 1995.
- [40] M. G. Knyazeva, D. C. Kiper, V. Y. Vildavski, P. A. Despland, M. Maeder-Ingvar, and G. M. Innocenti, "Visual stimulus-dependent changes in interhemispheric EEG coherence in humans," *Journal of Neurophysiology*, vol. 82, no. 6, pp. 3095–3107, 1999.
- [41] A. K. Engel, P. Konig, A. K. Kreiter, and W. Singer, "Interhemispheric synchronization of oscillatory neuronal responses in cat visual cortex," *Science*, vol. 252, no. 5010, pp. 1177–1179, 1991.
- [42] C. Carmeli, L. Lopez-Aguadao, K. E. Schmidt, O. De Feo, and G. M. Innocenti, "A novel interhemispheric interaction: modulation of neuronal cooperativity in the visual areas," *PLoS ONE*, vol. 2, no. 12, article e1287, 2007.
- [43] N. Berardi, S. Bisti, A. Fiorentini, and L. Maffei, "The transfer of visual information across the corpus callosum in cats, monkeys and humans: spatial and temporal properties," *Progress in Brain Research*, vol. 75, pp. 181–185, 1988.
- [44] L. Maffei, N. Berardi, and S. Bisti, "Interocular transfer of adaptation after effect in neurons of area 17 and 18 of split chiasm cats," *Journal of Neurophysiology*, vol. 55, no. 5, pp. 966–976, 1986.
- [45] D. E. Mitchell and C. Blakemore, "Binocular depth perception and the corpus callosum," *Vision Research*, vol. 10, no. 1, pp. 49–54, 1970.
- [46] T. Bocci, M. Caleo, E. Giorli et al., "Transcallosal inhibition dampens neural responses to high contrast stimuli in human visual cortex," *Neuroscience*, vol. 187, pp. 43–51, 2011.
- [47] J. B. Hellige, *Hemispheric Asymmetry: What's Right and What's Left*, Harvard University Press, Cambridge, Mass, USA, 1993.
- [48] H. Kennedy and C. Dehay, "Functional implications of the anatomical organization of the callosal projections of visual areas V1 and V2 in the macaque monkey," *Behavioural Brain Research*, vol. 29, no. 3, pp. 225–236, 1988.
- [49] M. A. Yates and J. M. Juraska, "Increases in size and myelination of the rat corpus callosum during adulthood are maintained into old age," *Brain Research*, vol. 1142, no. 1, pp. 13–18, 2007.
- [50] G. M. Innocenti and R. Caminiti, "Postnatal shaping of callosal connections from sensory areas," *Experimental Brain Research*, vol. 38, no. 4, pp. 381–394, 1980.
- [51] G. M. Innocenti, "Growth and reshaping of axons in the establishment of visual callosal connections," *Science*, vol. 212, no. 4496, pp. 824–827, 1981.
- [52] D. D. O'Leary, B. B. Stanfield, and W. M. Cowan, "Evidence that the early postnatal restriction of the cells of origin of the callosal projection is due to the elimination of axonal collaterals rather than to the death of neurons," *Brain Research*, vol. 227, no. 4, pp. 607–617, 1981.
- [53] G. M. Innocenti and D. J. Price, "Exuberance in the development of cortical networks," *Nature Reviews Neuroscience*, vol. 6, no. 12, pp. 955–965, 2005.
- [54] M. Caleo, L. Restani, L. Gianfranceschi et al., "Transient synaptic silencing of developing striate cortex has persistent effects on visual function and plasticity," *Journal of Neuroscience*, vol. 27, no. 17, pp. 4530–4540, 2007.
- [55] C. Cerri, L. Restani, and M. Caleo, "Callosal contribution to ocular dominance in rat primary visual cortex," *European Journal of Neuroscience*, vol. 32, no. 7, pp. 1163–1169, 2010.
- [56] G. M. Innocenti, S. Clarke, and R. Kraftsik, "Interchange of callosal and association projections in the developing visual cortex," *Journal of Neuroscience*, vol. 6, no. 5, pp. 1384–1409, 1986.
- [57] S. A. Sorensen, T. A. Jones, and J. F. Olavarria, "Neonatal enucleation reduces the proportion of callosal boutons forming multiple synaptic contacts in rat striate cortex," *Neuroscience Letters*, vol. 351, no. 1, pp. 17–20, 2003.
- [58] J. F. Olavarria and R. Hiroi, "Retinal influences specify cortico-cortical maps by postnatal day six in rats and mice," *Journal of Comparative Neurology*, vol. 459, no. 2, pp. 156–172, 2003.
- [59] B. Chen, S. S. Wang, A. M. Hattox, H. Rayburn, S. B. Nelson, and S. K. McConnell, "The Fezf2-Ctip2 genetic pathway regulates the fate choice of subcortical projection neurons in the developing cerebral cortex," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 32, pp. 11382–11387, 2008.
- [60] A. M. Hattox and S. B. Nelson, "Layer V neurons in mouse cortex projecting to different targets have distinct physiological properties," *Journal of Neurophysiology*, vol. 98, no. 6, pp. 3330–3340, 2007.
- [61] S. Lodato, C. Rouaux, K. B. Quast et al., "Excitatory projection neuron subtypes control the distribution of local

- inhibitory interneurons in the cerebral cortex," *Neuron*, vol. 69, no. 4, pp. 763–779, 2011.
- [62] D. O. Frost and Y. P. Moy, "Effects of dark rearing on the development of visual callosal connections," *Experimental Brain Research*, vol. 78, no. 1, pp. 203–213, 1989.
- [63] R. D. Lund and D. E. Mitchell, "The effects of dark-rearing on visual callosal connections of cats," *Brain Research*, vol. 167, no. 1, pp. 172–175, 1979.
- [64] G. M. Innocenti, D. O. Frost, and J. Illes, "Maturation of visual callosal connections in visually deprived kittens: a challenging critical period," *Journal of Neuroscience*, vol. 5, no. 2, pp. 255–267, 1985.
- [65] C. Milleret, "Visual callosal connections and strabismus," *Behavioural Brain Research*, vol. 64, no. 1-2, pp. 85–95, 1994.
- [66] N. E. Berman and B. R. Payne, "Alterations in connections of the corpus callosum following convergent and divergent strabismus," *Brain Research*, vol. 274, no. 2, pp. 201–212, 1983.
- [67] G. M. Innocenti and D. O. Frost, "Effects of visual experience on the maturation of the efferent system to the corpus callosum," *Nature*, vol. 280, no. 5719, pp. 231–234, 1979.
- [68] D. O. Frost, Y. P. Moy, and D. C. Smith, "Effects of alternating monocular occlusion on the development of visual callosal connections," *Experimental Brain Research*, vol. 83, no. 1, pp. 200–209, 1990.
- [69] L. Watroba, P. Buser, and C. Milleret, "Impairment of binocular vision in the adult cat induces plastic changes in the callosal cortical map," *European Journal of Neuroscience*, vol. 14, no. 6, pp. 1021–1029, 2001.
- [70] N. Berardi, T. Pizzorusso, and L. Maffei, "Critical periods during sensory development," *Current Opinion in Neurobiology*, vol. 10, no. 1, pp. 138–145, 2000.
- [71] M. Fagiolini, T. Pizzorusso, N. Berardi, L. Domenici, and L. Maffei, "Functional postnatal development of the rat primary visual cortex and the role of visual experience: dark rearing and monocular deprivation," *Vision Research*, vol. 34, no. 6, pp. 709–720, 1994.
- [72] L. C. Katz and C. J. Shatz, "Synaptic activity and the construction of cortical circuits," *Science*, vol. 274, no. 5290, pp. 1133–1138, 1996.
- [73] S. M. Sherman and P. D. Spear, "Organization of visual pathways in normal and visually deprived cats," *Physiological Reviews*, vol. 62, no. 2, pp. 738–855, 1982.
- [74] G. Schiavo, M. Matteoli, and C. Montecucco, "Neurotoxins affecting neuroexocytosis," *Physiological Reviews*, vol. 80, no. 2, pp. 717–766, 2000.
- [75] B. Davletov, M. Bajohrs, and T. Binz, "Beyond BOTOX: advantages and limitations of individual botulinum neurotoxins," *Trends in Neurosciences*, vol. 28, no. 8, pp. 446–452, 2005.
- [76] Y. Bozzi, L. Costantin, F. Antonucci, and M. Caleo, "Action of botulinum neurotoxins in the central nervous system: antiepileptic effects," *Neurotoxicity Research*, vol. 9, no. 2-3, pp. 197–203, 2006.
- [77] G. Berlucchi, "What is callosal plasticity?" in *Cortical Plasticity*, L. M. Chalupa et al., Ed., pp. 235–246, MIT Press, 2011.
- [78] L. Pinto, D. Drechsel, M. T. Schmid et al., "AP2 γ regulates basal progenitor fate in a region- and layer-specific manner in the developing cortex," *Nature Neuroscience*, vol. 12, no. 10, pp. 1229–1237, 2009.
- [79] A. J. Elberger, "The existence of a separate, brief critical period for the corpus callosum to affect visual development," *Behavioural Brain Research*, vol. 11, no. 3, pp. 223–231, 1984.
- [80] T. Moore, H. R. Rodman, A. B. Repp, C. G. Gross, and R. S. Mezrich, "Greater residual vision in monkeys after striate cortex damage in infancy," *Journal of Neurophysiology*, vol. 76, no. 6, pp. 3928–3933, 1996.
- [81] S. Lindqvist, J. Skranes, L. Eikenes, and et al, "Visual function and white matter microstructure in very-low-birth-weight (VLBW) adolescents—a DTI study," *Vision Research*, vol. 51, no. 18, pp. 2063–2070, 2011.
- [82] S. R. Afraz, L. Montaser-Kouhsari, M. Vaziri-Pashkam, and F. Moradi, "Interhemispheric visual interaction in a patient with posterior callosotomy," *Neuropsychologia*, vol. 41, no. 5, pp. 597–604, 2003.
- [83] B. Chapman and M. P. Stryker, "Development of orientation selectivity in ferret visual cortex and effects of deprivation," *Journal of Neuroscience*, vol. 13, no. 12, pp. 5251–5262, 1993.
- [84] M. Rizzo and D. A. Robin, "Bilateral effects of unilateral visual cortex lesions in human," *Brain*, vol. 119, no. 3, pp. 951–963, 1996.
- [85] R. F. Hess and J. S. Pointer, "Spatial and temporal contrast sensitivity in hemianopia: a comparative study of the sighted and blind hemifields," *Brain*, vol. 112, no. 4, pp. 871–894, 1989.
- [86] G. V. Paramei and B. A. Sabel, "Contour-integration deficits on the intact side of the visual field in hemianopia patients," *Behavioural Brain Research*, vol. 188, no. 1, pp. 109–124, 2008.
- [87] D. H. Hubel and T. N. Wiesel, "Cortical and callosal connections concerned with the vertical meridian of visual fields in the cat," *Journal of Neurophysiology*, vol. 30, no. 6, pp. 1561–1573, 1967.
- [88] D. Minciacchi and A. Antonini, "Binocularity in the visual cortex of the adult cat does not depend on the integrity of the corpus callosum," *Behavioural Brain Research*, vol. 13, no. 2, pp. 183–192, 1984.
- [89] B. R. Payne, A. J. Elberger, N. Berman, and E. H. Murphy, "Binocularity in the cat visual cortex is reduced by sectioning the corpus callosum," *Science*, vol. 207, no. 4435, pp. 1097–1099, 1980.
- [90] A. J. Elberger and E. L. Smith III, "The critical period for corpus callosum section to affect cortical binocularity," *Experimental Brain Research*, vol. 57, no. 2, pp. 213–223, 1985.
- [91] U. Yinon, M. Chen, and S. Gelerstein, "Binocularity and excitability loss in visual cortex cells of corpus callosum transected kittens and cats," *Brain Research Bulletin*, vol. 29, no. 5, pp. 541–552, 1992.
- [92] J. A. Gordon and M. P. Stryker, "Experience-dependent plasticity of binocular responses in the primary visual cortex of the mouse," *Journal of Neuroscience*, vol. 16, no. 10, pp. 3274–3286, 1996.
- [93] T. K. Hensch, M. Fagiolini, N. Mataga, M. P. Stryker, S. Baekkeskov, and S. F. Kash, "Local GABA circuit control of experience-dependent plasticity in developing visual cortex," *Science*, vol. 282, no. 5393, pp. 1504–1508, 1998.
- [94] N. B. Sawtell, M. Y. Frenkel, B. D. Philpot, K. Nakazawa, S. Tonegawa, and M. F. Bear, "NMDA receptor-dependent ocular dominance plasticity in adult visual cortex," *Neuron*, vol. 38, no. 6, pp. 977–985, 2003.
- [95] A. Sefton and B. Dreher, "Visual system," in *The Rat Nervous System*, G. Paxinos, Ed., pp. 833–898, Academic Press, 1995.
- [96] M. Caleo, C. Lodovichi, T. Pizzorusso, and L. Maffei, "Expression of the transcription factor Zif268 in the visual cortex of monocularly deprived rats: effects of nerve growth factor," *Neuroscience*, vol. 91, no. 3, pp. 1017–1026, 1999.

- [97] Y. C. Diao, Y. K. Wang, and M. L. Pu, "Binocular responses of cortical cells and the callosal projection in the albino rat," *Experimental Brain Research*, vol. 49, no. 3, pp. 410–418, 1983.
- [98] W. Zheng and E. I. Knudsen, "GABAergic inhibition antagonizes adaptive adjustment of the owl's auditory space map during the initial phase of plasticity," *Journal of Neuroscience*, vol. 21, no. 12, pp. 4356–4365, 2001.
- [99] A. Maffei, K. Nataraj, S. B. Nelson, and G. G. Turrigiano, "Potentiation of cortical inhibition by visual deprivation," *Nature*, vol. 443, no. 7107, pp. 81–84, 2006.
- [100] S. Fecteau, A. Pascual-Leone, and H. Théoret, "Paradoxical facilitation of attention in healthy humans," *Behavioural Neurology*, vol. 17, no. 3-4, pp. 159–162, 2006.
- [101] B. Fierro, F. Brighina, and E. Bisiach, "Improving neglect by TMS," *Behavioural Neurology*, vol. 17, no. 3-4, pp. 169–176, 2006.

Research Article

Cross-Modal Recruitment of Primary Visual Cortex by Auditory Stimuli in the Nonhuman Primate Brain: A Molecular Mapping Study

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Received 1 February 2012; Revised 17 April 2012; Accepted 7 May 2012

Academic Editor: Ron Kupers

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Recent studies suggest that exposure to only one component of audiovisual events can lead to cross-modal cortical activation. However, it is not certain whether such crossmodal recruitment can occur in the absence of explicit conditioning, semantic factors, or long-term associations. A recent study demonstrated that crossmodal cortical recruitment can occur even after a brief exposure to bimodal stimuli without semantic association. In addition, the authors showed that the primary visual cortex is under such crossmodal influence. In the present study, we used molecular activity mapping of the immediate early gene *zif268*. We found that animals, which had previously been exposed to a combination of auditory and visual stimuli, showed increased number of active neurons in the primary visual cortex when presented with sounds alone. As previously implied, this crossmodal activation appears to be the result of implicit associations of the two stimuli, likely driven by their spatiotemporal characteristics; it was observed after a relatively short period of exposure (~45 min) and lasted for a relatively long period after the initial exposure (~1 day). These results suggest that the previously reported findings may be directly rooted in the increased activity of the neurons occupying the primary visual cortex.

1. Introduction

Sensory processing of environmental stimuli starts at the level of specialized peripheral organs (e.g., skin, eyes, ears, etc.) and follows segregated information processing pathways in the central nervous system. To have coherent and unified percepts of multimodal events (e.g., containing sound and light as in the case of a moving car or a vocalizing conspecific), sensory information across these apparently divergent pathways needs to be integrated. Integration of information across two or more sensory channels involves multiple subcortical structures [1–4], as well as cortical regions (e.g., parietal cortex [5, 6], the superior temporal sulcus [7–9], and the insular cortex [10, 11]).

In more recent accounts of sensory processing, interactions between modality-specific channels are emphasized. For example, recent work shows that the activity of unisensory cortices can be under a cross-modal influence. For

example, recent studies show that a primary sensory cortex can be under inhibitory [12–17] or excitatory cross modal influence [18–21]. Findings from single-unit recordings of visual influence on early auditory cortical processing [22, 23] also demonstrate that activity in nominally unisensory auditory cortex can be modulated by the presence of a concurrent visual stimulus; similar data have been reported in human neuroimaging studies, where modulation of one sensory cortex occurs due to multisensory costimulation (for review see [24–26]).

In addition, it has been shown through several lines of evidence that a stimulus presented through only one sensory modality affects the processing and perception of a stimulus presented in another modality. The flash-beep illusion introduced by Shams et al. [27] is a clear example of such multisensory interactions whereby the *perceived* number of visual flashes appears to be positively linked to the *actual* number of simultaneous beeps for a single flash of light.

Subsequent work on the neurophysiological underpinnings of this illusion has revealed the involvement of the primary visual cortex (V1) and/or other early visual cortical areas [28–31]. These phenomena occur over very short time scales (typically a few tens/hundreds of milliseconds) and most likely reflect a process of integration of the information coming from the two modalities.

When the time scale of such interactions is expanded beyond minutes and hours, one comes across situations where a stimulus presented in only one sensory modality recruits regions pertaining to a different modality (such as in the case of lipreading [32–35] or the case where visual cortical areas have been shown to respond to auditory components of typically bimodal events with a close semantic relationship, such as tools and their sounds [7, 8] or voices and faces [36, 37]). In addition, a number of neuroimaging studies have also directly investigated the nature of cross-modal activity following learning or conditioning paradigms in which arbitrary pairings of unrelated auditory or visual stimuli [21, 38, 39] are shown to lead to cross-modal recruitment.

Immediate early genes (IEGs) are a group of genes that are transiently activated following sensory stimulation [40]. IEG *zif268* encodes a transcription factor that has a regulatory role in neuronal processes such as excitability, neurotransmitter release, and metabolism [40], and the time course of its mRNA and protein expression has been studied [41]. The inducible expression of IEG protein products is commonly used to map neural activity at the cellular level by immunohistochemistry (for extensive reviews see [40, 42–47]). One benefit of using *zif268* expression in activity mapping is that it has considerable staining reliability and availability of antibodies [48]. Furthermore, cellular resolution mapping provides a precise localization of neural activity over large areas of the cortex. This provides a framework for large-scale analysis with single-cell resolution, a combination that is difficult to achieve by any other method [48, 49]. Since analysis is performed postmortem, the technique permits the experimental animals to be treated with a flexible behavioral schedule. Unlike functional neuroimaging or electrophysiological techniques, a molecular mapping study requires little preparatory procedures. The animals are therefore permitted to behave more naturally in an unrestricted environment prior to experimentation [48, 49].

Molecular mapping can be used to reveal activation maps of brain areas in response to each component of a stimulus sequence. This allows for the visualization of segregated populations of neurons that are each responsive to a different part of the sequence [41]. Given that the time course of *zif268* induction has been determined, if an animal is exposed to a compound stimulus followed by euthanasia at a precise time point, one can establish the point within the sequence that a given population of neurons became active by assessing the level of *zif268* expression in the tissue. Based on the temporal properties of *zif268* induction, neurons that respond to the first part of the compound stimulus should contain detectable levels of the protein, whereas neurons responsive to the second part of the sequence should not

[41]. On this basis, if an animal is preexposed to the bimodal stimulus and subsequently experiences auditory followed by visual stimulation before euthanasia, *zif268* expression in the primary visual cortex in addition to the auditory cortex would constitute evidence for cross-modal recruitment.

A recent functional positron emission tomography (PET) study [50] has explored the circumstances under which cross-modal cortical recruitment can occur with unimodal stimuli in the absence of a semantic association, an explicit conditioning paradigm, or prolonged, habitual cooccurrence of bimodal stimuli. It was found that human subjects who had been preexposed to audiovisual stimuli showed increased cerebral blood flow in the primary visual cortex in response to the auditory component of the bimodal stimulus alone, whereas naïve subjects showed only modality-specific activation. The results indicate that inputs to the auditory system can drive activity in the primary visual cortex (V1) 1 day after brief exposure and persist for 16–24 hours [50]. However, due to the inherent limitations of correlating changes in blood flow to underlying neural activity as in the case of functional PET imaging, it still remains unclear whether or not the observed cross-modal phenomenon is directly linked to increased neural activity in area V1.

Thus, the goal of the present study was to investigate the previously described cross-modal recruitment of V1 in response to auditory stimuli using a more direct method of visualizing brain activity, namely, molecular activity mapping of the IEG *zif268*, in order to describe a more direct link between nonrelevant sensory input and cross-modal cortical activation.

2. Materials and Methods

2.1. Animals. The subjects were four adult vervet monkeys (*Chlorocebus sabaues*). Animal experimentation, conducted in accordance with the Animal Use Protocol of the McGill University Animal Care Committee, was performed at the Behavioural Sciences Foundation on the Island of St Kitts. This facility is fully accredited by the Canadian Council on Animal Care. Externalized brain tissue was transported back to McGill University where histological processing and data analysis were conducted.

2.2. Apparatus and Stimuli. Both auditory and visual stimuli were presented using a Sony 37" LCD Digital TV with integrated stereospeakers connected to a MacBook Air computer (Apple Inc.). Monkeys were seated in the primate chair facing the center of the monitor at a viewing distance of 60 cm.

Auditory (A) and visual (V) stimuli (Figure 1(a)) were each presented in the form of a sequence of five elements for a total of 2 s. However, the duration of the elements was determined randomly using Matlab software on each trial such that the total length of each trial did not exceed 2 s. Three seconds of silence followed each 2 s trial. The content of the auditory stimuli was white-noise bursts whereas the visual stimuli were made up of flashes of a random dot pattern. Each 2 s stimulus was presented from one of

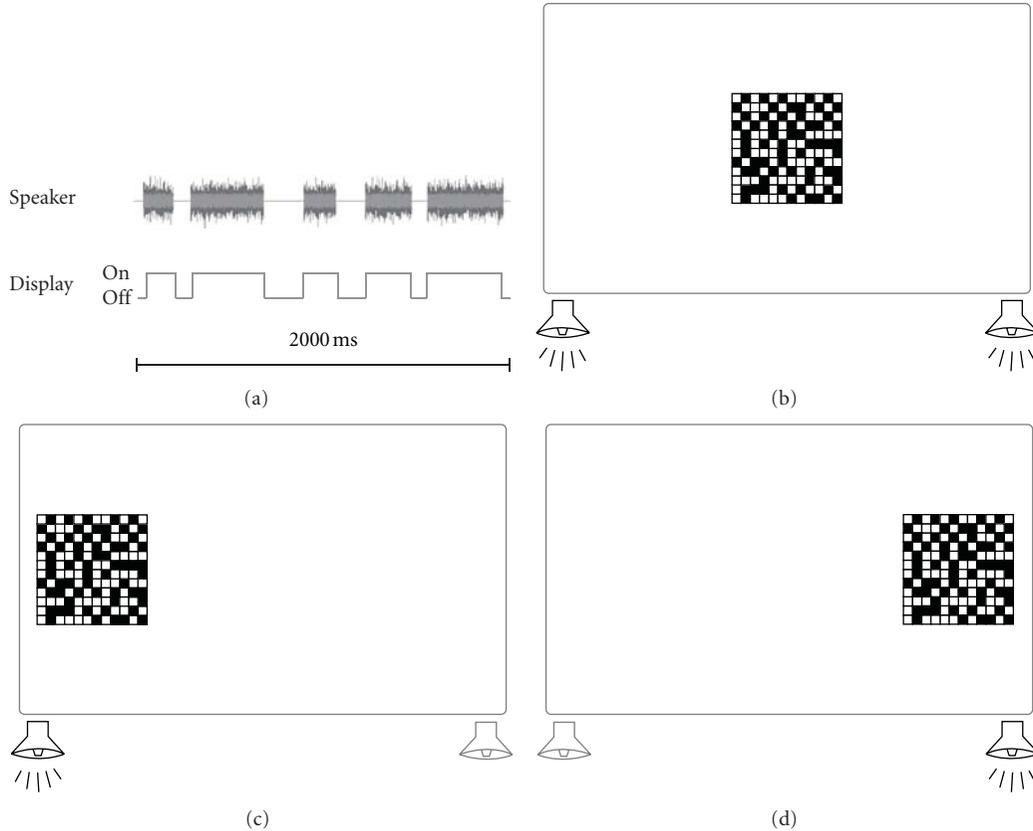


FIGURE 1: Schematics of stimuli and experimental apparatus. The stimuli presented were white-noise bursts and/or flashes of a single light source that lasted a total of 2000 msec (a), which were presented from the monitor and/or speakers at the centre (b), left (c), or right (d) side of the animals sensory space.

three discrete locations (left, center, or right; Figures 1(b)–1(d)) within the confines of the sensory space as defined by the limits of the monitor and location of speakers. Those auditory and visual stimuli were also paired together and presented to a subset of monkeys as a compound bimodal stimulus for 45 minutes in order to establish implicit associations between auditory and visual modalities based on the temporal and spatial attributes of the stimuli. This category of stimuli was used to visualize the cross-modal recruitment phenomenon previously reported in humans.

2.3. Stimulation Procedure. Each monkey was exposed to a sequence of A followed by V stimuli (or vice versa) in order to visualize zif268 expression in response to those stimuli in primary sensory areas of the brain. The total duration of the stimulus blocks was designed such that the first stimulus lasted 60 minutes followed by 30 minutes of exposure to stimuli of the other modality. The rationale behind the choice of those periods was based on the peak of zif268 protein (henceforth Zif268) expression, which occurs at 90 minutes following the onset of sensory stimulation. Thus, two animals received the auditory followed by visual stimulation sequence (i.e., AV), and the other two animals received the reverse sequence (i.e., VA). In addition, animals in each stimulation group were further divided into two

categories of Naïve (N) and Experienced (E), where N signifies the lack of experience of compound bimodal stimuli and E signifies the presence of such experience. Monkeys in group E received those compound stimuli 24 hours prior to receiving the AV or VA stimulation sequence and immediately before being euthanized for the purpose of visualizing Zif268 expression.

2.4. Animal Treatment and Tissue Collection. For 7 days prior to the start of this study, each day the animals experienced a two-hour habituation to the primate chair and the testing room that were subsequently used during the experiment. During these sessions they received orange- and lime-flavored juice as a reward. On the day of the experiment the monkeys were placed in the primate chair. Each animal was dark adapted while wearing a pair of foam earplugs for three hours to ensure baseline levels of Zif268 expression. Following stimulus presentation, animals received ketamine hydrochloride (10 mg/kg) sedation and were subsequently euthanized by an overdose of intravenously administered sodium pentobarbital (25 mg/kg), followed by transcardial perfusion of 0.1 M PBS to produce exsanguination. The brains were then externalized and sectioned along the coronal plane. Each hemisphere was blocked, and each block was flash-frozen in an isopentane bath cooled in a dry ice

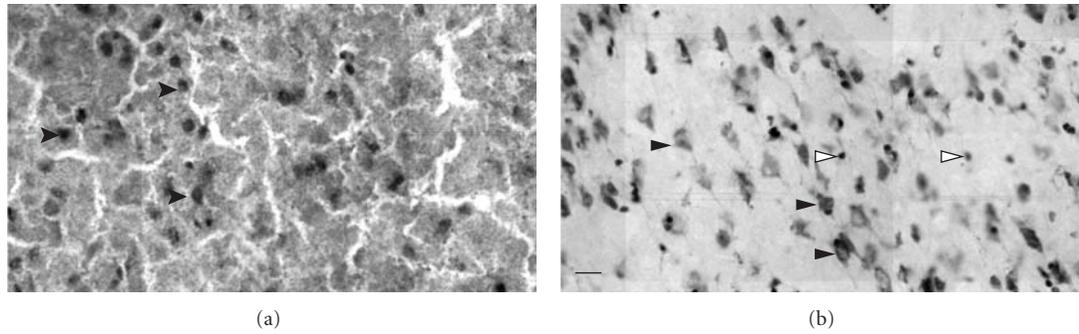


FIGURE 2: A representative sample of digitalized immunostained (a) and cresyl violet-stained (b) tissues. The black arrowheads show objects that were included in the count while the white arrowheads show objects that do not fit counting criteria and were discarded. The scale bar in panel (b) represents $20\ \mu\text{m}$.

chamber and maintained at -80°C . Twenty-micrometer-thick sections were cut from the frozen blocks at -20°C on a Leica CM3050 cryostat and mounted onto Vectabond (Vector Labs) subbed glass slides. The slide-mounted tissue sections were stored at -80°C until histological processing.

2.5. Immunohistochemistry. Slide-mounted twenty-micron fresh-frozen tissue sections were thawed on a slide warmer at 37°C for ~ 5 mins. A PAP pen was used to create a hydrophobic barrier surrounding the slide-mounted tissue in order to keep staining reagents localized on the tissue section. Sections were then fixed with a 4% paraformaldehyde-in-phosphate-buffered saline (PBS) solution for 10 minutes, followed by a 5-minute PBS rinse. Sections were then washed with 0.3% hydrogen peroxide in PBS for 15 minutes to block endogenous peroxidase activity. Following another 5-minute PBS rinse, sections were acetylated in 0.25% acetic anhydride in 10 mM triethanolamine (TEA) for 10 minutes each time. Sections were then rinsed in PBS for 5 minutes and then blocked for 30 minutes with a solution of PBS and 3% normal goat serum (NGS). Each section was incubated overnight at 4°C 1 mL of rabbit anti-Zif268 polyclonal antibody (courtesy of Bravo) solution at a concentration of 1 : 10,000. Next day, sections underwent three 10-minute PBS washes, followed by incubation in a secondary antibody solution (biotinylated goat-anti-rabbit antibody diluted 1 : 500 in PBS containing 3% NGS) for 1.5 hours. The sections were then given three consecutive 10-minute PBS washes before undergoing 1-hour incubation in avidin-biotin-conjugated horseradish peroxidase (Vectastain ABC kit) solution (1 : 500). Following three subsequent 10-minute washes in PBS, the sections were treated with a 3,3-diaminobenzidine (DAB) substrate kit. The sections were then rinsed in PBS three times for 5 minutes each and subsequently underwent dehydration in graded ethanol steps, cleared in xylene, and coverslipped with Permount.

2.6. Cresyl Violet Stain. Cresyl violet staining of Nissl bodies was conducted on adjacent tissue sections to those that were immunostained. The purpose of the Nissl stain was to reveal anatomical landmarks for delineating auditory and visual

cortical areas and to provide an estimate of the density of neurons in each brain area. The staining protocol was as follows. After removing the designated sections from freezer storage, they were left at room temperature to dry for 5 minutes. The sections then underwent graded ethanol dehydration followed by staining and rehydration. The slides were then coverslipped with Permount mounting medium and left to dry at room temperature under the fume hood.

2.7. Digitization of Histological Data. Following histological processing, all sections from the primary auditory and primary visual cortices were scanned using a MiraxDesk slide scanner and Mirax Viewer software (Carl Zeiss MicroImaging Inc, Thornwood, New York). Three frames were taken from each of the five scanned sections per brain area, with a sufficient number of high-magnification captures per frame (equivalent to the magnification using a 40x objective lens) to span all cortical layers. The necessary number of captures per frame depended on cortical thickness but ranged from three to six captures. Captures of the Nissl-stained scanned sections were taken from approximately the same segments of the brain areas of interest as on the corresponding immunostained scanned sections. A stereotaxic atlas was used as a reference for determining the boundaries of the areas of interest.

2.8. Cell Counting and Statistical Analyses. A counting frame with both inclusion and exclusion boundaries was fixed onto each captured frame. The counting frame area was approximately $310,000\ \text{pixels}^2$ which converts to $38,990\ \mu\text{m}^2$ and spanned the entire length of the scanned area. Manual counts of objects were performed in each counting frame, once for the immunostained nuclei and once for the Nissl-stained cell bodies in immediately adjacent sections. Figure 2 represents examples of counted objects. Criteria for Nissl cell counting were large endoplasmic staining with a visible nuclear envelope and one or more nucleoli. Objects such as glial cells that did not fit these criteria were discarded from the count. The density of cells was calculated by dividing the cell count by the area for each counting frame. The ratio of immunopositive neurons to the average number of neurons

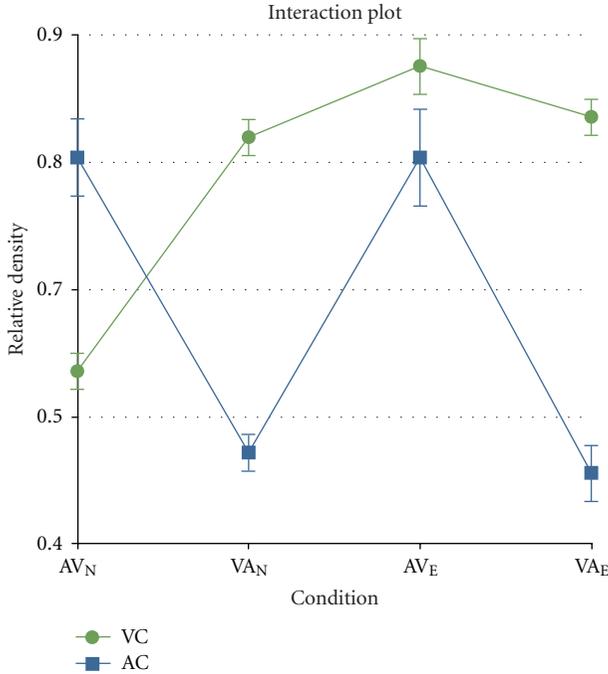


FIGURE 3: Interaction plot of the ANOVA conducted on cell counts. The primary visual cortex (V1) shows high expression of Zif268 when exposed to conditions VA_N, AV_E, and VA_E compared to baseline expression when exposed to condition VA_N. In contrast, the primary auditory cortex (A1) shows high protein expression only when exposed to conditions AV_N and AV_E and baseline expression when exposed to VA_N and VA_E. VA: visual stimulation followed by auditory stimulation; AV: auditory followed by visual stimulation; N: naïve group (no prior experience with the association of auditory and visual stimuli); E: experienced group (45 minutes of exposure to the association of auditory and visual stimuli, 24 hours prior to experiencing the stimulus sequence). Relative density measures are represented as Mean \pm SEM.

in the tissue section was calculated by dividing the density of immunopositive neurons by the density of Nissl-stained neurons in adjacent sections. A nonsignificant difference was found between the total cell densities of a given brain area across the four subjects. Counting was done for 40 frames from each brain area of each animal, for a total of 320 counted frames. The dependent variable was expressed as the resulting ratios and was transferred to ezANOVA statistical analysis software (<http://bit.ly/vZfGCO>) for the analysis of variance (ANOVA).

3. Results and Discussion

A mixed design 2-way ANOVA was performed on the ratio of cell densities of immunopositive neurons to total neurons, with Brain Region as the within-subjects variable and Condition as the between-subjects variable. The Condition variable contained four levels: AV_N, VA_N, AV_E, VA_E, where N represents naïve, E represents experienced, AV represents auditory followed by visual stimulation, and VA represents visual followed by auditory stimulation. The variable Brain Area contained two levels: AC (auditory cortex) and VC

(visual cortex). The ANOVA revealed a significant interaction between Brain Region and Condition ($F_{(3,156,0.05)} = 38.7$; $P < 0.000001$) as well as significant main effects of both Brain Area ($F_{(1,156,0.05)} = 44.6$; $P < 0.000001$) and Condition ($F_{(3,156,0.05)} = 14.4$; $P < 0.000001$). The interaction plot is summarized in Figure 3.

The analysis of Zif268 expression in the visual cortex was restricted to the striate cortex or V1 in order to be able to draw a direct comparison between our findings with those of Zangenehpour and Zatorre [50]. We first compared the modality-specific response of V1 between the two groups of animals (i.e., Experienced vs. Naïve) and found a nonsignificant difference between Zif268 expressions across the two conditions [VA_N (mean \pm SE = 0.82 ± 0.02) versus VA_E (mean \pm SE = 0.84 ± 0.02)]. This finding followed our expectation, as we had no *a priori* reason to anticipate a change in V1 activity in response to visual stimuli as a consequence of prior exposure to audiovisual stimuli. The result of this analysis also served as a valuable internal control because it demonstrates that despite individual differences, a great deal of consistency is observed in terms of the extent of cortical activity in response to visual stimuli.

We then focused our analyses any cross-modal effects in the visual cortex. We found that V1 expression of Zif268 in condition AV_E (mean \pm SE = 0.87 ± 0.03) was significantly higher than that found in condition AV_N (mean \pm SE = 0.57 ± 0.02 ; $t_{(78,0.05)} = 8.30$; $P < 0.0001$) and nonsignificantly different from that found in conditions VA_N and VA_E. The higher level of Zif268 expression in condition AV_E compared to AV_N suggests that the auditory component of the stimulation sequence (i.e., auditory followed by visual) was the driving force of V1 protein expression in condition AV_E, while it had little effect on the activity of V1 in condition AV_N. These analyses further revealed that the V1 of group E animals responded in the same manner to visual and auditory stimuli, while the V1 of group N animals responded in a modality-specific way to the same stimuli. This observation thus constitutes the main finding of our study, namely, that in addition to modality-specific activation of V1 and A1 in all conditions, V1 was crossmodally recruited by auditory stimuli in experienced but not naïve subjects. Figure 4 shows representative micrographs of the observed activation patterns.

The analysis of Zif268 expression in the auditory cortex was restricted to the core region (also referred to as A1, as defined by [51, 52], e.g.) in order to have an analogous framework for comparisons with V1. Auditory cortical expression of Zif268 in all experimental conditions was found to be modality specific. There was no significant difference between A1 expression of Zif268 in condition AV_N (mean \pm SE = 0.78 ± 0.04) and that of condition AV_E (mean \pm SE = 0.78 ± 0.05). Likewise, there was no significant difference between A1 expression of Zif268 in condition VA_N (mean \pm SE = 0.49 ± 0.02) and that of condition VA_E (mean \pm SE = 0.47 ± 0.03), suggesting that the auditory component of the stimulation sequence was the only driving force of Zif268 expression in A1. Figure 5 shows representative micrographs of Zif268 expression obtained from A1 of all four experimental conditions.

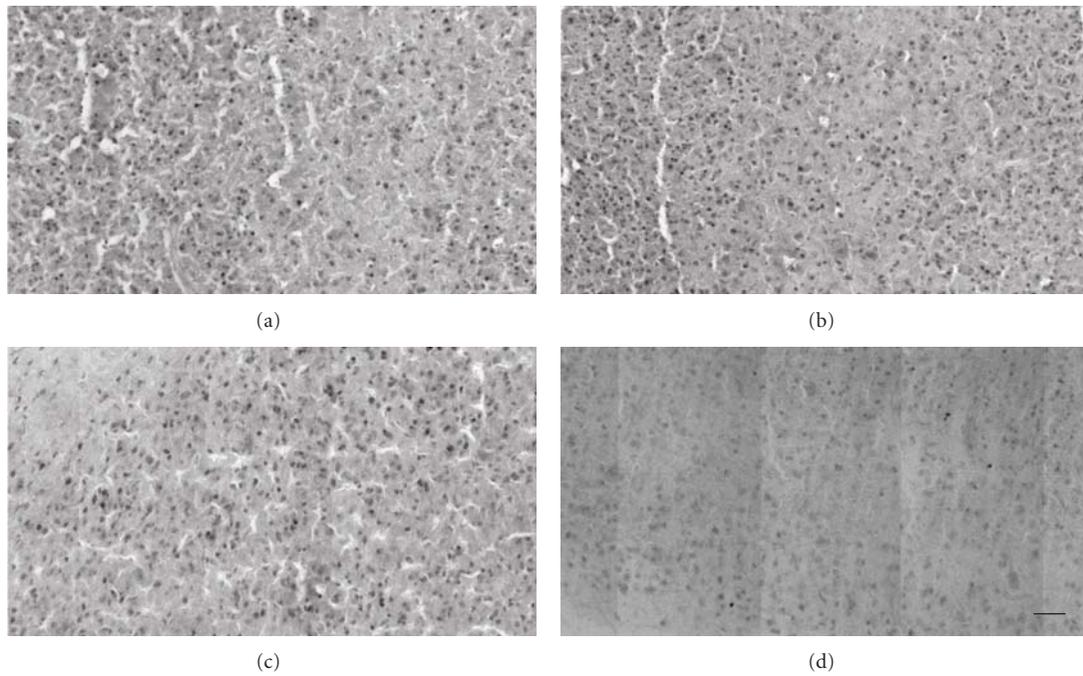


FIGURE 4: Sample micrographs of Zif268 immunoreactivity in the primary visual cortex (V1). There are a high number of Zif268 protein-positive neurons in V1 in conditions VA_E (a), AV_E (b), and VA_N (c), but not in condition AV_N (d). This observation implies that V1 neurons in the animals that were preexposed to audiovisual stimuli were recruited by *both* auditory *and* visual stimuli. The scale bar in (d) represents $50\ \mu\text{m}$.

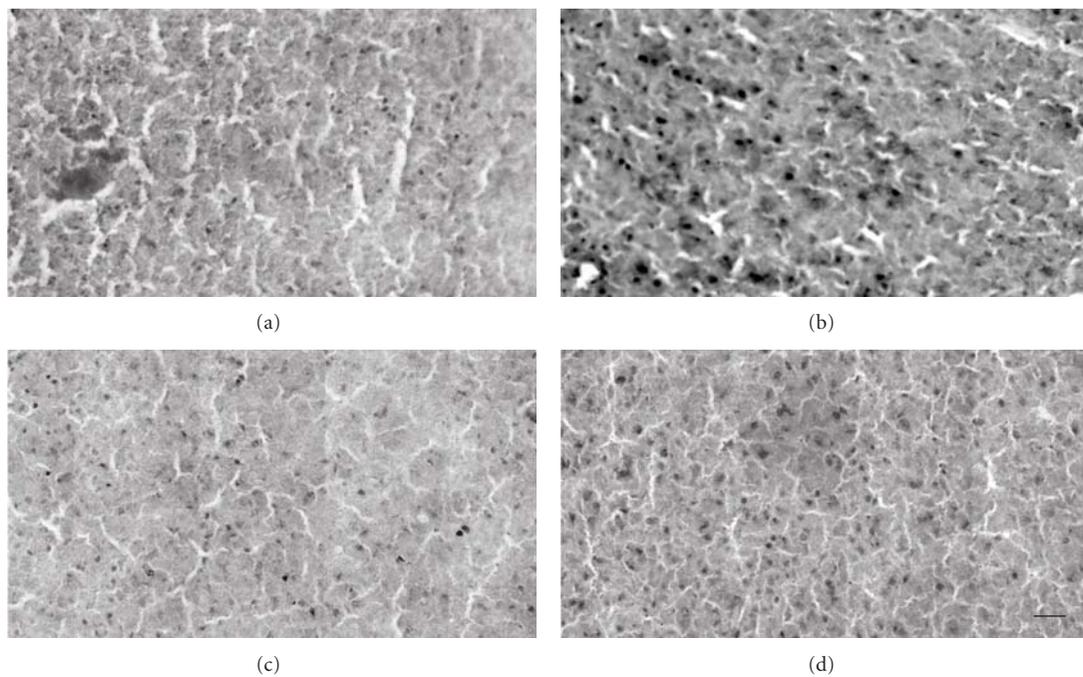


FIGURE 5: Sample micrographs of Zif268 immunoreactivity in the primary auditory cortex (A1). There are a low number of Zif268 protein-positive neurons in A1 under conditions VA_E (a) and VA_N (c). Conversely, protein expression appears to be much higher under conditions AV_E (b) and AV_N (d). Unlike in V1, neurons in A1 appear to be driven in a modality-specific manner (i.e., by auditory stimuli alone) irrespective of the preexposure to audiovisual stimuli. The scale bar in (d) represents $50\ \mu\text{m}$.

Thus far we have been able to find parallel evidence for cross-modal recruitment of visual cortex by auditory stimuli, *only* when those auditory stimuli were experienced *a priori* in the context of a compound audiovisual stimulus, using a nonhuman primate model. As in the Zangenehpour and Zatorre study [50], we did not observe symmetrical cross-modal recruitment; that is, the auditory cortex of the experienced subjects did not show a positive response to visual stimuli. Therefore, having demonstrated this phenomenon at cellular resolution in the nonhuman primate brain, we have provided converging evidence for the findings Zangenehpour and Zatorre in the human brain. The combined findings show that cross-modal recruitment can occur after brief exposure to an audiovisual event in the absence of semantic factors.

We have also replicated earlier findings in the rat brain [53] regarding the expression profile of Zif268 in the primary visual and auditory cortices following compound stimulation sequences. We confirmed the observations that as a result of auditory followed by visual stimulation, V1 will display baseline protein expression and A1 will display elevated protein expression and that the opposite pattern is obtained if the stimulation sequence is reversed. However, one important difference between our and those earlier findings is the amount of activity-induced protein expression compared to baseline. We found that protein expression in response to stimulation increased by a factor of 1.4 in V1 and by a factor of 1.6 in A1. This increase appears to be lower than the increase found in the rat brain [53], most likely because baseline protein expression in the vervet monkey brain is higher than that in the rat brain. Baseline Zif268 expression can be due to spontaneous translation or stimulus-driven expression caused by spontaneous neural activity [53]. It has previously been found that 30% of neurons in the rat visual cortex are Zif268 immunopositive at baseline [54], whereas we found this to be the case for 57% of neurons in the vervet monkey visual cortex. Although this could be due to interspecies variability, it is also plausible that the mere 3-hour sensory deprivation period in our study design contributed to high levels of baseline protein expression. Rodent studies that report lower baseline expression had sensory deprivation periods of several days to weeks [54, 55]. The sensory deprivation period in primate studies must be limited due to ethical and practical considerations.

A number of recent functional neuroimaging [32, 33, 35], event-related potential (ERP) recordings [56–60] and magnetoencephalography (MEG [61, 62]) experiments have shown the human auditory cortex to be a site of interaction of audiotactile and audiovisual information. Intracranial recording studies in monkeys [36, 63–67] support those noninvasive human studies by showing that the response of auditory cortical neurons may be influenced by visual and/or somatosensory information. Similarly, both early [68–70] and higher-level visual cortical areas [37, 71–79] have been shown to be under auditory and somatosensory influences. However, there are few studies showing that auditory stimuli alone result in activation of visual cortex (or vice versa) outside of situations involving explicit association

or habitual coexposure. In fact, there is considerable evidence for reciprocal inhibition across sensory cortices such that activity in one leads to suppression in the other. The present findings help to clarify the conditions under which cross-modal recruitment may be obtained.

A rich body of literature has been compiled around the topic of cross-modal plasticity in the context of structural manipulations of the nervous system in animal models and sensory deficits in humans. For example, surgical damage of ascending auditory pathways has been shown to lead to the formation of novel retinal projections into the medial geniculate nucleus and the auditory cortex [80–82]. Cross-modal plasticity has been documented in the human brain in the context of sensory deficit, such as the activation of visual cortical areas in blind subjects via tactile [83–87] or auditory tasks [88, 89], or auditory cortex recruitment by visual stimuli in deaf people [90–92]. In the present study, however, we observe a robust cross-modal recruitment of the primary visual cortex (V1) in the absence of similar deprivation-related reorganization. Others have shown the recruitment of extrastriate visual cortical regions after extensive training with visuohaptic object-related tasks [74] and in tactile discrimination of grating orientation [79] in normally sighted individuals. The present study documents that even *primary visual cortex* is subject to cross modal recruitment and that this can happen relatively rapidly. We did not observe symmetric cross-modal recruitment, since we did not detect any activity beyond baseline in A1 in response to the visual stimuli. Given the many studies reviewed above that have shown responsiveness of auditory regions to cross-modal inputs, however, we refrain from interpreting the lack of auditory recruitment as an indication that auditory cortical regions cannot be driven by nonauditory inputs under appropriate conditions.

There is one remaining question: how does V1 receive auditory information such that it is driven by sound? There are three plausible, and not necessarily mutually exclusive, scenarios for such activity to be mediated anatomically (as discussed in [9, 25, 26]): (i) auditory signals may be routed from auditory brainstem nuclei to subcortical visual structures (e.g., lateral geniculate nucleus) and thence to V1; (ii) cortical auditory signals may have influenced V1 indirectly through multimodal cortical regions; (iii) early auditory cortical regions may communicate directly with V1 via corticocortical pathways. There is evidence in support of direct, but sparse, anatomical links between the auditory and striate cortices of the nonhuman primate brain [22, 69, 93]. There is also evidence in support of various thalamic regions, such as the pulvinar [3, 4] and a number of other thalamic nuclei [4], exhibiting multisensory properties. Similar findings linking cortical parts of the brain, such as via the angular gyrus [94], STS [95], and the insular cortex [96], have been used to explain the observed cross-modal plasticity, such as those reported in sighted subjects [97]. Although our molecular mapping approach cannot be used to answer matters pertaining to connectivity directly, it can be combined with traditional tracing approaches in follow-up studies to help find a clearer answer to this question.

4. Conclusions

We used molecular activity mapping of the immediate early gene *zif268* to further study the nature of cross-modal recruitment of the visual cortex by auditory stimuli following a brief exposure to audiovisual events. When presented with only the auditory or visual components of the bimodal stimuli, naïve animals showed only modality-specific cortical activation. However, animals that had previously been exposed to a combination of auditory and visual stimuli showed increased number of active neurons in the primary visual cortex (V1) when presented with sounds alone. As previously implied, this cross-modal activation may be the result of implicit associations of the two stimuli, likely driven by their spatiotemporal characteristics; it was observed after a relatively short period of exposure (~45 min) and lasted for a relatively long period after the initial exposure (~1 day). These new findings suggest that the auditory and visual cortices interact far more extensively than typically assumed. Furthermore, they suggest that the previously reported findings may be directly rooted in the increased activity of the neurons occupying the primary visual cortex.

Acknowledgments

This work was supported by a start-up grant from the Faculty of Science at McGill University to SZ and infrastructure support from Behavioural Sciences Foundation (BSF), St Kitts, West Indies. The authors thank members of BSF staff, as well as Edward Wilson for their invaluable assistance in the primate handling and tissue collection. They also thank Frank Ervin and Roberta Palmour for their continued support.

References

- [1] B. E. Stein and M. A. Meredith, *The Merging of the Senses*, MIT Press, Cambridge, Mass, USA, 1993.
- [2] B. E. Stein, M. A. Meredith, W. S. Huneycutt, and L. McDade, "Behavioral indices of multisensory integration: orientation to visual cues is affected by auditory stimuli," *Journal of Cognitive Neuroscience*, vol. 1, no. 1, pp. 12–24, 1989.
- [3] C. Cappe, A. Morel, P. Barone, and E. M. Rouiller, "The thalamocortical projection systems in primate: an anatomical support for multisensory and sensorimotor interplay," *Cerebral Cortex*, vol. 19, no. 9, pp. 2025–2037, 2009.
- [4] T. A. Hackett, L. A. De La Mothe, I. Ulbert, G. Karmos, J. Smiley, and C. E. Schroeder, "Multisensory convergence in auditory cortex, II. Thalamocortical connections of the caudal superior temporal plane," *Journal of Comparative Neurology*, vol. 502, no. 6, pp. 924–952, 2007.
- [5] F. Bremmer, A. Schlack, J. R. Duhamel, W. Graf, and G. R. Fink, "Space coding in primate posterior parietal cortex," *NeuroImage*, vol. 14, no. 1, pp. S46–S51, 2001.
- [6] S. Nakashita, D. N. Saito, T. Kochiyama, M. Honda, H. C. Tanabe, and N. Sadato, "Tactile-visual integration in the posterior parietal cortex: a functional magnetic resonance imaging study," *Brain Research Bulletin*, vol. 75, no. 5, pp. 513–525, 2008.
- [7] M. S. Beauchamp, B. D. Argall, J. Bodurka, J. H. Duyn, and A. Martin, "Unraveling multisensory integration: patchy organization within human STS multisensory cortex," *Nature Neuroscience*, vol. 7, no. 11, pp. 1190–1192, 2004.
- [8] M. S. Beauchamp, K. E. Lee, J. V. Haxby, and A. Martin, "Parallel visual motion processing streams for manipulable objects and human movements," *Neuron*, vol. 34, no. 1, pp. 149–159, 2002.
- [9] T. Noesselt, J. W. Rieger, M. A. Schoenfeld et al., "Audiovisual temporal correspondence modulates human multisensory superior temporal sulcus plus primary sensory cortices," *Journal of Neuroscience*, vol. 27, no. 42, pp. 11431–11441, 2007.
- [10] R. B. Banati, G. W. Goerres, C. Tjoa, J. P. Aggleton, and P. Grasby, "The functional anatomy of visual-tactile integration in man: a study using positron emission tomography," *Neuropsychologia*, vol. 38, no. 2, pp. 115–124, 2000.
- [11] J. W. Lewis, M. S. Beauchamp, and E. A. Deyoe, "A comparison of visual and auditory motion processing in human cerebral cortex," *Cerebral Cortex*, vol. 10, no. 9, pp. 873–888, 2000.
- [12] J. A. Johnson and R. J. Zatorre, "Attention to simultaneous unrelated auditory and visual events: behavioral and neural correlates," *Cerebral Cortex*, vol. 15, no. 10, pp. 1609–1620, 2005.
- [13] J. A. Johnson and R. J. Zatorre, "Neural substrates for dividing and focusing attention between simultaneous auditory and visual events," *NeuroImage*, vol. 31, no. 4, pp. 1673–1681, 2006.
- [14] D. S. O'Leary, R. I. Block, J. A. Koeppel et al., "Effects of smoking marijuana on brain perfusion and cognition," *Neuropsychopharmacology*, vol. 26, no. 6, pp. 802–816, 2002.
- [15] P. W. R. Woodruff, R. R. Benson, P. A. Bandettini et al., "Modulation of auditory and visual cortex by selective attention is modality-dependent," *NeuroReport*, vol. 7, no. 12, pp. 1909–1913, 1996.
- [16] C. I. Petkov, X. Kang, K. Alho, O. Bertrand, E. W. Yund, and D. L. Woods, "Attentional modulation of human auditory cortex," *Nature Neuroscience*, vol. 7, no. 6, pp. 658–663, 2004.
- [17] R. Kawashima, B. T. O'Sullivan, and P. E. Roland, "Positron-emission tomography studies of cross-modality inhibition in selective attentional tasks: closing the "mind's eye"," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 92, no. 13, pp. 5969–5972, 1995.
- [18] G. A. Calvert, "Crossmodal processing in the human brain: insights from functional neuroimaging studies," *Cerebral Cortex*, vol. 11, no. 12, pp. 1110–1123, 2001.
- [19] R. Martuzzi, M. M. Murray, C. M. Michel et al., "Multisensory interactions within human primary cortices revealed by BOLD dynamics," *Cerebral Cortex*, vol. 17, no. 7, pp. 1672–1679, 2007.
- [20] J. Pekkola, V. Ojanen, T. Autti et al., "Primary auditory cortex activation by visual speech: an fMRI study at 3 T," *NeuroReport*, vol. 16, no. 2, pp. 125–128, 2005.
- [21] H. C. Tanabe, M. Honda, and N. Sadato, "Functionally segregated neural substrates for arbitrary audiovisual paired-association learning," *Journal of Neuroscience*, vol. 25, no. 27, pp. 6409–6418, 2005.
- [22] J. K. Bizley, F. R. Nodal, V. M. Bajo, I. Nelken, and A. J. King, "Physiological and anatomical evidence for multisensory interactions in auditory cortex," *Cerebral Cortex*, vol. 17, no. 9, pp. 2172–2189, 2007.
- [23] C. Kayser, C. I. Petkov, and N. K. Logothetis, "Visual modulation of neurons in auditory cortex," *Cerebral Cortex*, vol. 18, no. 7, pp. 1560–1574, 2008.
- [24] J. Driver and T. Noesselt, "Multisensory interplay reveals crossmodal influences on "sensory-specific" brain regions, neural responses, and judgments," *Neuron*, vol. 57, no. 1, pp. 11–23, 2008.

- [25] A. A. Ghazanfar and C. E. Schroeder, "Is neocortex essentially multisensory?" *Trends in Cognitive Sciences*, vol. 10, no. 6, pp. 278–285, 2006.
- [26] D. Senkowski, T. R. Schneider, J. J. Foxe, and A. K. Engel, "Crossmodal binding through neural coherence: implications for multisensory processing," *Trends in Neurosciences*, vol. 31, no. 8, pp. 401–409, 2008.
- [27] L. Shams, Y. Kamitani, and S. Shimojo, "Illusions: what you see is what you hear," *Nature*, vol. 408, no. 6814, p. 788, 2000.
- [28] J. Mishra, A. Martinez, and S. A. Hillyard, "Cortical processes underlying sound-induced flash fusion," *Brain Research*, vol. 1242, pp. 102–115, 2008.
- [29] J. Mishra, A. Martinez, T. J. Sejnowski, and S. A. Hillyard, "Early cross-modal interactions in auditory and visual cortex underlie a sound-induced visual illusion," *Journal of Neuroscience*, vol. 27, no. 15, pp. 4120–4131, 2007.
- [30] S. Watkins, L. Shams, O. Josephs, and G. Rees, "Activity in human V1 follows multisensory perception," *NeuroImage*, vol. 37, no. 2, pp. 572–578, 2007.
- [31] S. Watkins, L. Shams, S. Tanaka, J. D. Haynes, and G. Rees, "Sound alters activity in human V1 in association with illusory visual perception," *NeuroImage*, vol. 31, no. 3, pp. 1247–1256, 2006.
- [32] G. A. Calvert, E. T. Bullmore, M. J. Brammer et al., "Activation of auditory cortex during silent lipreading," *Science*, vol. 276, no. 5312, pp. 593–596, 1997.
- [33] J. Pekkola, V. Ojanen, T. Autti et al., "Primary auditory cortex activation by visual speech: an fMRI study at 3 T," *NeuroReport*, vol. 16, no. 2, pp. 125–128, 2005.
- [34] J. Besle, C. Fischer, A. Bidet-Caulet, F. Lecaigard, O. Bertrand, and M. H. Giard, "Visual activation and audiovisual interactions in the auditory cortex during speech perception: intracranial recordings in humans," *Journal of Neuroscience*, vol. 28, no. 52, pp. 14301–14310, 2008.
- [35] N. Van Atteveldt, E. Formisano, R. Goebel, and L. Blomert, "Integration of letters and speech sounds in the human brain," *Neuron*, vol. 43, no. 2, pp. 271–282, 2004.
- [36] A. A. Ghazanfar, J. X. Maier, K. L. Hoffman, and N. K. Logothetis, "Multisensory integration of dynamic faces and voices in rhesus monkey auditory cortex," *Journal of Neuroscience*, vol. 25, no. 20, pp. 5004–5012, 2005.
- [37] K. von Kriegstein, A. Kleinschmidt, P. Sterzer, and A. L. Giraud, "Interaction of face and voice areas during speaker recognition," *Journal of Cognitive Neuroscience*, vol. 17, no. 3, pp. 367–376, 2005.
- [38] A. R. McIntosh, R. E. Cabeza, and N. J. Lobaugh, "Analysis of neural interactions explains the activation of occipital cortex by an auditory stimulus," *Journal of Neurophysiology*, vol. 80, no. 5, pp. 2790–2796, 1998.
- [39] M. Meyer, S. Baumann, S. Marchina, and L. Jancke, "Hemodynamic responses in human multisensory and auditory association cortex to purely visual stimulation," *BMC Neuroscience*, vol. 8, article 14, 2007.
- [40] T. Terleph and L. Tremere, "The use of immediate early genes as mapping tools for neuronal activation: concepts and methods," in *Immediate Early Genes in Sensory Processing, Cognitive Performance and Neurological Disorders*, R. Pinaud and L. Tremere, Eds., pp. 1–10, Springer, New York, NY, USA, 2009.
- [41] S. Zangenehpour and A. Chaudhuri, "Differential induction and decay curves of c-fos and zif268 revealed through dual activity maps," *Molecular Brain Research*, vol. 109, no. 1-2, pp. 221–225, 2002.
- [42] A. Chaudhuri, "Neural activity mapping with inducible transcription factors," *NeuroReport*, vol. 8, no. 13, pp. 3–7, 1997.
- [43] R. Farivar, S. Zangenehpour, and A. Chaudhuri, "Cellular-resolution activity mapping of the brain using immediate-early gene expression," *Frontiers in Bioscience*, vol. 9, pp. 104–109, 2004.
- [44] R. K. Filipkowski, "Inducing gene expression in barrel cortex—focus on immediate early genes," *Acta Neurobiologiae Experimentalis*, vol. 60, no. 3, pp. 411–418, 2000.
- [45] J. F. Guzowski, "Insights into immediate-early gene function in hippocampal memory consolidation using antisense oligonucleotide and fluorescent imaging approaches," *Hippocampus*, vol. 12, no. 1, pp. 86–104, 2002.
- [46] T. Herdegen and J. D. Leah, "Inducible and constitutive transcription factors in the mammalian nervous system: control of gene expression by Jun, Fos and Krox, and CREB/ATF proteins," *Brain Research Reviews*, vol. 28, no. 3, pp. 370–490, 1998.
- [47] L. Kaczmarek and A. Chaudhuri, "Sensory regulation of immediate-early gene expression in mammalian visual cortex: implications for functional mapping and neural plasticity," *Brain Research Reviews*, vol. 23, no. 3, pp. 237–256, 1997.
- [48] R. Farivar, S. Zangenehpour, and A. Chaudhuri, "Cellular-resolution activity mapping of the brain using immediate-early gene expression," *Frontiers in Bioscience*, vol. 9, pp. 104–109, 2004.
- [49] S. Zangenehpour and A. Chaudhuri, "Patchy organization and asymmetric distribution of the neural correlates of face processing in monkey inferotemporal cortex," *Current Biology*, vol. 15, no. 11, pp. 993–1005, 2005.
- [50] S. Zangenehpour and R. J. Zatorre, "Crossmodal recruitment of primary visual cortex following brief exposure to bimodal audiovisual stimuli," *Neuropsychologia*, vol. 48, no. 2, pp. 591–600, 2010.
- [51] J. P. Rauschecker and S. K. Scott, "Maps and streams in the auditory cortex: nonhuman primates illuminate human speech processing," *Nature Neuroscience*, vol. 12, no. 6, pp. 718–724, 2009.
- [52] L. M. Romanski and B. B. Averbeck, "The primate cortical auditory system and neural representation of conspecific vocalizations," *Annual Review of Neuroscience*, vol. 32, pp. 315–346, 2009.
- [53] S. Zangenehpour and A. Chaudhuri, "Neural activity profiles of the neocortex and superior colliculus after bimodal sensory stimulation," *Cerebral Cortex*, vol. 11, no. 10, pp. 924–935, 2001.
- [54] P. F. Worley, B. A. Christy, Y. Nakabeppu, R. V. Bhat, A. J. Cole, and J. M. Baraban, "Constitutive expression of zif268 in neocortex is regulated by synaptic activity," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 88, no. 12, pp. 5106–5110, 1991.
- [55] S. Zangenehpour and A. Chaudhuri, "Neural activity profiles of the neocortex and superior colliculus after bimodal sensory stimulation," *Cerebral Cortex*, vol. 11, no. 10, pp. 924–935, 2001.
- [56] J. Besle, A. Fort, C. Delpuech, and M. H. Giard, "Bimodal speech: early suppressive visual effects in human auditory cortex," *European Journal of Neuroscience*, vol. 20, no. 8, pp. 2225–2234, 2004.
- [57] J. J. Foxe, I. A. Morocz, M. M. Murray, B. A. Higgins, D. C. Javitt, and C. E. Schroeder, "Multisensory auditory-somatosensory interactions in early cortical processing

- revealed by high-density electrical mapping,” *Cognitive Brain Research*, vol. 10, no. 1-2, pp. 77–83, 2000.
- [58] M. H. Giard and F. Peronnet, “Auditory-visual integration during multimodal object recognition in humans: a behavioral and electrophysiological study,” *Journal of Cognitive Neuroscience*, vol. 11, no. 5, pp. 473–490, 1999.
- [59] S. Molholm, W. Ritter, M. M. Murray, D. C. Javitt, C. E. Schroeder, and J. J. Foxe, “Multisensory auditory-visual interactions during early sensory processing in humans: a high-density electrical mapping study,” *Cognitive Brain Research*, vol. 14, no. 1, pp. 115–128, 2002.
- [60] V. van Wassenhove, K. W. Grant, and D. Poeppel, “Visual speech speeds up the neural processing of auditory speech,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 102, no. 4, pp. 1181–1186, 2005.
- [61] R. Gobbelé, M. Schürmann, N. Forss, K. Juottonen, H. Buchner, and R. Hari, “Activation of the human posterior parietal and temporoparietal cortices during audiotactile interaction,” *NeuroImage*, vol. 20, no. 1, pp. 503–511, 2003.
- [62] B. Lütkenhöner, C. Lammertmann, C. Simões, and R. Hari, “Magnetoencephalographic correlates of audiotactile interaction,” *NeuroImage*, vol. 15, no. 3, pp. 509–522, 2002.
- [63] M. Brosch, E. Selezneva, and H. Scheich, “Nonauditory events of a behavioral procedure activate auditory cortex of highly trained monkeys,” *Journal of Neuroscience*, vol. 25, no. 29, pp. 6797–6806, 2005.
- [64] K. M. G. Fu, T. A. Johnston, A. S. Shah et al., “Auditory cortical neurons respond to somatosensory stimulation,” *Journal of Neuroscience*, vol. 23, no. 20, pp. 7510–7515, 2003.
- [65] C. Kayser, C. I. Petkov, M. Augath, and N. K. Logothetis, “Integration of touch and sound in auditory cortex,” *Neuron*, vol. 48, no. 2, pp. 373–384, 2005.
- [66] C. E. Schroeder and J. J. Foxe, “The timing and laminar profile of converging inputs to multisensory areas of the macaque neocortex,” *Cognitive Brain Research*, vol. 14, no. 1, pp. 187–198, 2002.
- [67] C. E. Schroeder, R. W. Lindsley, C. Specht, A. Marcovici, J. F. Smiley, and D. C. Javitt, “Somatosensory input to auditory association cortex in the macaque monkey,” *Journal of Neurophysiology*, vol. 85, no. 3, pp. 1322–1327, 2001.
- [68] F. Morrell, “Visual system’s view of acoustic space,” *Nature*, vol. 238, no. 5358, pp. 44–46, 1972.
- [69] A. Falchier, S. Clavagnier, P. Barone, and H. Kennedy, “Anatomical evidence of multimodal integration in primate striate cortex,” *Journal of Neuroscience*, vol. 22, no. 13, pp. 5749–5759, 2002.
- [70] K. S. Rockland and H. Ojima, “Multisensory convergence in calcarine visual areas in macaque monkey,” *International Journal of Psychophysiology*, vol. 50, no. 1-2, pp. 19–26, 2003.
- [71] J. R. Gibson and J. H. R. Maunsell, “Sensory modality specificity of neural activity related to memory in visual cortex,” *Journal of Neurophysiology*, vol. 78, no. 3, pp. 1263–1275, 1997.
- [72] A. Poremba, R. C. Saunder, A. M. Crane, M. Cook, L. Sokoloff, and M. Mishkin, “Functional mapping of the primate auditory system,” *Science*, vol. 299, no. 5606, pp. 568–572, 2003.
- [73] K. H. Pribram, B. S. Rosner, and W. A. Rosenblith, “Electrical responses to acoustic clicks in monkey: extent of neocortex activated,” *Journal of neurophysiology*, vol. 17, no. 4, pp. 336–344, 1954.
- [74] A. Amedi, R. Malach, T. Hendler, S. Peled, and E. Zohary, “Visuo-haptic object-related activation in the ventral visual pathway,” *Nature Neuroscience*, vol. 4, no. 3, pp. 324–330, 2001.
- [75] R. Blake, K. V. Sobel, and T. W. James, “Neural synergy between kinetic vision and touch,” *Psychological Science*, vol. 15, no. 6, pp. 397–402, 2004.
- [76] M. C. Hagen, O. Franzén, F. McGlone, G. Essick, C. Dancer, and J. V. Pardo, “Tactile motion activates the human middle temporal/V5 (MT/V5) complex,” *European Journal of Neuroscience*, vol. 16, no. 5, pp. 957–964, 2002.
- [77] T. W. James, G. K. Humphrey, J. S. Gati, P. Servos, R. S. Menon, and M. A. Goodale, “Haptic study of three-dimensional objects activates extrastriate visual areas,” *Neuropsychologia*, vol. 40, no. 10, pp. 1706–1714, 2002.
- [78] P. Pietrini, M. L. Furey, E. Ricciardi et al., “Beyond sensory images: object-based representation in the human ventral pathway,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 101, no. 15, pp. 5658–5663, 2004.
- [79] K. Sathian and A. Zangaladze, “Feeling with the mind’s eye: contribution of visual cortex to tactile perception,” *Behavioural Brain Research*, vol. 135, no. 1-2, pp. 127–132, 2002.
- [80] M. Sur, P. E. Garraghty, and A. W. Roe, “Experimentally induced visual projections into auditory thalamus and cortex,” *Science*, vol. 242, no. 4884, pp. 1437–1441, 1988.
- [81] D. O. Frost, “Orderly anomalous retinal projections to the medial geniculate, ventrobasal, and lateral posterior nuclei of the hamster,” *Journal of Comparative Neurology*, vol. 203, no. 2, pp. 227–256, 1981.
- [82] D. O. Frost, D. Boire, G. Gingras, and M. Ptito, “Surgically created neural pathways mediate visual pattern discrimination,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 20, pp. 11068–11073, 2000.
- [83] C. Büchel, C. Price, R. S. J. Frackowiak, and K. Friston, “Different activation patterns in the visual cortex of late and congenitally blind subjects,” *Brain*, vol. 121, no. 3, pp. 409–419, 1998.
- [84] H. Burton, A. Z. Snyder, T. E. Conturo, E. Akbudak, J. M. Ollinger, and M. E. Raichle, “Adaptive changes in early and late blind: a fMRI study of Braille reading,” *Journal of Neurophysiology*, vol. 87, no. 1, pp. 589–607, 2002.
- [85] L. G. Cohen, R. A. Weeks, N. Sadato et al., “Period of susceptibility for cross-modal plasticity in the blind,” *Annals of Neurology*, vol. 45, pp. 451–460, 1999.
- [86] N. Sadato, A. Pascual-Leone, J. Grafman et al., “Activation of the primary visual cortex by Braille reading in blind subjects,” *Nature*, vol. 380, no. 6574, pp. 526–528, 1996.
- [87] M. Ptito, S. M. Moesgaard, A. Gjedde, and R. Kupers, “Cross-modal plasticity revealed by electrotactile stimulation of the tongue in the congenitally blind,” *Brain*, vol. 128, no. 3, pp. 606–614, 2005.
- [88] R. Weeks, B. Horwitz, A. Aziz-Sultan et al., “A positron emission tomographic study of auditory localization in the congenitally blind,” *Journal of Neuroscience*, vol. 20, no. 7, pp. 2664–2672, 2000.
- [89] F. Gougoux, R. J. Zatorre, M. Lassonde, P. Voss, and F. Lepore, “A functional neuroimaging study of sound localization: visual cortex activity predicts performance in early-blind individuals,” *PLoS Biology*, vol. 3, no. 2, article e27, 2005.
- [90] E. M. Finney, B. A. Clementz, G. Hickok, and K. R. Dobkins, “Visual stimuli activate auditory cortex in deaf subjects: evidence from MEG,” *NeuroReport*, vol. 14, no. 11, pp. 1425–1427, 2003.
- [91] E. M. Finney, I. Fine, and K. R. Dobkins, “Visual stimuli activate auditory cortex in the deaf,” *Nature Neuroscience*, vol. 4, no. 12, pp. 1171–1173, 2001.

- [92] L. A. Petitto, R. J. Zatorre, K. Gauna, E. J. Nikelski, D. Dostie, and A. C. Evans, "Speech-like cerebral activity in profoundly deaf people processing signed languages: implications for the neural basis of human language," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 25, pp. 13961–13966, 2000.
- [93] S. Clavagnier, A. Falchier, and H. Kennedy, "Long-distance feedback projections to area V1: implications for multisensory integration, spatial awareness, and visual consciousness," *Cognitive, Affective and Behavioral Neuroscience*, vol. 4, no. 2, pp. 117–126, 2004.
- [94] B. Horwitz, J. M. Rumsey, and B. C. Donohue, "Functional connectivity of the angular gyrus in normal reading and dyslexia," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 95, no. 15, pp. 8939–8944, 1998.
- [95] C. Cappe and P. Barone, "Heteromodal connections supporting multisensory integration at low levels of cortical processing in the monkey," *European Journal of Neuroscience*, vol. 22, no. 11, pp. 2886–2902, 2005.
- [96] J. F. Smiley, T. A. Hackett, I. Ulbert et al., "Multisensory convergence in auditory cortex, I. Cortical connections of the caudal superior temporal plane in macaque monkeys," *Journal of Comparative Neurology*, vol. 502, no. 6, pp. 894–923, 2007.
- [97] A. Amedi, G. Jacobson, T. Hendler, R. Malach, and E. Zohary, "Convergence of visual and tactile shape processing in the human lateral occipital complex," *Cerebral Cortex*, vol. 12, no. 11, pp. 1202–1212, 2002.

Clinical Study

Increased BOLD Variability in the Parietal Cortex and Enhanced Parieto-Occipital Connectivity during Tactile Perception in Congenitally Blind Individuals

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Received 20 January 2012; Revised 6 April 2012; Accepted 18 April 2012

Academic Editor: Ron Kupers

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Previous studies in early blind individuals posited a possible role of parieto-occipital connections in conveying nonvisual information to the visual occipital cortex. As a consequence of blindness, parietal areas would thus become able to integrate a greater amount of multimodal information than in sighted individuals. To verify this hypothesis, we compared fMRI-measured BOLD signal temporal variability, an index of efficiency in functional information integration, in congenitally blind and sighted individuals during tactile spatial discrimination and motion perception tasks. In both tasks, the BOLD variability analysis revealed many cortical regions with a significantly greater variability in the blind as compared to sighted individuals, with an overlapping cluster located in the left inferior parietal/anterior intraparietal cortex. A functional connectivity analysis using this region as seed showed stronger correlations in both tasks with occipital areas in the blind as compared to sighted individuals. As BOLD variability reflects neural integration and processing efficiency, these cross-modal plastic changes in the parietal cortex, even if described in a limited sample, reinforce the hypothesis that this region may play an important role in processing nonvisual information in blind subjects and act as a hub in the cortico-cortical pathway from somatosensory cortex to the reorganized occipital areas.

1. Introduction

The human cerebral cortex is capable of a high degree of plasticity, a phenomenon based on both functional and structural modifications that allow the brain to adapt to environmental changes as well as to physiological or pathological conditions that may affect the individual [1]. According to this definition, an alteration during brain development may lead to significant changes in brain functional response and network organization as compared to normally developed brains. In this perspective, the study of early sensory deprivation has emerged as an interesting field of research in neuroscience since it represents an exceptional condition to assess; on one side, to what extent the development of the brain functional architecture is independent from that given sensory experience, for example, vision (for a recent critical overview see [2]), on the other side, the potentialities

of neural plasticity in reorganizing brain regions primarily affected by sensory deprivation (recently reviewed in [3, 4]). In particular, studies on the congenital lack of vision or its loss at later stages in life have investigated how the absence of vision affects the functional and structural organization of the brain, and which modifications occur in the visual cortical areas as a consequence of the lack of any retinal input [2, 5, 6].

The absence of inputs from the retina since birth induces a cross-modal plastic reorganization in early visual brain areas and a functional rearrangement of their afferent and efferent connections [1, 2, 4, 5, 7, 8]. These primary visual areas are recruited in blind individuals to process stimuli conveyed by nonvisual sensory modalities, that is, the tactile, auditory, and olfactory senses [6, 9–13]. Interestingly, the activation of specific areas in the occipital cortex of blind individuals is not merely an epiphenomenon but rather is

fundamental to the new sensory processing, as virtual functional lesions of these areas via transcranial magnetic stimulation (TMS) impair nonvisual performances, such as tactile perception, verb generation, or Braille reading [4, 14–16].

In addition to these cross-modal plastic modifications, the combined study of congenitally blind and sighted individuals also has demonstrated that cortical areas in the ventral and dorsal visual pathways are able to process sensory information regardless of the sensory modality through which such information has been acquired [2, 17]. In fact, while the activation of visual areas during non-visual processing could be ascribed to a visually based imagery in sighted individuals, the observation of an identical response pattern in a group of congenitally blind individuals, who by definition lack vision since birth and therefore do not possess any visually based mental imagery, indicates that these *supramodal* brain regions rely on a more abstract representation of the perceived stimuli, as, for example, in the cases of object category recognition [18, 19], spatial representation [20–24], or motion discrimination [25–27]. While plastic modifications do take place in the blind brain and lead visual areas that are *unimodal* in nature to process stimuli carried by different sensory modalities (*cross-modal plasticity*), at the same time *supramodal* areas develop within visual cortical regions that are ordinarily able to process also non-visual information both in sighted and blind individuals [2]. This more abstract nature of functional cortical organization may enable congenitally blind individuals to acquire knowledge, form mental representations of and interact effectively with an external world that they have never seen. In addition, the concept of *supramodal* organization has been recently extended much beyond the “what” and “where” visual pathways to brain areas associated with other cognitive and affective functions [2].

These observations have raised the question of which neural pathways are responsible for conveying non-visual sensory information to the *supramodal* and to the *cross-modal* specific areas within the visual cortex in congenitally blind individuals [2, 6]. It has been suggested that a rearrangement, or the potentiation, of preexisting cortico-cortical connections, such as in the case of a parieto-occipital pathway for tactile perception, may play a fundamental role in visual cortex recruitment in the blind brain [1, 2, 4, 6]. Consistent with this hypothesis, blind individuals show a specific functional association between parietal and occipital areas as compared to sighted subjects during tactile tasks [16, 28, 29], suggesting that parietal cortex may become a central hub for a more efficient exchange of multimodal information between somatosensory and occipital cortices in the absence of any visual experience.

To test this hypothesis we used a new measure, namely, blood-oxygenation-level-dependent (BOLD) signal variability, that has been recently proposed as an index of brain operative efficiency [30]. Indeed, as shown in some earlier experiments that employed EEG and MEG [31, 32], moment-to-moment variability in brain activity increases with the functional complexity of the cortical networks that subserve

a specific function. Thus, local changes in BOLD variability may represent an expression of an increase in the amount of information processed consequent to modifications in the specific functional network configuration [32].

For this reason, we hypothesized that areas within the parietal cortex, because of their enhanced multimodal information processing in congenitally blind as compared to sighted individuals, would show increased BOLD signal variability during tactile processing and a stronger functional correlation with occipital areas, consequent to cross-modal plastic modifications. To this aim, we performed a mean squared successive difference (MSSD) analysis [33, 34] to estimate BOLD temporal variability on data previously collected during a tactile spatial discrimination [20, 24] and a tactile motion perception task [26, 27]. In addition, differences in functional connectivity (FC) of brain areas showing an increased MSSD were also measured in sighted and congenitally blind individuals.

2. Methods

2.1. Subjects. Seven sighted (2 females, 29 ± 3 yrs) and 4 blind (1 female, 35 ± 15 yrs) right-handed healthy volunteers participated in the tactile spatial discrimination experiment. A partially distinct group of seven sighted (2 females, 27 ± 2 yrs) and four blind (1 female, 37 ± 14 yrs) individuals participated in the tactile motion perception protocol. All blind individuals were blind since birth, except one who became blind within the first two years of life and had no recollection of any visual experience (causes of blindness: congenital glaucoma, retinopathy of prematurity, and congenital optic atrophy). One sighted and two blind volunteers (including the early blind individual) participated in both experiments. All subjects received medical, neurological, and psychiatric examinations and a structural magnetic resonance imaging (MRI) brain scan to exclude any disorder that could affect brain function (other than blindness in the blind group). No subject was taking any psychotropic medication. All subjects gave their written informed consent after the study procedures, and risks involved had been explained (protocol no. 1616/2003 approved by the Ethical Committee of the University of Pisa).

2.2. Image Acquisition. We used fMRI to measure brain activity while subjects performed the two experimental paradigms described below. Gradient echo echoplanar images (GRE-EPI-) were acquired with a GE Signa 1.5-T scanner (General Electric, Milwaukee, WI) using the following parameters: repetition time = 3000 ms, 22–26 axial slices, slice thickness = 5 mm, field of view = 24 cm, echo time = 40 ms, flip angle = 90, image plane resolution = 64×64 pixels. Voxels were $3.75 \times 3.75 \times 5$ mm. High-resolution T1-weighted spoiled gradient recall images were obtained for each subject to provide detailed brain anatomy.

2.3. Tactile Spatial Discrimination. Tactile stimuli were wooden squares and cubes with three or five Velcro-covered

target-squares/cubes [24]. Each matrix was randomly presented to the subjects by using a wooden pole with a Plexiglas platform on one end upon which the wooden matrix was attached with Velcro. In this study, we used two-dimensional (2D) 5×5 matrices and three-dimensional (3D) $3 \times 3 \times 3$ matrices, comparable in terms of number of targets and number of potential combinations of target location. In order to control for any sequence effects, the presentation of different matrices (2D versus 3D) and number of targets (3 versus 5), as well as the foot code response, were randomized within and between subjects. Each stimulus was created in order to be different from the others, even if translated or rotated. Matrices were presented for 10 s during the tactile task, with an interstimulus interval of 5 s. Each time series consisted of 16 consecutive matrices with three/five targets in counterbalanced order. Subjects were instructed to explore the matrices with both hands, to generate a mental image of the perceived stimulus and to maintain it in memory for a comparison with the next stimulus. During baseline and interstimuli periods, subjects were asked to rest their arms along their side and keep them still. Sighted subjects were required to keep their eyes closed while performing the task. During the one-back tactile recognition task, volunteers tactily explored 16 sequential matrices in 6 to 8 experimental runs and indicated whether the last matrix was the same or different as compared to the previous one, by pressing foot pedals with the right (e.g., same) or left (e.g., different) foot.

2.4. Tactile Motion Perception. Tactile stimuli were moving or static Braille-like dot patterns (diameter = 1 mm, height = 1 mm) randomly displaced on a plastic flat surface. The average distance was 9 mm so the stimuli could not recall any letter of the Braille alphabet in blind subjects. Two types of motion were used during the task: horizontal translation (left to right and right to left at about 2.2 cm/s) and rotation (clockwise and counterclockwise at about $93.5^\circ/s$). Tactile stimuli were randomly presented using an MRI compatible device on a polystyrene table placed over the subjects' legs. Participants' hands lay on the table with the index and middle fingers touching the plastic surface with dot patterns. Moving stimuli were presented in 8 to 40 s blocks separated by intervals with static stimuli of varying duration (11 ± 10 s). Type of movement, direction of movement, and side of stimulation (right hand or left hand) were randomized and counterbalanced within and across subjects. Each time series began and ended with 30 s of static stimuli [26, 27].

2.5. Data Preprocessing. We used AFNI and SUMA software packages to preprocess, analyze, and view functional imaging data (<http://afni.nimh.nih.gov/afni/> [35]). Acquired raw data were reconstructed, coregistered to the volume collected nearest in time to the high-resolution anatomy, phase-shifted using Fourier transformation to correct for slice acquisition time, spatially smoothed (FWHM = 8 mm), and normalized to estimate the percent signal change at each time point. Individual preprocessed data were then transformed into the Talairach and Tournoux Atlas [36] coordinate system

and resampled into 3 mm^3 voxels. Finally, voxel time series were further adjusted by regressing out motion correction parameters, a polynomial function modeling the BOLD drifting effect and white matter (WM) and cerebrospinal fluid (CSF) time series [30]. For each experiment WM and CSF time courses were extracted from two single voxels, respectively, located in the corpus callosum and the ventricles of a common template obtained by merging spatially normalized anatomical images from all the participants. Finally, a low pass filter was applied. Results obtained in subsequent analyses were anatomically localized on the group-averaged Talairach-transformed T1-weighted images.

2.6. BOLD Temporal Variability Analysis. We used the mean squared successive difference (MSSD) measure to calculate BOLD signal temporal variability in every subject and for each experimental condition. For each individual run, MSSD was computed over the entire preprocessed activation time course using a custom-built function in MATLAB (The MathWorks, Inc.).

For each subject, MSSD values were averaged across different runs of the same experimental protocol, and nonparametric Mann-Whitney tests were used to look for any potential difference between blind and sighted subjects. Significance threshold was set at corrected $P < 0.05$, calculated with a Monte-Carlo simulation run via *3dClustSim* program in AFNI, with a voxelwise threshold of $P < 0.02$, which resulted in a minimum cluster volume of $k < 111$ voxels.

Thresholded sighted versus blind MSSD comparison maps of the two different experiments were used to compute a conjunction map (logical AND) to identify regions that showed similar differences in signal variability across conditions (small volume correction P value < 0.001 , minimum cluster volume $k > 28$ voxels, as calculated on the sighted versus blind MSSD comparison map of the spatial discrimination condition).

2.7. Functional Connectivity Analysis. Cortical areas whose BOLD signal variability was significantly different between sighted and blind individuals during both the tactile spatial discrimination and the tactile motion perception experiments were used as seed regions of interest (ROI) for a functional connectivity analysis. Specifically, for each subject and condition the Pearson's correlation coefficient was computed between the BOLD signal time course (obtained concatenating all task-related functional runs) of the ROI and the time course of all the other voxels of the brain.

To identify the significant pattern of functional connectivity for each group and each experimental condition, we converted correlation coefficients of each subject into Z scores using Fisher's Z transformation and then performed one-sample group t -tests. Significance threshold was set at corrected $P < 0.05$, obtained using a voxelwise threshold of $P < 0.01$ and a cluster greater than 66 voxels. Furthermore, in order to determine significant differences in functional connectivity between congenitally blind and sighted individuals in each condition, unpaired nonparametric Mann-Whitney

tests were performed to compare the two groups during both tactile experiments (voxel-wise $P < 0.05$, $k > 30$ voxels).

3. Results

3.1. BOLD Signal Variability in Congenitally Blind and Sighted Individuals. During both experiments the blind subjects showed a significantly higher MSSD measure in a number of cortical brain areas as compared to the sighted individuals. Specifically, during the tactile spatial discrimination task, blind individuals had a significantly greater (corrected $P < 0.05$) signal variability in the left superior and bilateral inferior parietal, left superior frontal, right middle temporal, bilateral superior temporal, lingual, medial frontal and cingulate cortex, precuneus and cuneus (Figure 1(a)). During the tactile motion perception task, blind individuals showed a significantly greater (corrected $P < 0.05$) signal variability in the bilateral superior and inferior parietal cortex, left lingual, right superior temporal, postcentral, inferior frontal, and anterior cingulate cortex, as compared to the sighted group (Figure 1(b)).

The conjunction map computed from the sighted-versus-blind MSSD contrast maps obtained for the two tactile experimental protocols revealed a common cluster (small volume correction, $P < 0.02$; Figure 1(c)) of greater BOLD variability in the blind as compared to sighted individuals located in left inferior parietal and anterior intraparietal cortex (Talairach coordinates of the center of mass were $x = -40$, $y = -43$, $z = 49$).

3.2. Functional Correlation between the Left Parietal Cortex and Occipital Areas in the Congenitally Blind and Sighted Individuals. We performed a functional connectivity analysis using the left inferior parietal and anterior intraparietal cluster as a seed ROI for each group and each experimental condition. In both the blind and sighted individuals, during the tactile spatial (Figure 2(a)) and tactile motion (Figure 2(b)) discrimination tasks, the parietal ROI was significantly correlated (corrected $P < 0.05$) with a number of brain areas including inferior/superior parietal, middle/superior temporal, middle/superior and anterior frontal, motor and cingulate cortex, and clusters in the occipital regions. Interestingly, blind subjects showed a greater correlation between the left inferior parietal lobule and visual areas, comprising bilateral cuneus/superior occipital (Talairach coordinates of the peak voxels: $x = -16$, $y = -82$, $z = 32$; $x = 7$, $y = -85$, $z = 41$) during the spatial discrimination task and bilateral cuneus, superior and middle occipital (Talairach coordinates of the peak voxels: $x = 23$, $y = -82$, $z = 26$; $x = -25$; $y = -70$, $z = 38$), precuneus (Talairach coordinates $x = -5$, $y = -56$, $z = 29$), and right supramarginal (Talairach coordinates $x = 44$, $y = -55$, $z = 29$) cortex during the motion perception task (Figures 2(c)–2(d)). Additional regions in bilateral sensorimotor and left anterior temporal cortex (spatial discrimination) and in bilateral inferior parietal cortex and anterior cingulate (motion perception) showed a greater correlation with the seed ROI in blind individuals, while in bilateral medial

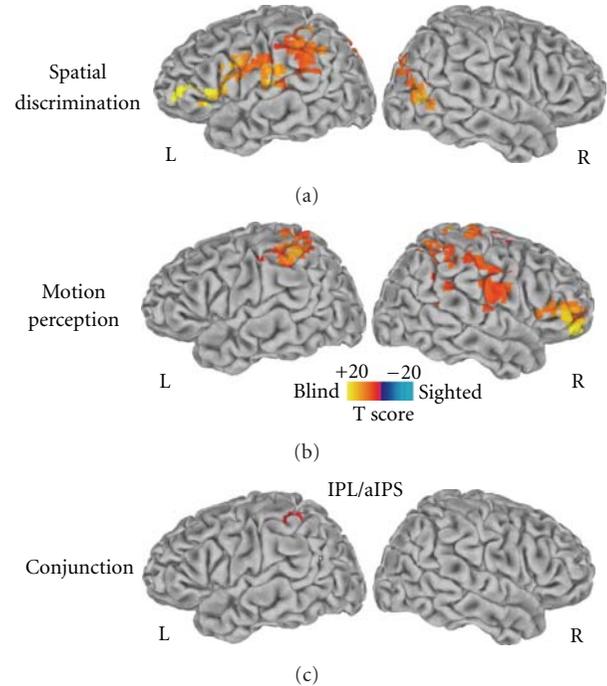


FIGURE 1: Mean squared successive difference (MSSD) significant (corrected $P < 0.05$) differences between blind and sighted individuals during (a) the tactile spatial discrimination and (b) tactile motion perception experiments. (c) The conjunction map (logical AND) obtained from the thresholded sighted-versus-blind MSSD maps revealed a region located in left inferior parietal and anterior intraparietal cortex (IPL/aIPS) that showed similar differences in signal variability in the tactile spatial and motion discrimination experiments ($k > 30$ voxels). Spatially normalized MSSD differences are projected onto single-subject left and right hemisphere templates in the Talairach space.

prefrontal and right superior temporal cortex during the spatial discrimination task in sighted individuals (Figures 2(c)–2(d)).

4. Discussion

The aim of the present study was to examine whether the lack of visual experience since birth may lead to changes in neural efficiency, as measured by the BOLD signal temporal variability, within the parietal cortex and in its connections with occipital cortical areas in relation to the processing of non-visual sensory information. As a matter of fact, despite the growing number of studies indicating the existence also in blind individuals of *supramodal* areas within the visual cortex capable of processing information conveyed by different sensory modalities [2] and the potential additional development of *cross-modally*-reorganized occipital clusters [3, 5, 21], the specific pathway(s) that carry such non-visual sensory information to the occipital cortex are still a matter of wide debate. Indeed, direct connections between different primary sensory areas, subcortico-cortical loops, or cortico-cortical pathway(s) are all considered potential mechanisms to explain how non-visual sensory inputs reach the “visual” cortex [3, 6].

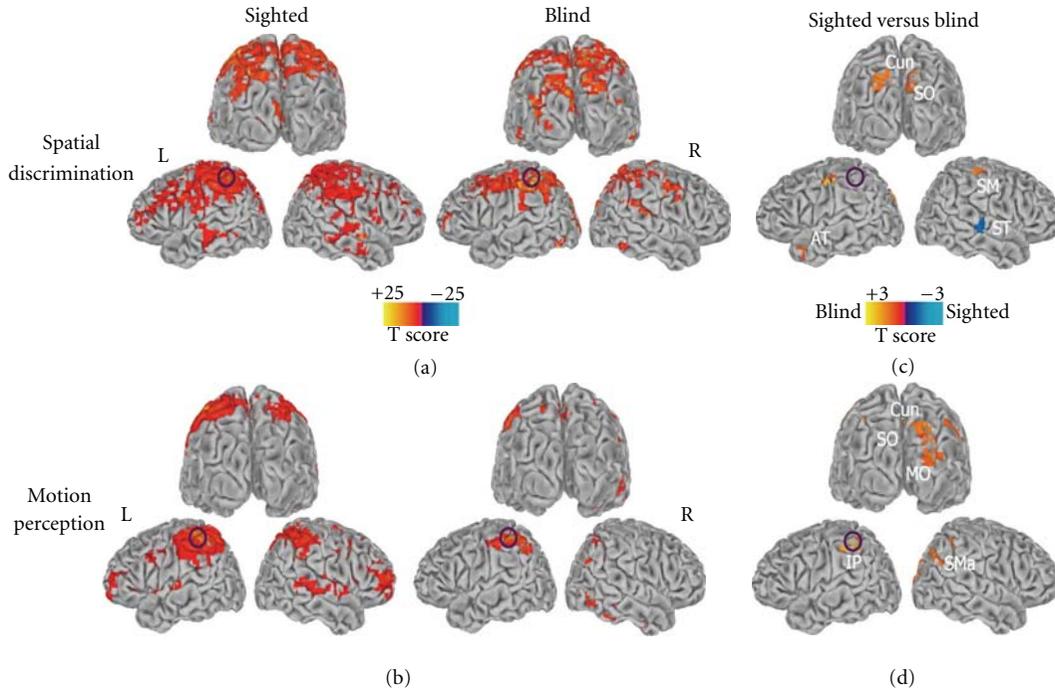


FIGURE 2: Group functional connectivity (FC) maps obtained for sighted (first column) and blind (second column) individuals during (a) the tactile spatial discrimination and (b) the tactile motion perception tasks (corrected $P < 0.05$). The third column (c)-(d) shows the differences in functional connectivity between the two groups (uncorrected $P < 0.05$, $k > 30$ voxels). The left inferior parietal seed ROI has been indicated with a purple circle. Spatially normalized maps are projected onto single-subject left and right hemisphere templates in the Talairach space in frontal and lateral views. Cun: cuneus; SO: superior occipital; MO: middle occipital; SMA: supramarginal; IP: inferior parietal; ST: superior temporal; AT: anterior temporal.

As far as tactile processing in the occipital areas is concerned, the hypothesis of a cortico-cortical connection has been supported by the results of experiments that used TMS to induce temporary functional lesions during Braille reading [4, 15, 37]. In congenitally blind individuals, parietal activation in response to tactile letter detection precedes occipital activation associated with letter identification, thus suggesting that tactile information reaches the occipital cortex through a parieto-occipital pathway [1]. Additionally, other tactile experimental paradigms in early blind participants confirmed a reinforced functional coupling between occipital and parietal cortical areas, without any subcortical involvement [4, 15, 27–29]. For instance, a connectivity analysis during electro-tactile stimulation of the tongue showed that anterior areas of the parietal cortex had an increased correlation in activity with posterior parietal cortex and the visual occipital cortex of trained blind individuals [28]. Consistently, in a distinct tactile motion perception experiment in which subjects had to discriminate motion of plastic dots under their fingertips [25], we found that somatosensory areas showed extensive bilateral connections with contiguous posterior parietal and intraparietal regions, and with middle temporal and lateral occipital areas in both sighted and congenitally blind individuals, supporting a cortico-cortical pathway from primary somatosensory cortex through parietal regions to occipital areas [27].

On the other hand, we cannot exclude that other sensory modalities, such as auditory inputs, may follow distinct pathways of sensory integration. Indeed, the spatial processing of sounds in early blind and sighted individuals was impaired only when TMS was applied at short latencies to the right dorsal occipital cortex, but not when it was applied to the right intraparietal region [22, 38], thus suggesting that sounds may reach occipital regions in the blind brain either via subcortical connections or direct projections from the auditory cortex.

In light of these preliminary observations on the occipital processing of somatosensory information, we hypothesized that, as a consequence of plastic rearrangements, parietal areas in congenitally blind individuals would elaborate a greater amount of information during tactile tasks and would be characterized by specific functional networks with strengthened occipitoparietal connectivity. Parietal areas, including the superior parietal and the intraparietal cortex, have been proposed as hubs of integration for multimodal inputs, and many studies have found connections of these subregions with primary areas of different sensory modalities both in monkeys and in humans (reviewed in [39, 40]). Therefore, parietal regions multisensory in nature which are expected to receive and integrate different inputs may show a more efficient processing and a greater capacity to switch between different network configurations. To test this hypothesis, in the present study we compared BOLD signal

temporal variability, a recently proposed index of functional brain efficiency [32, 41], across sighted and congenitally blind individuals during two different tactile tasks: spatial discrimination and motion perception.

Variability in BOLD signal is thought to increase with the functional complexity of the cortical networks that subserve a specific function, as shown in some earlier experiments which employed EEG and MEG [31, 32, 42], though the physiological meaning of this measure still remains to be fully understood [42]. For instance, by studying populations at different ages, Garrett and colleagues recently showed that the noise (i.e., temporal variability) in BOLD signal is greater in young highly performing adults as compared to older adults [30, 32]. Moreover, a higher variability in certain brain areas was associated with superior behavioral performances, suggesting a correlation between this parameter and operative brain functional efficiency [41]. In the present study, BOLD signal variability was assessed by computing the mean of the square differences between the values of BOLD signal at two successive time points (i.e., MSSD) [33, 34].

Our results showed that during both the tactile spatial discrimination and motion perception conditions, the blind individuals were characterized by increased signal variability in brain areas distributed over prefrontal, parietal, occipital and temporal cortex. Differences in MSSD scores between the two groups were more evident in the tactile spatial discrimination task than in the motion perception condition, mainly in temporal, prefrontal and occipitoparietal areas. In order to avoid specific effects dependent on the distinct experimental conditions, we performed a conjunction analysis to identify common areas of increased MSSD in the congenitally blind group. A significant cluster of overlapping in group MSSD differences was localized in the left inferior parietal and anterior intraparietal cortex.

To verify whether such differences in BOLD signal variability were also associated with changes in the functional network organization, a functional connectivity analysis using the identified parietal cluster as a seed region of interest also was carried out. Results showed a more extended inclusion of occipital regions in the connectivity network of this parietal region in the blind as compared to sighted individuals. In general, congenitally blind individuals demonstrated enhanced connectivity between brain areas processing different sensory inputs as compared to sighted [43, 44] or novel pattern of correlations between cross-modal reorganized occipital areas and cortical areas conveying or elaborating non-visual stimuli [27].

Taken together, our MSSD and functional connectivity results suggest a reinforced integrative role of the parietal cortex during tactile perception in congenitally blind individuals as compared to the sighted ones. Moreover, our results provide novel support to the potential role of the parietal cortex in conveying tactile and maybe other non-visual information to striate and extrastriate occipital regions in blind individuals. In fact, although a concomitant involvement of a subcortical loop between the somatosensory and occipital areas may exist [2, 45], these results suggest that important functional plastic modifications occur in the parietal cortex in congenitally blind individuals, including changes in the

functional connectivity network, which is rewired to reach occipital areas.

Notably, it should be emphasized that the measures of BOLD temporal variability are independent of task-related BOLD responses and appear more to provide information about the level of functional integration other than the recruitment of a specific brain region during distinct experimental tasks [30, 41, 42]. Therefore, these measures do not directly reflect task-related perceptual and cognitive processing. Furthermore, the original studies on BOLD signal variability evaluated the effects of different cognitive tasks on this measure [30, 41, 42]. The authors showed a substantial independence of the age-related differences in BOLD variability from the specific task employed. Here, to properly avoid any specific effects dependent on the two experimental conditions, we also performed a conjunction analysis to identify common areas of increased MSSD in the congenitally blind groups.

Sample sizes may represent a limitation of our analysis, as four blind individuals per experimental protocol is a relatively small number of subject are for an fMRI study nowadays, and the results reported here would need to be replicated in a larger cohort. Nonetheless, congenitally blind individuals represent an exceptionally rare population that should comply with strict selection criteria from a medical (e.g., no other neurological disorders, drug-free) and demographic (e.g., independent living conditions, socially integrated) perspective. Even if limited samples could be sufficient to respond to specific hypothesis-driven protocols (e.g., [18]), some methodological considerations have been however taken into account, such as the use of nonparametric comparisons.

5. Conclusions

In summary, these findings, though acquired in a relatively limited sample and therefore in need of replication in larger groups, expand the current knowledge on the functional reorganization that occurs in the brain of congenitally blind individuals, as they demonstrate that during both tactile spatial discrimination and motion perception tasks, the inferior parietal and anterior intraparietal cortices were characterized by increased BOLD signal temporal variability and relevant functional network modifications in the congenitally blind as compared to the sighted subjects. Overall, these findings support a role for parietal cortex in conveying nonvisual-information to visual cortex after cross-modal plastic modifications in the brain of visually deprived individuals.

Acknowledgments

The authors wish to thank all the volunteers who took part in their studies and the Unione Italiana Ciechi for its support to these projects and the MRI Laboratory at the *Fondazione Regione Toscana/CNR "G. Monasterio"* (Pisa, Italy) coordinated by Massimo Lombardi. This research was supported by the Italian Ministero dell'Istruzione, dell'Università e della

Ricerca (PRIN 2009 to P. Pietrini,) and by the Medical School of the University of Pisa (Research Grant to E. Ricciardi).

References

- [1] A. Pascual-Leone, A. Amedi, F. Fregni, and L. B. Merabet, “The plastic human brain cortex,” *Annual Review of Neuroscience*, vol. 28, pp. 377–401, 2005.
- [2] E. Ricciardi and P. Pietrini, “New light from the dark: what blindness can teach us about brain function,” *Current Opinion in Neurology*, vol. 24, no. 4, pp. 357–363, 2011.
- [3] J. Frasnelli, O. Collignon, P. Voss, and F. Lepore, “Crossmodal plasticity in sensory loss,” *Progress in Brain Research*, vol. 191, pp. 233–249, 2011.
- [4] U. Noppeney, “The effects of visual deprivation on functional and structural organization of the human brain,” *Neuroscience and Biobehavioral Reviews*, vol. 31, no. 8, pp. 1169–1180, 2007.
- [5] A. Amedi, L. B. Merabet, F. Bempohl, and A. Pascual-Leone, “The occipital cortex in the blind lessons about plasticity and vision,” *Current Directions in Psychological Science*, vol. 14, no. 6, pp. 306–311, 2005.
- [6] R. Kupers, P. Pietrini, E. Ricciardi, and M. Ptito, “The nature of consciousness in the visually deprived brain,” *Frontiers in Psychology*, vol. 2, pp. 1–14, 2011.
- [7] M. Ptito, J. F. Giguère, D. Boire, D. O. Frost, and C. Casanova, “When the auditory cortex turns visual,” *Progress in Brain Research*, vol. 134, pp. 447–458, 2001.
- [8] A. Ptito, A. Fortin, and M. Ptito, “‘Seeing’ in the blind hemi-field following hemispherectomy,” *Progress in Brain Research*, vol. 134, pp. 367–378, 2001.
- [9] H. Burton, A. Z. Snyder, T. E. Conturo, E. Akbudak, J. M. Ollinger, and M. E. Raichle, “Adaptive changes in early and late blind: a fMRI study of Braille reading,” *Journal of Neurophysiology*, vol. 87, no. 1, pp. 589–607, 2002.
- [10] N. Sadato, A. Pascual-Leone, J. Grafman, M. P. Deiber, V. Ibañez, and M. Hallett, “Neural networks for Braille reading by the blind,” *Brain*, vol. 121, no. 7, pp. 1213–1229, 1998.
- [11] N. Sadato, A. Pascual-Leone, J. Grafman et al., “Activation of the primary visual cortex by Braille reading in blind subjects,” *Nature*, vol. 380, no. 6574, pp. 526–528, 1996.
- [12] A. A. Stevens, M. Snodgrass, D. Schwartz, and K. Weaver, “Preparatory activity in occipital cortex in early blind humans predicts auditory perceptual performance,” *Journal of Neuroscience*, vol. 27, no. 40, pp. 10734–10741, 2007.
- [13] R. Weeks, B. Horwitz, A. Aziz-Sultan et al., “A positron emission tomographic study of auditory localization in the congenitally blind,” *Journal of Neuroscience*, vol. 20, no. 7, pp. 2664–2672, 2000.
- [14] O. Collignon, F. Champoux, P. Voss, and F. Lepore, “Sensory rehabilitation in the plastic brain,” *Progress in Brain Research*, vol. 191, pp. 211–231, 2011.
- [15] A. Pascual-Leone, V. Walsh, and J. Rothwell, “Transcranial magnetic stimulation in cognitive neuroscience—virtual lesion, chronometry, and functional connectivity,” *Current Opinion in Neurobiology*, vol. 10, no. 2, pp. 232–237, 2000.
- [16] R. Kupers, A. Fumal, A. M. de Noordhout, A. Gjedde, J. Schoenen, and M. Ptito, “Transcranial magnetic stimulation of the visual cortex induces somatotopically organized qualia in blind subjects,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 103, no. 35, pp. 13256–13260, 2006.
- [17] A. Pascual-Leone and R. Hamilton, “The metamodal organization of the brain,” *Progress in Brain Research*, vol. 134, pp. 427–445, 2001.
- [18] B. Z. Mahon, S. Anzellotti, J. Schwarzbach, M. Zampini, and A. Caramazza, “Category-specific organization in the human brain does not require visual experience,” *Neuron*, vol. 63, no. 3, pp. 397–405, 2009.
- [19] P. Pietrini, M. L. Furey, E. Ricciardi et al., “Beyond sensory images: object-based representation in the human ventral pathway,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 101, no. 15, pp. 5658–5663, 2004.
- [20] D. Bonino, E. Ricciardi, L. Sani et al., “Tactile spatial working memory activates the dorsal extrastriate cortical pathway in congenitally blind individuals,” *Archives Italiennes de Biologie*, vol. 146, no. 3-4, pp. 133–146, 2008.
- [21] O. Collignon, G. Vandewalle, P. Voss et al., “Functional specialization for auditory-spatial processing in the occipital cortex of congenitally blind humans,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 11, pp. 4435–4440, 2011.
- [22] O. Collignon, P. Voss, M. Lassonde, and F. Lepore, “Cross-modal plasticity for the spatial processing of sounds in visually deprived subjects,” *Experimental Brain Research*, vol. 192, no. 3, pp. 343–358, 2009.
- [23] F. Gougoux, R. J. Zatorre, M. Lassonde, P. Voss, and F. Lepore, “A functional neuroimaging study of sound localization: visual cortex activity predicts performance in early-blind individuals,” *PLoS biology*, vol. 3, no. 2, p. e27, 2005.
- [24] E. Ricciardi, D. Bonino, C. Gentili, L. Sani, P. Pietrini, and T. Vecchi, “Neural correlates of spatial working memory in humans: a functional magnetic resonance imaging study comparing visual and tactile processes,” *Neuroscience*, vol. 139, no. 1, pp. 339–349, 2006.
- [25] C. Poirier, O. Collignon, C. Scheiber et al., “Auditory motion perception activates visual motion areas in early blind subjects,” *NeuroImage*, vol. 31, no. 1, pp. 279–285, 2006.
- [26] E. Ricciardi, N. Vanello, L. Sani et al., “The effect of visual experience on the development of functional architecture in hMT+,” *Cerebral Cortex*, vol. 17, no. 12, pp. 2933–2939, 2007.
- [27] L. Sani, E. Ricciardi, C. Gentili, N. Vanello, J. V. Haxby, and P. Pietrini, “Effects of visual experience on the human MT+ functional connectivity networks: an fMRI study of motion perception in sighted and congenitally blind individuals,” *Frontiers in Systems Neuroscience*, vol. 4, article 159, 2010.
- [28] M. Ptito, S. M. Moesgaard, A. Gjedde, and R. Kupers, “Cross-modal plasticity revealed by electrotactile stimulation of the tongue in the congenitally blind,” *Brain*, vol. 128, no. 3, pp. 606–614, 2005.
- [29] G. F. Wittenberg, K. J. Werhahn, E. M. Wassermann, P. Herscovitch, and L. G. Cohen, “Functional connectivity between somatosensory and visual cortex in early blind humans,” *European Journal of Neuroscience*, vol. 20, no. 7, pp. 1923–1927, 2004.
- [30] D. D. Garrett, N. Kovacevic, A. R. McIntosh, and C. L. Grady, “Blood oxygen level-dependent signal variability is more than just noise,” *Journal of Neuroscience*, vol. 30, no. 14, pp. 4914–4921, 2010.
- [31] A. R. McIntosh, N. Kovacevic, and R. J. Itier, “Increased brain signal variability accompanies lower behavioral variability in development,” *PLoS Computational Biology*, vol. 4, no. 7, Article ID e1000106, 2008.

- [32] A. R. McIntosh, N. Kovacevic, S. Lippe, D. Garrett, C. Grady, and V. Jirsa, "The development of a noisy brain," *Archives Italiennes de Biologie*, vol. 148, no. 3, pp. 323–337, 2010.
- [33] G. R. Samanez-Larkin, C. M. Kuhnen, D. J. Yoo, and B. Knutson, "Variability in nucleus accumbens activity mediates age-related suboptimal financial risk taking," *Journal of Neuroscience*, vol. 30, no. 4, pp. 1426–1434, 2010.
- [34] J. von Neumann, R. H. Kent, H. R. Bellinson, and B. I. Hart, "The mean square successive difference," *The Annals of Mathematical Statistics*, vol. 12, pp. 153–162, 1941.
- [35] R. W. Cox, "AFNI: software for analysis and visualization of functional magnetic resonance neuroimages," *Computers and Biomedical Research*, vol. 29, no. 3, pp. 162–173, 1996.
- [36] J. Tàilairach and P. Tournoux, "Co-planar stereotaxic atlas of the human brain," pp. Thieme Medical Publisher–New York, NY, USA, 1988.
- [37] R. Kupers, M. Pappens, A. M. de Noordhout, J. Schoenen, M. Ptito, and A. Fumal, "rTMS of the occipital cortex abolishes Braille reading and repetition priming in blind subjects," *Neurology*, vol. 68, no. 9, pp. 691–693, 2007.
- [38] O. Collignon, M. Davare, A. G. de Volder, C. Poirier, E. Olivier, and C. Veraart, "Time-course of posterior parietal and occipital cortex contribution to sound localization," *Journal of Cognitive Neuroscience*, vol. 20, no. 8, pp. 1454–1463, 2008.
- [39] C. Cappe, E. M. Rouiller, and P. Barone, "Multisensory anatomical pathways," *Hearing Research*, vol. 258, no. 1–2, pp. 28–36, 2009.
- [40] J. Klemen and C. D. Chambers, "Current perspectives and methods in studying neural mechanisms of multisensory interactions," *Neuroscience and Biobehavioral Reviews*, vol. 36, pp. 111–133, 2012.
- [41] D. D. Garrett, N. Kovacevic, A. R. McIntosh, and C. L. Grady, "The importance of being variable," *Journal of Neuroscience*, vol. 31, no. 12, pp. 4496–4503, 2011.
- [42] P. N. C. Mohr and I. E. Nagel, "Variability in brain activity as an individual difference measure in neuroscience?" *Journal of Neuroscience*, vol. 30, no. 23, pp. 7755–7757, 2010.
- [43] T. Fujii, H. C. Tanabe, T. Kochiyama, and N. Sadato, "An investigation of cross-modal plasticity of effective connectivity in the blind by dynamic causal modeling of functional MRI data," *Neuroscience Research*, vol. 65, no. 2, pp. 175–186, 2009.
- [44] C. Klinge, F. Eippert, B. Röder, and C. Büchel, "Cortico-cortical connections mediate primary visual cortex responses to auditory stimulation in the blind," *Journal of Neuroscience*, vol. 30, no. 38, pp. 12798–12805, 2010.
- [45] A. Cowey, "Visual system: how does blindsight arise?" *Current Biology*, vol. 20, no. 17, pp. R702–R704, 2010.

Research Article

Crossmodal Recruitment of the Ventral Visual Stream in Congenital Blindness

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Received 22 January 2012; Accepted 19 April 2012

Academic Editor: Pietro Pietrini

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We used functional MRI (fMRI) to test the hypothesis that blind subjects recruit the ventral visual stream during nonhaptic tactile-form recognition. Congenitally blind and blindfolded sighted control subjects were scanned after they had been trained during four consecutive days to perform a tactile-form recognition task with the tongue display unit (TDU). Both groups learned the task at the same rate. In line with our hypothesis, the fMRI data showed that during nonhaptic shape recognition, blind subjects activated large portions of the ventral visual stream, including the cuneus, precuneus, inferotemporal (IT), cortex, lateral occipital tactile vision area (LOtv), and fusiform gyrus. Control subjects activated area LOtv and precuneus but not cuneus, IT and fusiform gyrus. These results indicate that congenitally blind subjects recruit key regions in the ventral visual pathway during nonhaptic tactile shape discrimination. The activation of LOtv by nonhaptic tactile shape processing in blind and sighted subjects adds further support to the notion that this area subserves an abstract or supramodal representation of shape. Together with our previous findings, our data suggest that the segregation of the efferent projections of the primary visual cortex into a dorsal and ventral visual stream is preserved in individuals blind from birth.

1. Introduction

It is well established that early-onset blindness leads to widespread neuroplastic changes. For instance, studies have shown that the senses of hearing and touch are more developed in blind than sighted individuals [1–6], probably due to training-induced plasticity. The enlargement of the somatic and motor area representation of the index finger in proficient Braille readers is a clear example of this experience-dependent plastic process [7]. Brain imaging studies using 18F-fluoro-deoxyglucose-positron emission tomography (FDG-PET) have shown that, despite the absence of visual input, the occipital cortex of congenitally blind individuals shows a supranormal metabolism at rest [8, 9]. This indicates that the visually deprived cortex is still functionally

active and can be recruited by other modalities such as touch, hearing, and smell. Indeed, studies using a variety of brain imaging tools such as PET, functional, magnetic resonance imaging (fMRI), event-related potentials and magnetoencephalography, all concur on a recruitment of the visual cortex of early blind individuals during various nonvisual tasks (e.g., [10–14]).

Numerous brain imaging studies have consistently found activations of occipital cortical areas when blind subjects perform a range of tactile tasks such as Braille character recognition, vibrotactile discrimination, and haptic object exploration [15–20]. Previous work from our laboratory showed that blind subjects who had been trained to use the TDU in an orientation and motion discrimination task [13, 14] or in a navigation task [21] activate their visual cortex. The

observed activation patterns within the visual cortex were remarkably similar to those observed in sighted individuals performing corresponding visual tasks. The modularity of these activations is further substantiated by the observation that in early blind individuals, transcranial magnetic stimulation (TMS) of the reorganized visual cortex elicits somatotopically organized tactile sensations [22, 23]. TMS studies have also provided evidence for the functional implication of the occipital cortex in tactile processing. For example, the demonstration that repetitive TMS of the occipital cortex disrupts Braille reading performance in the blind [24, 25] suggests that the contribution of the visually-deprived occipital cortex to nonvisual functions is indeed functionally relevant. Together, these findings indicate that the visually deprived posterior cortical regions are much more adaptable than previously thought and may act either as a high-level multisensory area [26] or undergoes a cross-modal plastic reorganization [27].

The visual system is grossly subdivided into a dorsal and a ventral processing stream [28]. Area hMT+, a critical part of the dorsal visual pathway, involved in visual motion processing, is recruited during tactile and auditory motion discrimination task in early blind subjects [14, 29–31], suggesting that the dorsal “where” processing stream is functionally preserved in subjects lacking vision from birth. This raises the question whether the ventral processing stream, known to participate in object and shape recognition [30–39], is also preserved in blind subjects. Evidence in support of this hypothesis comes from a study by Pietrini et al. [12] that showed category-specific recruitment of the ventral temporal cortex by haptic exploration of objects in congenitally blind and sighted individuals. Therefore, the aim of this study was to investigate whether the ventral stream will also be activated by nonhaptic exploration of shapes.

2. Materials and Methods

2.1. Subjects. Ten sighted control (five females; mean age: 21 ± 11 y.) and eight blind (seven congenitally and one early blind) individuals with no recollection of any visual experience (four females; mean age: 31 ± 10 y.) participated in this study. Causes of blindness were retinopathy of prematurity [7] and Leber’s congenital amaurosis [1]. Visual inspection of the structural brain MRI scans by a trained neuroradiologist did not reveal macroscopic abnormalities and none of the subjects had a history of psychiatric or neurological illness. The study protocol was approved by the local ethics committee (Project ID: KF-01328723) and all subjects provided written informed consent.

2.2. Electro-Tactile Stimulation of the Tongue. The apparatus has been described in detail elsewhere [13]. Briefly, it consists of a tongue display unit (TDU, Wicab Inc.), an electrode array (3×3 cm) with 144 gold-plated contacts arranged in a 12×12 matrix and a laptop with custom-made software (Figure 1(a)). Computer-generated geometric shapes were converted into electrical pulses and delivered to the tongue via the electrode array. Stimulation intensity was controlled

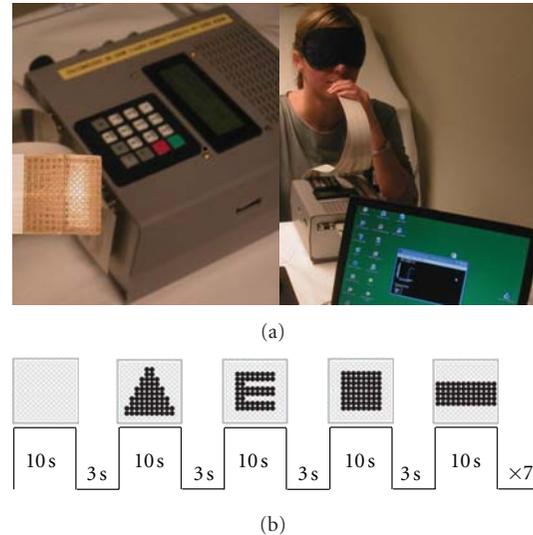


FIGURE 1: Experimental setup. (a) The tongue display unit (TDU) and its components. (b) The fMRI block design. During each of the two fMRI runs, 7 stimulation blocks were presented. One block consisted of a rest period and Shape stimuli used during training and fMRI.

by the subject and could be adjusted at any time to allow optimal perception of the stimuli.

2.3. Behavioural Training. Both blind and blindfolded control subjects were trained during 10 sessions, stretched over 4 consecutive days. Each session lasted around 15 minutes and comprised 28 trials. During training, subjects learned to use the TDU to recognize four different shapes that were randomly presented: a square, a triangle, a rectangle, and the letter E (Figure 1(b)). Participants were given a maximum of 30 seconds to identify each stimulus and they received immediate feedback about the correctness of their response. Both the reaction time and the response accuracy were measured. It was stressed that correctness of responses was more important than speed. Stimuli were presented statically and participants could not explore the images by using exploratory movements of a computer mouse as was the case in our previous PET study on orientation discrimination [13]. Prior to the training sessions, participants were familiarized with the TDU and the experimental procedures. They were told the forms that were going to be used and that their task was to correctly identify the shape that was presented. Blind participants were asked whether they were familiar with the shapes that were going to be presented and they were given the opportunity to explore haptically plastic copies of the four shapes if necessary. Training sessions were limited to a maximum of 15 minutes to avoid habituation to tongue stimulation. Participants were given two or maximum 3 training sessions per day. Between two successive sessions there was a minimum time interval of 30 minutes. We chose to work with a limited set of shapes in order not to overload memory and cognitive processing since during the fMRI session. Subjects had to indicate their response by pressing one of the four keys on a response pad (1: square;

2: triangle; 3: rectangle and; 4: letter E). All participants were trained by the same experimenter (IM). The criterion for successful learning was set to 85% correct responses in two consecutive sessions. Participants who reached this criterion could participate the next day in the fMRI examination. Statistical analysis of the behavioural data was carried out using ANOVA (SPSS18, Chicago, Ill, US). Values of $P < 0.05$ were considered as statistically significant.

2.4. MRI Experimental Design. Following behavioural training, subjects performed the shape recognition task during whole-brain fMRI. We used an fMRI block design with periods of rest (the electrode array was placed on the tongue but no electrotactile stimulation was administered) and task (i.e., nonhaptic shape recognition). The same shapes were presented in the fMRI session as during behavioural training. Two fMRI runs were carried out, each lasting 7 minutes and 40 seconds. Each run consisted of alternating rest and task blocks (Figure 1(b)). During a task block, four stimuli, one for each form, were presented in a random order. This was repeated seven times, resulting in 35 blocks per fMRI run. Each stimulus lasted 10 s and was followed by a 3 s interval during which subjects had to indicate which form had been presented by pressing one of 4 buttons on a keypad with their right hand. Each button corresponded to one of the stimulus forms. Prior to scanning, subjects practiced to use the appropriate corresponding response button.

2.5. Image Acquisition and Analysis. Task-related changes in the (blood oxygenation level-dependent BOLD signal were measured with whole-brain fMRI using a Siemens Trio 3 Tesla MR Scanner (Siemens, Erlangen, Germany), equipped with an 8-channel head coil. The multislice gradient echo-planar imaging sequence had a repetition time (TR) = 2500 ms, echo time (TE) = 50 ms, flip angle (FA) = 90°, and field of view (FOV) of 192 mm (matrix: 64 × 64). Each volume consisted of 42 slices in an inclined axial plane, aligned to the AC-PC axis, with a slice thickness of 4 mm, resulting in a voxel size of 4 × 4 × 4 mm. A total of 368 functional brain volumes were acquired per subject. After the fMRI session, a high-resolution structural T1-weighted three-dimensional brain scan (MPRAGE) was acquired using a gradient echo pulse sequence (TE = 9.20 ms; flipangle = 30°; FOV = 256 mm; matrix = 256 × 256; voxelsize = 1 mm³).

The MRI data were analyzed using Statistical Parametric Mapping software (SPM5, Wellcome Department of Cognitive Neurology, London, UK). Functional volumes were motion-corrected using SINC interpolation and spatially normalized to the reference space defined by the MRI template supplied by the Montreal Neurological Institute (MNI). Images were spatially smoothed with an 8-mm wide Gaussian kernel to improve the signal-to-noise ratio.

For the statistical analysis, active conditions were fitted with a box-car function convolved with the hemodynamic response function. Low-frequency temporal drifts were removed by applying a 128-s high-pass filter. The duration of all conditions was modelled, except for the 10 s rest periods,

which served as baseline. In order to estimate the effects associated with the experimental design, we evaluated BOLD signal changes associated with the contrast active task (shapes) compared to the control task (rest). Following single subject analyses, we performed a random-effect analysis within and between groups using the individual contrast estimates for each functional run. Activation maps were thresholded at $P < 0.01$, corrected for multiple comparisons using the false discovery rate (FDR) [40]. We applied a conservative extent threshold of 20 contiguous voxels.

3. Results

3.1. Behavioural Training. Both blind and sighted control subjects learned the tactile form discrimination task within the 10 sessions. Figure 2 illustrates the learning curves for percentage of correct responses and reaction times. A statistical analysis of the time × group interactions yielded no significant differences in the percentage of correct responses ($F = 0.728$; $P > 0.05$) or reaction times at the end of the training ($F = 1.016$; $P > 0.05$).

3.2. Functional MRI. Blind subjects but not blindfolded sighted controls activated large areas of occipital (cuneus, inferior and middle occipital gyri and lingual gyrus) and occipito-temporal (fusiform gyrus) cortices (Figure 3). Both blind and sighted controls showed increased BOLD responses in the inferotemporal cortex (including area LOtv), post-central gyrus, superior and inferior parietal lobule, precuneus, prefrontal cortex, cingulate gyrus, insula, and cerebellum. Task-related activations for blind and control subjects are listed in Table 1. A direct comparison of the activation maps in both groups showed that BOLD increases in the inferior temporal gyrus, middle occipital cortex, and precuneus were significantly stronger in blind subjects (Figure 4). In contrast, blindfolded-sighted control subjects showed a relative larger BOLD response increase in the right postcentral gyrus (BA3) and the left anterior cingulate cortex (BA24) only (data not shown). We also observed activation in both blind and controls in left and right premotor areas that are probably due to the subject's preparation to respond to the tactile stimulation. An increased BOLD response was also found in bilateral somatosensory cortex for both groups of subjects.

4. Discussion

In this study, we report that congenitally blind but not sighted subjects activated large parts of the occipital cortex when performing a nonhaptic shape recognition task. Our data further showed that both groups recruited the inferotemporal cortex, including area LOtv, in response to 2D tactile shape information extracted from electrotactile stimulation of the tongue. Previous studies showed that area LOtv processes form information in the absence of visual input through haptic [12] or auditory modalities [15, 30]. The present data extend these findings by showing that area LOtv processes form information even when tactile stimuli

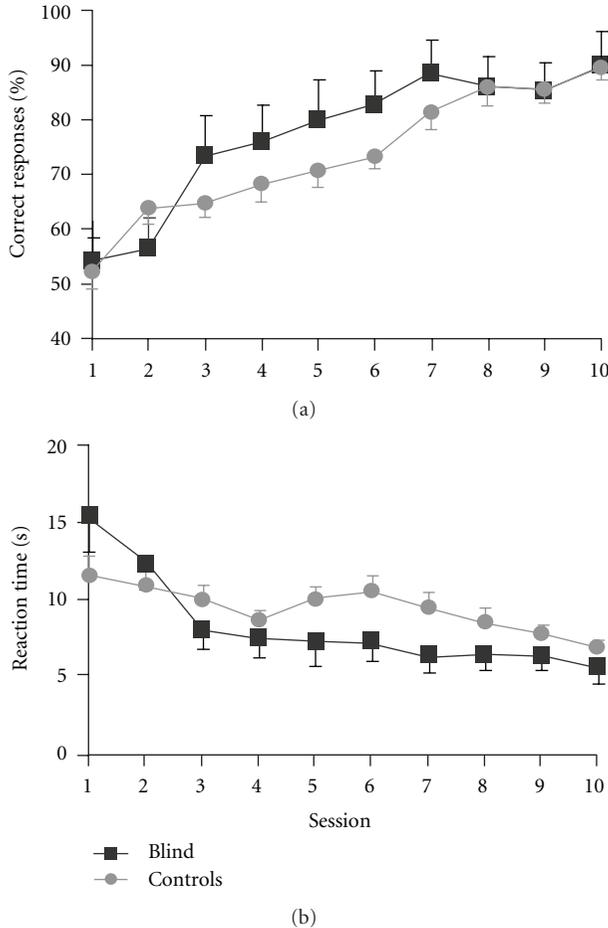


FIGURE 2: Learning curves for shape recognition in congenitally blind and blindfolded control subjects. (a) Mean percentage changes \pm SEM of correct responses and (b) mean reaction times \pm SEM. No significant differences in performance were observed between the groups.

are delivered nonhaptically and to a body part, like the tongue, that is not primarily devoted to shape recognition.

4.1. Activation of IT/LOtv Complex. We found strong task-related activation along the occipital/inferior temporal cortical border in both sighted and blind subjects. Whereas IT/LOtv was activated bilaterally in blind subjects, it was activated only in the right hemisphere in sighted participants. This might be due to the relatively small sample size. Indeed, when using a less stringent criterion for statistical significance ($P < 0.01$, uncorrected), an increased BOLD response was also noted in the left hemisphere. Moreover, a conjunction analysis of the activation patterns in both groups confirms the bilateral activation of IT/LOtv although the cluster size was markedly larger in the right compared to the left hemisphere (data not shown). The activation pattern in both groups encompassed a region that Amedi and coworkers [36, 37] have coined the lateral occipital tactile visual area. The stereotactic coordinates of our LOtv activation in both groups (see Table 1) are very close to

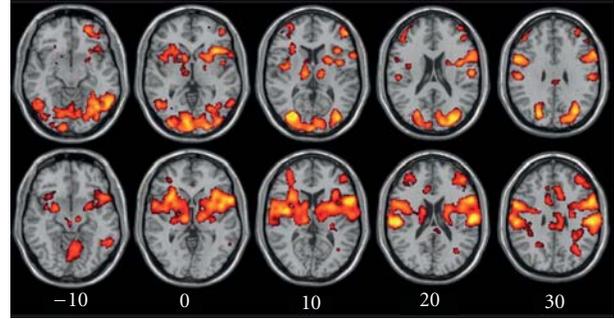


FIGURE 3: Axial maps showing brain activations for the contrast “shapes-rest” in congenitally blind (upper row) and blindfolded control (lower row) subjects. The color-coded t-maps illustrate voxels showing a task-related increase in activation at $P < 0.01$, FDR-corrected. Right side of the brain is to the right of the image.

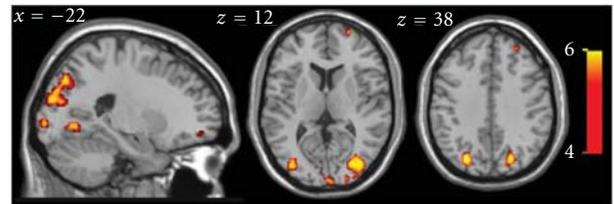


FIGURE 4: Cortical maps showing brain areas where activity was significantly larger in congenitally blind compared to blindfolded control subjects. The color-coded t-map shows the voxels with a relative increase in task-related activation in the blind group relative to controls at $P < 0.01$, FDR-corrected. Right side of the brain is to the right of the image.

those reported by others [12, 36, 41]. LOtv is a subregion within the human lateral occipital cortex (LOC) that is robustly activated during both visual and tactile object recognition. Amedi and coworkers [37] demonstrated that for both modalities, LOtv has a preference for objects compared to textures and scrambled objects; this area is only weakly activated by the motor, naming and visual imagery components of object recognition [37]. Area LOtv is also recruited by tactile exploration of novel, meaningless three-dimensional clay objects, suggesting that it responds more to form than to semantic features of objects [39]. Our finding of LOtv activation in both groups during the presentation of tactile stimuli is in accordance with previous results reported in normally sighted [12, 36, 36, 42–44] and blind [12] participants. The results further show for the first time that not only three-dimensional tactile stimuli but also two-dimensional nonhaptic tactile information can recruit area LOtv, adding further support that this area subserves an abstract or supramodal representation of shape information [45].

4.2. Occipital Cortex. Only blind subjects showed a significant BOLD response in several regions within the occipital cortex including the cuneus, the lingual gyrus and the inferior, middle, and superior occipital gyri. The activation pattern in the blind following training shows remarkable

TABLE 1: Activation clusters for “shapes versus rest” in blind and sighted subjects.

Anatomical area of activation	BA	Congenitally blind				Sighted controls			
		Talairach coordinates				Talairach coordinates			
		<i>x</i>	<i>y</i>	<i>z</i>	<i>t</i>	<i>x</i>	<i>y</i>	<i>z</i>	<i>t</i>
<i>Occipital cortex</i>									
Cuneus	18	-26	-75	15	8.12				
		12	-85	19	6.13				
Lingual gyrus	17	-8	-87	4	5.84				
		8	-85	3	4.38				
Inferior occipital gyrus	19	-48	-80	-3	4.02				
		34	-76	-8	4.78				
Middle occipital gyrus	19	-36	-85	6	4.71				
		37	53	-61	-9	6.36			
Superior occipital gyrus	19	36	-73	24	4.54				
<i>Temporal cortex</i>									
Fusiform gyrus	19	-36	-74	-10	3.48				
		38	-64	-5	5.76				
Inferior temporal gyrus (LOtv)	19	-51	-66	-2	4.60				
		37/20	55	-53	-4	5.36	51	-52	-7
Middle temporal gyrus	37	-50	-60	0	5.41	-55	-51	-4	5.43
		53	-62	0	4.59				
Superior temporal gyrus	22					-57	4	2	6.29
						55	10	3	8.49
<i>Parietal Cortex</i>									
Precuneus	31/7	-26	-75	15	8.12	-10	-64	44	5.63
		19/7	26	-70	35	7.57	12	-62	49
Inferior parietal lobule	40	-46	-39	44	6.96	-44	-31	49	9.73
		42	-31	44	5.92	46	-33	44	8.72
Superior parietal lobule	7	-30	-54	51	6.82	-20	-53	58	5.75
		30	-54	52	6.50	32	-52	49	6.82
Postcentral gyrus	2	-53	-25	40	5.65	-50	-25	44	9.49
		3/2	59	-20	34	5.73	51	-28	53
<i>Prefrontal and frontal cortices</i>									
Precentral gyrus	6/44	-61	0	35	4.48	-50	2	11	6.81
		62	5	29	3.73	50	8	9	8.09
Inferior frontal gyrus	9	-55	5	29	5.91	-57	5	26	6.98
		47/9	34	17	-3	6.02	53	9	28
Middle frontal gyrus	6	-26	-2	46	4.87	-38	-3	54	5.87
		30	-1	48	6.37	40	2	50	7.00
Medial frontal gyrus	6					-2	-1	50	6.63
						4	1	50	6.52
Superior frontal gyrus	6					-18	5	62	4.33
						18	5	62	5.08
<i>Cingulate and Insular cortices</i>									
Cingulate gyrus	32	2	10	42	6.31	2	10	42	8.53
Insula	13	-40	5	13	4.76	-34	14	1	8.28
		38	3	15	5.43	34	21	3	7.55
<i>Cerebellum</i>		-32	-74	-22	5.36	-28	-67	-15	8.16
		4	-73	-13	4.34	28	-67	-15	6.35

similarities with that observed in normal seeing subjects during the performance of visual form discrimination tasks (see review by [45]). Pietrini and coworkers [12] reported activations in portions of the ventral stream such as the lingual and fusiform gyri and inferior occipital cortex during haptic object recognition in blind subjects. Other studies have shown that the visual cortex in blind subjects can also be recruited by auditory and olfactory stimuli and cognitive processes [38, 46–48] providing further evidence that the visual cortex can be reorganized to mediate a variety of non-visual functions in the blind.

Another issue is the potential role of mental imagery in the visual cortex activation. A number of previous brain imaging studies on haptic processing [49, 50] and auditory-based sensory substitution [51] in blindfolded sighted subjects have suggested that neural activity related to visual imagery may account for the activation in the occipital cortex. For the following reasons, it is unlikely that visual imagery explains the current findings. First, our blind subjects never had visual experiences and during the debriefing following the experiments, they did not report that they had engaged in visual imagery during the orientation detection task (see also [12]). Secondly, if mental imagery would be at the basis of the activation in the occipital cortex, sighted controls should activate the visual cortex to a larger extent compared to congenitally blind participants, which was clearly not the case. Finally, the question whether congenitally blind subjects have true “visual imagery” (instead of imagery) remains a matter of debate [52].

In accordance with several other studies (e.g., [13, 14, 53, 54]), we report here a lack of activation in the occipital cortex of our blindfolded controls. Previous neuroimaging studies in normally sighted subjects have yielded inconsistent results regarding the implication of V1 in tactile processing: some studies showing no activation of V1, others showing activation of V1 only [18, 55], activation of V1 accompanied by a deactivation of extrastriate areas [56], or activation of extrastriate cortical areas only [12]. Of note, most studies showing V1 activation in tactile processing in normal subjects used 3D stimuli that were palpated haptically with the hand or fingers. In our study, we used 2-D shape stimuli presented passively to the tongue and thus, required no active haptic exploration.

4.3. Possible Mechanisms for Cross-Modal Responses. A critical question in the study of cross-modal processing in the blind is whether the recruitment of the occipital cortex occurs through changes of existing neural network or through the formation of new neural connections. In this study, as well as in our previous studies using the same sensory substitution device, cross-modal responses were already observed after only a four to seven day period of intensive training [13, 22]. The speed with which these neuroplastic changes occur suggest that they are mediated by the unmasking or strengthening of preexisting cortico-cortical connections [13, 22]. The observed striate and extrastriate activations in the blind have been attributed to a cortico-cortical feedback pathway from primary somatosensory cortex (S1) through the posterior parietal cortex [22, 23].

The posterior parietal cortex is a highly multimodal association area. Investigations in macaque and humans have demonstrated that the anterior intraparietal area (AIP) and ventral intraparietal area (VIP) are likely regions where visuotactile multimodal information of object features and motion processing is integrated in sighted participants. Neurons in the macaque AIP, for instance, are sensitive to three-dimensional features of objects such as size, shape, and orientation during object manipulation under visual control [57, 58]. Neuroimaging studies in humans have also demonstrated recruitment of AIP during tactile shape processing [59, 60] and during orientation discrimination of visual stimuli [13, 61]. In blind subjects, who lack bottom-up visual processing, tactile inputs from these multimodal areas may then lead to a recruitment of the visual cortex via these multimodal areas. This assumption is supported by the strong activation of the posterior parietal cortex observed in the blind in the present study and is moreover reinforced by the results of several additional neuroimaging studies [13, 17, 38]. This hypothesis is also in line with a recent report that used dynamic causal modeling of fMRI data to investigate the cross-modal plasticity of effective connectivity in the blind during a Braille reading task [62]. It is also possible that new aberrant subcortical projections could be responsible for the evoked activity in the visual cortex of congenitally blind individuals. For example, animal models of bilateral enucleation in hamsters [60], congenital blindness in mice [63, 64], and natural very low vision like the blind mole rat [65] have indicated the formation of new ectopic projections from the inferior colliculus to the lateral geniculate nucleus, the thalamic primary visual relay. More advanced methods, such as functional connectivity analysis, will be helpful to better understand through which pathways nonhaptic tactile information is funnelled to the visual cortex of the blind.

4.4. Methodological Considerations. The main limitation of this study is the sample size. While eight subjects are considered to be a relatively small sample size for a classical fMRI study, we would like to emphasize that congenitally blind individuals represent an exceptionally rare population, even more so when strict selection requirements are enforced, as in this study. We would further like to stress that sample sizes of congenitally blind individuals reported in most fMRI studies in the literature are similar or smaller as compared to the present one [12–18, 29–31, 38, 39, 54, 55, 62, 66]. Larger numbers of subjects certainly are required to make rigorous statistical comparisons between the sighted and congenitally blind groups in terms of distribution and extent of brain response to shape recognition following stimulation by TDU. Nevertheless, the data were obtained using a random-effects analysis and FDR-corrected statistical thresholds. A final limitation is that we did not use functional localizer scans in our sighted subjects to identify subregions within the ventral stream.

5. Conclusion

The question we have addressed in this and our previous studies is whether the functional segregation of the visual

cortex in a dorsal and ventral visual pathway is preserved in individuals who were born without vision or who lost their sight at a very early age. The present results significantly extend to our previously published data on motion processing via the tongue in the blind [14], showing that both pathways are preserved in this population and add to growing evidence that the visual cortex can be reorganized to mediate non-visual functions in the blind.

Acknowledgments

This work was supported by the Harland Sanders foundation (MP), the Lundbeck Foundation (R. Kupers), Danish Medical Research Council (M. Ptito and RK; Grant no. 09-065632). A. Z. Wang and I. Matteau held a studentship from, respectively, the Dana Foundation and the Canadian Institutes of Health Research (CIHR). The 3 Tesla MRI scanner was donated by the Simon Spies foundation.

References

- [1] O. Collignon, G. Charbonneau, M. Lassonde, and F. Lepore, "Early visual deprivation alters multisensory processing in peripersonal space," *Neuropsychologia*, vol. 47, no. 14, pp. 3236–3243, 2009.
- [2] D. R. Chebat, J. K. Chen, F. Schneider, A. Ptito, R. Kupers, and M. Ptito, "Alterations in right posterior hippocampus in early blind individuals," *NeuroReport*, vol. 18, no. 4, pp. 329–333, 2007.
- [3] A. Fieger, B. Röder, W. Teder-Sälejärvi, S. A. Hillyard, and H. J. Neville, "Auditory spatial tuning in late-onset blindness in humans," *Journal of Cognitive Neuroscience*, vol. 18, no. 2, pp. 149–157, 2006.
- [4] D. Goldreich and I. M. Kanics, "Tactile acuity is enhanced in blindness," *Journal of Neuroscience*, vol. 23, no. 8, pp. 3439–3445, 2003.
- [5] D. Goldreich and I. M. Kanics, "Performance of blind and sighted humans on a tactile grating detection task," *Perception and Psychophysics*, vol. 68, no. 8, pp. 1363–1371, 2006.
- [6] R. W. Van Boven, J. E. Ingeholm, M. S. Beauchamp, P. C. Bikle, and L. G. Ungerleider, "Tactile form and location processing in the human brain," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 102, no. 35, pp. 12601–12605, 2005.
- [7] A. Pascual-Leone and F. Torres, "Plasticity of the sensorimotor cortex representation of the reading finger in Braille readers," *Brain*, vol. 116, no. 1, pp. 39–52, 1993.
- [8] A. G. De Volder, A. Bol, J. Blin et al., "Brain energy metabolism in early blind subjects: neural activity in the visual cortex," *Brain Research*, vol. 750, no. 1-2, pp. 235–244, 1997.
- [9] R. Kupers, P. Pietrini, E. Ricciardi, and M. Ptito, "The nature of consciousness in the visually-deprived brain," *Frontiers in Psychology*, vol. 2, no. 4, 2011.
- [10] L. B. Merabet, J. F. Rizzo, A. Amedi, D. C. Somers, and A. Pascual-Leone, "What blindness can tell us about seeing again: merging neuroplasticity and neuroprostheses," *Nature Reviews Neuroscience*, vol. 6, no. 1, pp. 71–77, 2005.
- [11] U. Noppeney, "The effects of visual deprivation on functional and structural organization of the human brain," *Neuroscience and Biobehavioral Reviews*, vol. 31, no. 8, pp. 1169–1180, 2007.
- [12] P. Pietrini, M. L. Furey, E. Ricciardi et al., "Beyond sensory images: object-based representation in the human ventral pathway," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 101, no. 15, pp. 5658–5663, 2004.
- [13] M. Ptito, S. M. Moesgaard, A. Gjedde, and R. Kupers, "Cross-modal plasticity revealed by electrotactile stimulation of the tongue in the congenitally blind," *Brain*, vol. 128, no. 3, pp. 606–614, 2005.
- [14] M. Ptito, I. Matteau, A. Gjedde, and R. Kupers, "Recruitment of the middle temporal area by tactile motion in congenital blindness," *NeuroReport*, vol. 20, no. 6, pp. 543–547, 2009.
- [15] A. Amedi, W. M. Stern, J. A. Camprodon et al., "Shape conveyed by visual-to-auditory sensory substitution activates the lateral occipital complex," *Nature Neuroscience*, vol. 10, no. 6, pp. 687–689, 2007.
- [16] C. Büchel, C. Price, R. S. J. Frackowiak, and K. Friston, "Different activation patterns in the visual cortex of late and congenitally blind subjects," *Brain*, vol. 121, no. 3, pp. 409–419, 1998.
- [17] H. Burton, "Visual cortex activity in early and late blind people," *Journal of Neuroscience*, vol. 23, no. 10, pp. 4005–4011, 2003.
- [18] H. Burton, D. G. McLaren, and R. J. Sinclair, "Reading embossed capital letters: an fMRI study in blind and sighted individuals," *Human Brain Mapping*, vol. 27, no. 4, pp. 325–339, 2006.
- [19] N. Sadato, A. Pascual-Leone, J. Grafman et al., "Activation of the primary visual cortex by Braille reading in blind subjects," *Nature*, vol. 380, no. 6574, pp. 526–528, 1996.
- [20] N. Sadato, A. Pascual-Leone, J. Grafman, M. P. Deiber, V. Ibañez, and M. Hallett, "Neural networks for Braille reading by the blind," *Brain*, vol. 121, no. 7, pp. 1213–1229, 1998.
- [21] R. Kupers, D. R. Chebat, K. H. Madsen, O. B. Paulson, and M. Ptito, "Neural correlates of virtual route recognition in congenital blindness," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 107, no. 28, pp. 12716–12721, 2010.
- [22] R. Kupers, A. Fumal, A. M. De Noordhout, A. Gjedde, J. Schoenen, and M. Ptito, "Transcranial magnetic stimulation of the visual cortex induces somatotopically organized qualia in blind subjects," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 103, no. 35, pp. 13256–13260, 2006.
- [23] M. Ptito, F. C. G. Schneider, O. B. Paulson, and R. Kupers, "Alterations of the visual pathways in congenital blindness," *Experimental Brain Research*, vol. 187, no. 1, pp. 41–49, 2008.
- [24] L. G. Cohen, P. Celnik, A. Pascual-Leone et al., "Functional relevance of cross-modal plasticity in blind humans," *Nature*, vol. 389, no. 6647, pp. 180–183, 1997.
- [25] R. Kupers, M. Pappens, A. M. De Noordhout, J. Schoenen, M. Ptito, and A. Fumal, "rTMS of the occipital cortex abolishes Braille reading and repetition priming in blind subjects," *Neurology*, vol. 68, no. 9, pp. 691–693, 2007.
- [26] C. Büchel, "Cortical hierarchy turned on its head," *Nature Neuroscience*, vol. 6, no. 7, pp. 657–658, 2003.
- [27] P. Pietrini, M. Ptito, and R. Kupers, "Blindness and consciousness: new lights from the dark," in *The Neurology of Consciousness*, S. Laureys and G. Tononi, Eds., pp. 360–374, Elsevier, Amsterdam, The Netherlands, 2009.
- [28] M. A. Goodale and A. David Milner, "Separate visual pathways for perception and action," *Trends in Neurosciences*, vol. 15, no. 1, pp. 20–25, 1992.
- [29] C. Poirier, O. Collignon, C. Scheiber et al., "Auditory motion perception activates visual motion areas in early blind subjects," *NeuroImage*, vol. 31, no. 1, pp. 279–285, 2006.

- [30] C. C. Poirier, A. G. De Volder, D. Tranduy, and C. Scheiber, "Neural changes in the ventral and dorsal visual streams during pattern recognition learning," *Neurobiology of Learning and Memory*, vol. 85, no. 1, pp. 36–43, 2006.
- [31] E. Ricciardi, N. Vanello, L. Sani et al., "The effect of visual experience on the development of functional architecture in hMT+," *Cerebral Cortex*, vol. 17, no. 12, pp. 2933–2939, 2007.
- [32] J. V. Haxby, M. I. Gobbini, M. L. Furey, A. Ishai, J. L. Schouten, and P. Pietrini, "Distributed and overlapping representations of faces and objects in ventral temporal cortex," *Science*, vol. 293, no. 5539, pp. 2425–2430, 2001.
- [33] I. Matteau, R. Kupers, E. Ricciardi, P. Pietrini, and M. Ptito, "Beyond visual, aural and haptic movement perception: hMT+ is activated by electrotactile motion stimulation of the tongue in sighted and in congenitally blind individuals," *Brain Research Bulletin*, vol. 82, no. 5-6, pp. 264–270, 2010.
- [34] L. G. Underleider and M. Mishkin, "Two cortical visual systems," in *Analysis of Visual Behavior*, M. A. Ingle, M. I. Goodale, and R. J. W. Masfield, Eds., pp. 549–586, MIT Press, Cambridge, Mass, USA, 1982.
- [35] R. Farivar, "Dorsal-ventral integration in object recognition," *Brain Research Reviews*, vol. 61, no. 2, pp. 144–153, 2009.
- [36] A. Amedi, G. Jacobson, T. Hendler, R. Malach, and E. Zohary, "Convergence of visual and tactile shape processing in the human lateral occipital complex," *Cerebral Cortex*, vol. 12, no. 11, pp. 1202–1212, 2002.
- [37] A. Amedi, R. Malach, T. Hendler, S. Peled, and E. Zohary, "Visuo-haptic object-related activation in the ventral visual pathway," *Nature Neuroscience*, vol. 4, no. 3, pp. 324–330, 2001.
- [38] A. Amedi, N. Raz, P. Pianka, R. Malach, and E. Zohary, "Early 'visual' cortex activation correlates with superior verbal memory performance in the blind," *Nature Neuroscience*, vol. 6, no. 7, pp. 758–766, 2003.
- [39] T. W. James, G. K. Humphrey, J. S. Gati, P. Servos, R. S. Menon, and M. A. Goodale, "Haptic study of three-dimensional objects activates extrastriate visual areas," *Neuropsychologia*, vol. 40, no. 10, pp. 1706–1714, 2002.
- [40] C. R. Genovese, N. A. Lazar, and T. Nichols, "Thresholding of statistical maps in functional neuroimaging using the false discovery rate," *NeuroImage*, vol. 15, no. 4, pp. 870–878, 2002.
- [41] T. Kassuba, C. Klinge, C. Hölig et al., "The left fusiform gyrus hosts trisensory representations of manipulable objects," *NeuroImage*, vol. 56, no. 3, pp. 1566–1577, 2011.
- [42] S. C. Prather and K. Sathian, "Mental rotation of tactile stimuli," *Cognitive Brain Research*, vol. 14, no. 1, pp. 91–98, 2002.
- [43] C. L. Reed, S. Shoham, and E. Halgren, "Neural substrates of tactile object recognition: an fMRI study," *Human Brain Mapping*, vol. 21, no. 4, pp. 236–246, 2004.
- [44] M. R. Stoesz, M. Zhang, V. D. Weisser, S. C. Prather, H. Mao, and K. Sathian, "Neural networks active during tactile form perception: common and differential activity during macrospatial and microspatial tasks," *International Journal of Psychophysiology*, vol. 50, no. 1-2, pp. 41–49, 2003.
- [45] A. Amedi, K. Von Kriegstein, N. M. Van Atteveldt, M. S. Beauchamp, and M. J. Naumer, "Functional imaging of human crossmodal identification and object recognition," *Experimental Brain Research*, vol. 166, no. 3-4, pp. 559–571, 2005.
- [46] B. Röder, O. Stock, S. Bien, H. Neville, and F. Röslér, "Speech processing activates visual cortex in congenitally blind humans," *European Journal of Neuroscience*, vol. 16, no. 5, pp. 930–936, 2002.
- [47] F. Gougoux, F. Lepore, M. Lassonde, P. Voss, R. J. Zatorre, and P. Belin, "Neuropsychology: pitch discrimination in the early blind," *Nature*, vol. 430, no. 6997, p. 309, 2004.
- [48] M. Beaulieu-Lefebvre, F. C. Schneider, R. Kupers, and M. Ptito, "Odor perception and odor awareness in congenital blindness," *Brain Research Bulletin*, vol. 84, no. 3, pp. 206–209, 2011.
- [49] M. Zhang, V. D. Weisser, R. Stilla, S. C. Prather, and K. Sathian, "Multisensory cortical processing of object shape and its relation to mental imagery," *Cognitive, Affective and Behavioral Neuroscience*, vol. 4, no. 2, pp. 251–259, 2004.
- [50] S. Lacey, P. Flueckiger, R. Stilla, M. Lava, and K. Sathian, "Object familiarity modulates the relationship between visual object imagery and haptic shape perception," *NeuroImage*, vol. 49, no. 3, pp. 1977–1990, 2010.
- [51] C. Poirier, A. De Volder, D. Tranduy, and C. Scheiber, "Pattern recognition using a device substituting audition for vision in blindfolded sighted subjects," *Neuropsychologia*, vol. 45, no. 5, pp. 1108–1121, 2007.
- [52] D. Kaski, "Revision: is visual perception a requisite for visual imagery?" *Perception*, vol. 31, no. 6, pp. 717–731, 2002.
- [53] N. Sadato, T. Okada, M. Honda, and Y. Yonekura, "Critical period for cross-modal plasticity in blind humans: a functional MRI study," *NeuroImage*, vol. 16, no. 2, pp. 389–400, 2002.
- [54] T. Harada, D. N. Saito, K. I. Kashikura et al., "Asymmetrical neural substrates of tactile discrimination humans: a functional magnetic resonance imaging study," *Journal of Neuroscience*, vol. 24, no. 34, pp. 7524–7530, 2004.
- [55] D. N. Saito, T. Okada, M. Honda, Y. Yonekura, and N. Sadato, "Practice makes perfect: the neural substrates of tactile discrimination by Mah-Jong experts include the primary visual cortex," *BMC Neuroscience*, vol. 7, article 79, 2006.
- [56] L. B. Merabet, J. D. Swisher, S. A. McMains et al., "Combined activation and deactivation of visual cortex during tactile sensory processing," *Journal of Neurophysiology*, vol. 97, no. 2, pp. 1633–1641, 2007.
- [57] A. Murata, V. Gallese, M. Kaseda, and H. Sakata, "Parietal neurons related to memory-guided hand manipulation," *Journal of Neurophysiology*, vol. 75, no. 5, pp. 2180–2186, 1996.
- [58] H. Sakata, M. Taira, A. Murata, and S. Mine, "Neural mechanisms of visual guidance of hand action in the parietal cortex of the monkey," *Cerebral Cortex*, vol. 5, no. 5, pp. 429–438, 1995.
- [59] A. Bodegård, S. Geyer, C. Grefkes, K. Zilles, and P. E. Roland, "Hierarchical processing of tactile shape in the human brain," *Neuron*, vol. 31, no. 2, pp. 317–328, 2001.
- [60] L. Jäncke, A. Kleinschmidt, S. Mirzazade, N. J. Shah, and H. J. Freund, "The role of the inferior parietal cortex in linking the tactile perception and manual construction of object shapes," *Cerebral Cortex*, vol. 11, no. 2, pp. 114–121, 2001.
- [61] E. Shikata, F. Hamzei, V. Glauche et al., "Surface orientation discrimination activates caudal and anterior intraparietal sulcus in humans: an event-related fMRI study," *Journal of Neurophysiology*, vol. 85, no. 3, pp. 1309–1314, 2001.
- [62] T. Fujii, H. C. Tanabe, T. Kochiyama, and N. Sadato, "An investigation of cross-modal plasticity of effective connectivity in the blind by dynamic causal modeling of functional MRI data," *Neuroscience Research*, vol. 65, no. 2, pp. 175–186, 2009.
- [63] N. Chabot, S. Robert, R. Tremblay, D. Miceli, D. Boire, and G. Bronchti, "Audition differently activates the visual system in neonatally enucleated mice compared with anophthalmic mutants," *European Journal of Neuroscience*, vol. 26, no. 8, pp. 2334–2348, 2007.
- [64] N. Chabot, V. Charbonneau, M. E. Laramée, R. Tremblay, D. Boire, and G. Bronchti, "Subcortical auditory input to the

primary visual cortex in anophthalmic mice,” *Neuroscience Letters*, vol. 433, no. 2, pp. 129–134, 2008.

- [65] G. Bronchti, P. Heil, R. Sadka, A. Hess, H. Scheich, and Z. Wollberg, “Auditory activation of ‘visual’ cortical areas in the blind mole rat (*Spalax ehrenbergi*),” *European Journal of Neuroscience*, vol. 16, no. 2, pp. 311–329, 2002.
- [66] H. Burton, R. J. Sinclair, and D. G. McLaren, “Cortical activity to vibrotactile stimulation: an fMRI study in blind and sighted individuals,” *Human Brain Mapping*, vol. 23, no. 4, pp. 210–228, 2004.

Review Article

Cortical GABAergic Interneurons in Cross-Modal Plasticity following Early Blindness

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Received 7 February 2012; Accepted 4 April 2012

Academic Editor: Ron Kupers

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Early loss of a given sensory input in mammals causes anatomical and functional modifications in the brain via a process called cross-modal plasticity. In the past four decades, several animal models have illuminated our understanding of the biological substrates involved in cross-modal plasticity. Progressively, studies are now starting to emphasise on cell-specific mechanisms that may be responsible for this intermodal sensory plasticity. Inhibitory interneurons expressing γ -aminobutyric acid (GABA) play an important role in maintaining the appropriate dynamic range of cortical excitation, in critical periods of developmental plasticity, in receptive field refinement, and in treatment of sensory information reaching the cerebral cortex. The diverse interneuron population is very sensitive to sensory experience during development. GABAergic neurons are therefore well suited to act as a gate for mediating cross-modal plasticity. This paper attempts to highlight the links between early sensory deprivation, cortical GABAergic interneuron alterations, and cross-modal plasticity, discuss its implications, and further provide insights for future research in the field.

1. Introduction

Patterns of activity from the peripheral sensory receptor arrays can dramatically influence the development of connectivity and functional organization of cortical fields in mammals. In some species, evolution in relation to specific environmental cues has nurtured the brain's blueprint in such a way that a sensory cortex processing specific survival needs has been enlarged over time as compared to other modalities (Figure 1) [1–5]. Similarly, when a sensory function is lost during development, spared senses compensate by taking more cortical space and recruiting the deafferented areas, to maintain homeostasis of sensory function. This reorganization optimizes and secures the individual's survival and awareness to future environmental changes. For example, the loss of sight at birth or during early life in humans leads to important anatomical and functional

reorganization of the visually deprived cortex that will become activated by a wide variety of nonvisual stimuli involving touch, audition, and olfaction [6–11]. Enhanced spatiotemporal functions in the remaining sensory modalities have also been reported [12–16]. It seems therefore that the visual cortex of the blind is not lifeless and is capable of adapting in order to accommodate these nonvisual inputs through cross-modal plasticity.

But how does a visually deprived cortical area signal its loss of sensory inputs to, or be recruited by, areas of other sensory modalities? Two main hypotheses have been proposed to explain cross-modal plasticity in a visually deprived brain. The first hypothesis proposes that early deprived visual cortical circuits can be rewired and/or cross-wired with other modalities following the initial insult [17–19]. This rewiring stipulates the formation of new and permanent aberrant connections from the sensory receptors of spared modalities

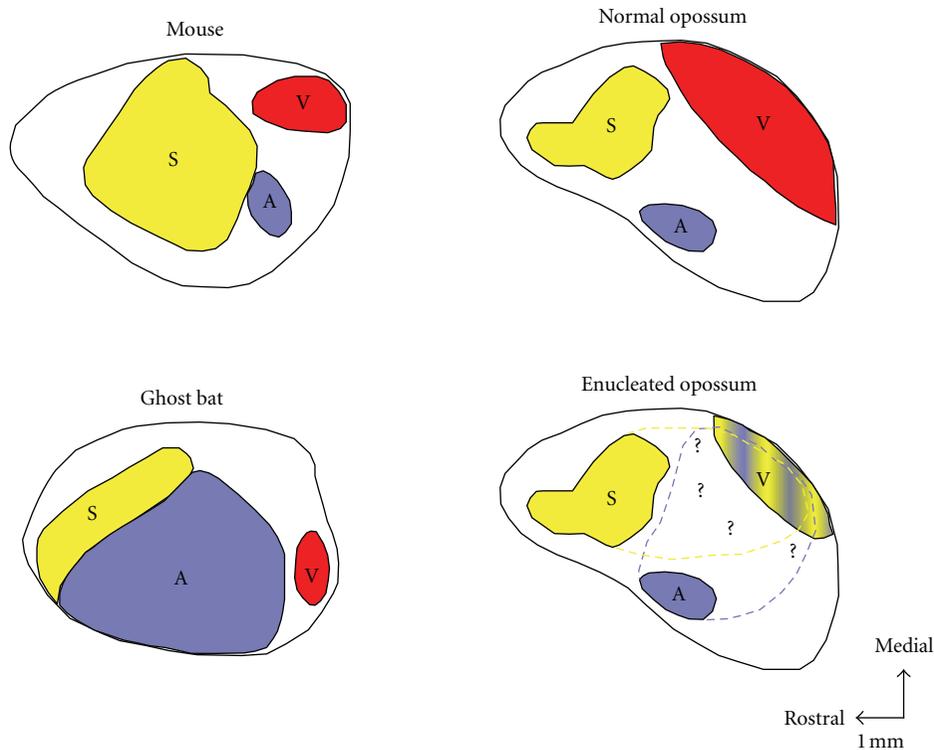


FIGURE 1: Primary cortical areas in three species of mammals (i.e., Mouse, Ghost Bat and Opossum) that have approximately the same size cortical sheet, but different amounts of cortex allowed to different sensory modality (S: Somatosensory system, A: Auditory system and V: Visual system), related to use of particular sensory receptor arrays. In the mouse (top left), which relies heavily on tactile inputs from the whiskers for survival, the somatosensory cortex (S) is enlarged, compared with the ghost bat (bottom left) and normal opossum (top right). The auditory cortex (A) in the neocortex of the echolocating ghost bat is expanded, while the visual area (V) and S is relatively small. Similarly, the cortex of the highly visual opossum have a dominant visual cortex. Finally, for example, in the enucleated at birth opossum (bottom right) the V cortex becomes smaller and is recruited by the A and S modalities. Similarity in relative location of sensory cortical fields in all these mammals suggests that the topographic organization and overall pattern of thalamocortical projections of the brain is constrained by developmental mechanisms. Conversely, the differences in size, shape, and detailed organization of sensory cortical fields indicate that input from the periphery is a crucial factor in guiding many of the details of organization of the neocortex. Rostral is to the left and medial is up. Scale bar = 1 mm. Adapted from Kahn and Krubitzer, 2002 [48].

to visual thalamic relays and up into the visual cortices. The second hypothesis stipulates the activation, formation, and/or enhancement of corticocortical connections that involve local connectivity modifications in the deprived cortex as well as physically present but functionally silent connections between sensory cortices that could therefore be activated and/or sprout following a specific sensory loss. Thus, early blindness could lead either to abnormal thalamocortical or corticocortical connections. These connections are not yet fully understood but would explain, in part, how the afferents of the remaining modalities could reach the deprived cortex. In order to clarify these hypotheses at the microscale level and to better understand the biological underpinnings of cross-modal plasticity, several early developmental models have been developed in the past four decades.

Even if it is now widely accepted that cross-modal plasticity involves important anatomical and functional changes in the neocortex, its cellular mechanisms are still ill-defined. Inhibitory GABAergic interneurons are believed to

subserve cross-modal plasticity processes such as in re-establishing homeostasis when the excitation-inhibition balance is perturbed. For example, GABAergic neuronal activity coordinates the rhythmic behavior of principal (excitatory) neurons in the cortical networks. GABAergic neurons are also critically involved in neuronal growth, fine tuning of sensory receptive fields, visual plasticity, and the formation of critical periods in development. In addition, GABAergic interneurons especially those expressing calcium-binding proteins like calbindin (CB), calretinin (CR), and particularly parvalbumin (PV) have a protracted development reaching their neurochemical and innervation maturity only during early postnatal life making them very sensitive to sensory experience, sensory deprivation, and noxious environmental changes. Finally, GABAergic interneurons play a pivotal role in gating sensory thalamocortical feed-forward inputs [20–22], cortico-cortical [23–25], and corticothalamocortical connectivities between visual cortices [26] which is of prime interest for cross-modal plasticity following an early sensory loss. Further, significant morphological alterations

in inhibitory networks are found in animal models of sensory deprivation, early blindness, rewiring, and cross-modal plasticity [1, 27–30]. Since interneurons play such a significant role in activity-dependent modification of developing sensory circuits, it is thus important to study specific implications of the various GABAergic subpopulations in cross-modal plasticity paradigms.

In this paper we will first discuss general anatomical and functional findings in different animal models of cross-modal plasticity (in particular hamsters, ferrets, opossum, and mice) and the effects of loss of sensory function on GABAergic cortical networks. We will then focus on how aberrations in inhibitory circuitry could explain cross-modal plasticity and briefly discuss future research directions in the field. Thus, the main objective of this paper is to stress out the importance of studying the GABAergic networks in animal models of cross-modal plasticity for future experimental work because information on the possible mechanisms involved is presently lacking.

2. Animal Models of Cross-Modal Plasticity

During development, activity pattern amongst different sensory modalities determines the relative size and organization of its representative subcortical and cortical areas. The loss or decrease of any one modality leads to the invasion of the deprived cortical area by inputs originating from other modalities, illustrating the remarkable capacity of the cerebral cortex for plasticity resulting in anatomical reorganization, functional and behavioral recovery. As mentioned before, in blindness, cross-modal changes most probably require or involve rewiring and cross-wiring of cortices. The remaining modalities could colonize the deprived visual cortex directly through changes at the subcortical level (modified thalamo-cortical afferences) and via cortico-cortical connections.

2.1. Early Sensory Privation, Binocular Enucleation, and Congenital Blindness Models. Several studies have shown that the total or partial loss of a sensory modality like vision leads to changes in the anatomical and functional organization of the structures associated with the affected sensory input as well as from the spared modalities [31, 32]. In the 1970s, Rebillard et al. reported for the first time that the primary auditory cortex in congenitally deaf cats could be driven by visual or somatosensory stimuli [33] and was later shown also in a congenitally deaf mouse strain [34]. Furthermore, following bilateral lid sutures at birth in kittens it was also found that the neurons in the visual part of the anterior ectosylvian cortex (AEV) could respond to other modalities [35, 36]. Thus, areas normally dedicated to vision could be taken over by neighbouring auditory and somatosensory areas leading to superior performance in localization discrimination tasks relying on these remaining senses. These changes were attributed to the expansion of the auditory and somatosensory areas to the detriment of extrastriate or associative visual cortices [37, 38]. Cross-modal plasticity has also been observed in primary sensory

cortices. In rats enucleated at birth, the primary somatosensory cortex (S1) can recruit the rostral part of primary visual cortex (V1) conferring functional tactile neuronal responses in that area. These rats showed better exploring skills and higher whisker responsiveness than control siblings [39, 40]. Similar anatomical and electrophysiological findings have been reported in early postnatal and adult enucleated mice and rabbits [41–43]. Enucleation at birth or congenital microphthalmia in kittens induces auditory activation of the visual cortex, principally area V1 [44, 45], which has also been shown in hamsters, opossums, and mice [46–49].

In very low-sighted mammals like the blind mole rat (*Spalax ehrenbergi*), the primary visual thalamic relay, the dorsal lateral geniculate nucleus (dLGN), receives direct atypical subcortical projections from the inferior colliculus (IC) which gets its auditory input from the cochlea [50]. Hence, in these animals, an auditory stimulus can activate neurons in both the dLGN and area V1 [51–53]. Congenitally anophthalmic mutant mice (strain ZRDCT-An) show similar responses. When the dLGN-V1 connectivity is preserved in these mice, there is an increase in the thalamo-cortical projections coming from the lateral posterior (LP) (ancestor of the pulvinar in rodents) and the somatosensory ventro-posterior (VP) nuclei [54]. Further, there is development of ectopic innervations of the dLGN and the LP by inputs originating in the dorsal column nuclei (DCN) of the somatosensory system and the IC [55–57]. Chabot et al. found that an auditory stimulation provoked a strong c-fos response in cells of the dLGN and V1 and to a lesser extent in secondary associative visual cortices (V2M and V2L) in these anophthalmic mutants only, compared to C57BL/6 normal controls and enucleated at birth mice that do not develop aberrant projections between IC and the dLGN [46, 56]. More recently, an elegant study using mice mutants that lacked functional rods (*Gnat-/-*), but had normal cone function, reported that cortical connections of V1 in these animals were similar to those of normal siblings, but there were sparse inputs from the auditory cortex (AC) to area V1. This region also received some abnormal subcortical inputs from the anterior thalamic nuclei, the ventral posterior, the ventral lateral, and the posterior nuclei. While vision generated from a small number of cones appeared to be sufficient to maintain most of the patterns of normal connectivity, the sparse abnormal thalamic inputs to V1, existing inputs from AC, and possibly abnormal inputs to LG and LP may be responsible for generating alterations in the functional organization of V1 of these mice [32]. Taken together, these studies suggest that developmental timing and age at which sensory loss happens are of prime importance to the strength of rewiring and cross-wiring that occurs in cross-modal plasticity. These studies also imply that a prenatal period of spontaneous retinal and/or basic postnatal retinal activities may play a role in shaping differences in sensory reorganization in mammals. This corroborates results obtained in prematurely born animals like hamsters (E15) and opossums (E13,5) in which subcortical reconnections are generally more important. For example, binocular enucleation at birth in these animals also induces strong auditory responses in the primary visual

cortex [31, 47, 58, 59]. In enucleated hamsters, single cell electrophysiological recordings have shown that 63% of neurons in the V1 are now responsive to auditory stimulation. This manipulation leads to the formation of direct new ectopic auditory projections from the IC to the dLGN while the connectivity between this nucleus and the visual cortex remains unchanged (Figures 2(a) and 2(c)). These new auditory inputs to the visual thalamus and cortico-cortical connections from A1 are thought to be responsible for the auditory activities found in V1 [47]. Interestingly, reducing or ablating visual thalamo-cortical inputs on the day of birth in normal hamsters significantly increases the number of cortico-cortical projections to V1 arising from both primary nonvisual and associative visual areas in the adult [60]. In enucleated opossums, the cross-modal plasticity and alterations of the subcortical and cortico-cortical afferent circuits are stronger. As a result, in these marsupials, area V1 can now receive ectopic projections from the primary thalamic auditory (medial geniculate nucleus (MG)) and somatosensory (ventrobasal nucleus (VB)) relays as well as new inputs from primary auditory (A1) and somatosensory (S1) cortices. However, no projections between the IC and the dLGN were seen in this model [31]. For comparison purposes, a similar enucleation paradigm in the rat reported a reduction in thalamo-cortical afferents from the dLGN to the visual cortex, a significant increase in the projections from the LP to the V1, and more cortico-thalamic projections between the S1 and the LP in these rats [61].

As previously mentioned, it is possible for auditory and/or somatosensory information to reach area V1 in blind mammals via modifications of the cortico-cortical connectivity. Several animal studies in the past decade show the existence of direct anatomical connections between the auditory and visual cortices, more particularly in normal-sighted cats and nonhuman primates [62–66]. Long projections relaying the V1 to other primary sensory cortices are found in several mammalian species such as rats [67], gerbils [68], hamsters [47, 60], and ferrets [69]. Recently, an indirect pathway between the primary auditory and visual cortices through layer V pyramidal neurons in V2L has been identified in the mouse and can be amplified by enucleation at birth. The authors suggest that this A1-V2L-V1 pathway may be involved in multisensory processing and contribute to the auditory activation of the occipital cortex in blind rodents [70]. It is possible that such cortico-cortical connections in normal animals contribute to cross-modal plasticity by being stabilized, reorganized and/or being amplified following any form of sensory loss during development. Taken as a whole, studies so far highlight the importance of putative reorganization of subcortical, thalamo-cortical, and cortico-cortical pathways in the blind brain.

2.2. Artificial Rewiring Paradigms in Hamsters and Ferrets Neonates. Cross-modal plasticity changes have also been studied by surgically creating new visual circuits. Schneider pioneered this approach and showed that a lesion of the visual and superficial layers of the superior colliculus (SC) at birth in hamsters (that constantly give birth prematurely

at E15) could produce ectopic retinal projections, from surviving ganglionic cells to subcortical sensory relays that normally receive small or no visual inputs [71]. For example, a bilateral lesion of the stratum opticum on postnatal day 1 leads to a fourfold amplification of retinal synapses in the lateral posterior nucleus (LP) of the thalamus, which is a secondary associative visual relay connected to the lateral secondary visual cortex (V2L) in rodents [72, 73] (Figures 2(a) and 2(b)). Frost (in hamsters) and Sur (in ferrets) were the first to optimize this model and demonstrate that, in combination to the superficial SC lesion at birth, surgically cutting the auditory (i.e., the inferior colliculus brachium) or somatosensory (i.e., the medial lemniscus) afferents could lead to the formation of new robust ectopic retinal projections to the auditory medial geniculate nucleus (MG) (Figures 3(a), 3(b)) or to the somatosensory ventrobasal nucleus (VB), respectively [74–80]. At birth thalamo-cortical projections from primary sensory thalamic relays have not yet reached the cortical subplate (this happens at P1 in hamsters and P14 in ferrets). Therefore, by using this experimental paradigm one can alter the nature of sensory activity that reaches the primary auditory or somatosensory cortices during development without changing the original thalamo-cortical connectivity. The new retinal projections inducted in MG or VB are from the three main classical ganglion cell types, form functional synapses, and are retinotopically organized in the host primary auditory (A1 (Figure 3(c))) or somatosensory (S1) cortex, respectively [78–84]. Nevertheless, the molecular and cellular mechanisms involved in the formation of these new ectopic connexions are still unanswered. In the ferret, the morphology of the retinal synapses in the MG are similar to the ones found in the visual CS and dLGN in control animals [85, 86]. Further, retinal afferents conserve their visual organization in the auditory relays [87, 88]. Although the tonotopic organization of thalamo-cortical projections is preserved between MG and A1 in these rewired ferrets [89, 90], it has been shown that the horizontal network in the auditory cortex as well as its contralateral callosal projections is largely modified and is very similar to those normally found in the V1 [91, 92]. *In vivo* electrophysiological recordings of single neurons in both the A1 and S1 indicated that these cells have acquired functional receptive field properties of the visual cortex (i.e., orientation selectivity, motion and direction sensitivity) (see Figure 4) and some also show a bimodal response [83, 93–97]. Using intrinsic signal optical imaging in the A1 of rewired ferrets, visual orientation selectivity columns were found to be similar but broader than those in the V1 of control animals [98–102]. Furthermore, at the behavioral level, these rewired animals can learn visual discrimination tasks and perceive vision with the rewired auditory cortex [95, 103, 104]. Rewired hamsters with no visual cortex can learn visual tasks as well as normal animals, and a lesion of the auditory cortex abolishes this ability and function (Figures 5(a) and 5(b)). In fact, rewired hamsters with auditory cortex lesions exhibit cortical blindness similar to nonrewired hamsters with visual cortex ablations. Overall these results involving intermodal rewiring in neonatal hamsters and ferrets show

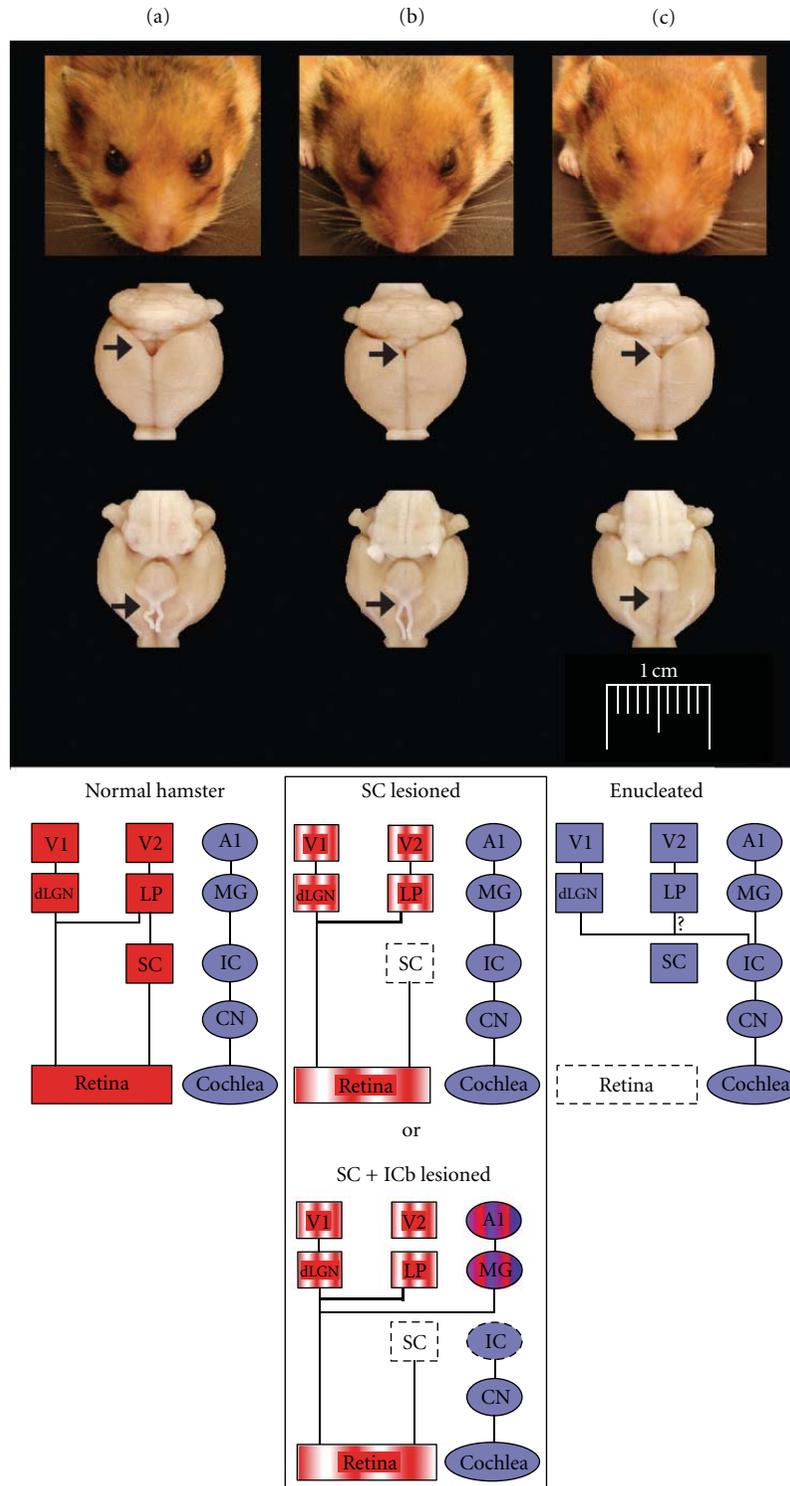


FIGURE 2: Hamster models of cross-modal plasticity. Photomicrographs examples of normal (a), SC lesioned (b) and enucleated (c) hamsters. Column a: top figure showing normal hamster brain with intact superior colliculus and optic chiasm (black arrow heads). At the bottom a simplified schematic representation of the normal visual and auditory pathways. Column b: At the top Superior colliculus (SC) lesioned hamster brain were the SC and optic chiasm are atrophied (black arrow heads). Underneath, diagrams showing the new ectopic retinal projections to the LP in the SC lesioned and to the MG in the SC + ICb lesioned animals. Column c: Enucleated case with an evident SC but complete absence of optic nerves and optic chiasm (arrows). Bottom diagram illustrating the new ectopic auditory projections between the IC and the dLGN to V1. V1, primary visual cortex; V2L, lateral secondary visual cortex; A1, primary auditory cortex; dLGN, dorsal lateral geniculate nucleus; LP, lateral posterior nucleus; MG, medial geniculate nucleus; SC, superior colliculus; IC, inferior colliculus; ICb, inferior colliculus brachium; CN, cochlear nucleus.

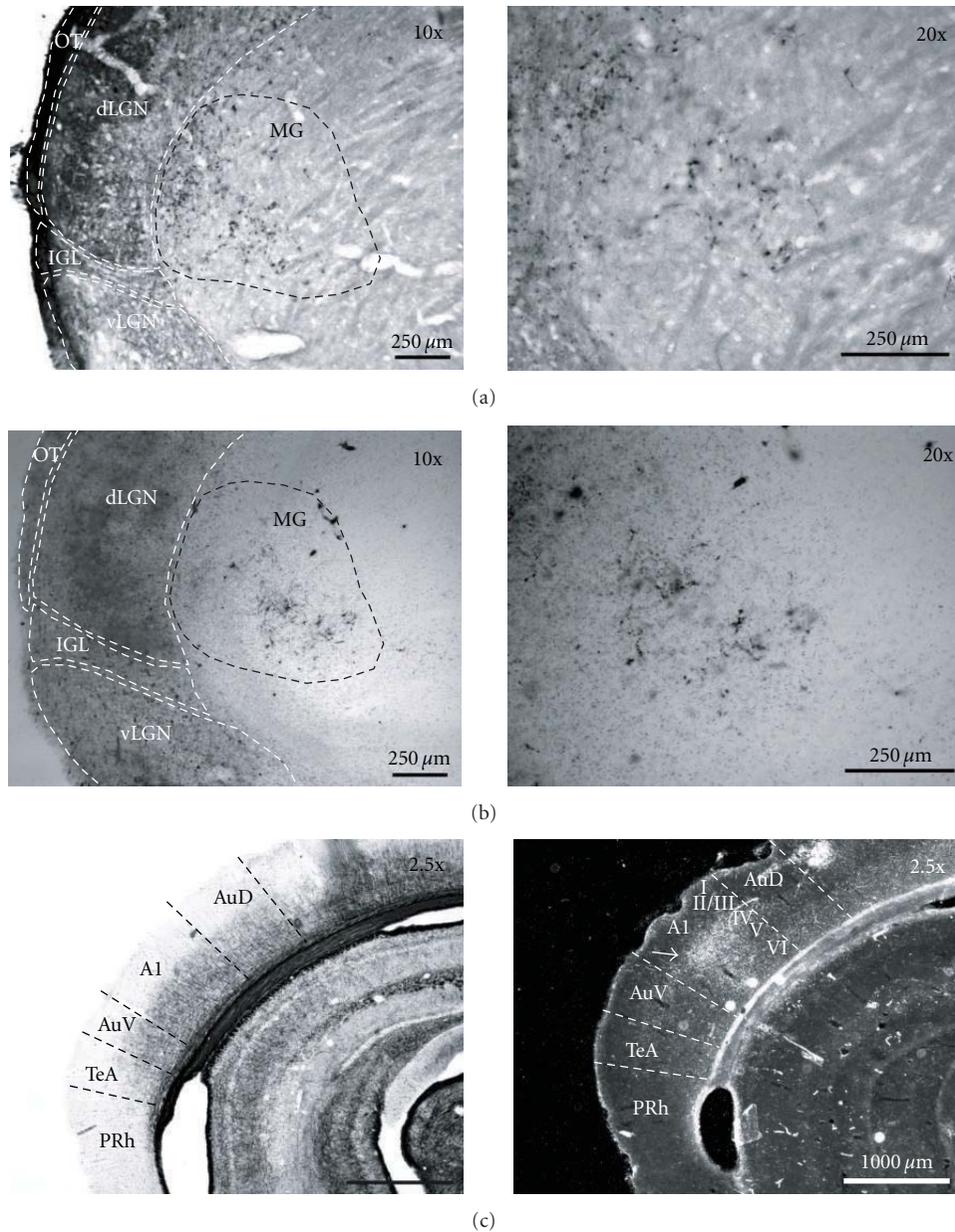


FIGURE 3: Ectopic retinal projections to the medial geniculate nucleus (MG) in the SC + ICb lesioned (or rewired) hamster. (a) Retinal projections in the MG labelled by intraocular injection of the cholera toxin β fragment and (b) co-tagged with wheat germ agglutinin-horse radish peroxidase (WGA-HRP). (c) Gold chloride myelin staining (left panel) and transneuronal labelling, with WGA-HRP (right panel), of new visual thalamo-cortical afferences reaching namely cortical layer IV in the primary auditory cortex (A1) (white arrow head). A1, primary auditory cortex; AuD, dorsal secondary auditory cortex; AuV, ventral secondary auditory cortex; TeA, temporal association cortex; PRh, perirhinal cortex; dLGN, dorsal lateral geniculate nucleus; vLGN, ventral geniculate nucleus; LP, lateral posterior nucleus; MG, medial geniculate nucleus; ot, optic track; IGL, intergeniculate leaflet.

that sensory information via subcortical thalamic afferents play an important role in shaping anatomical and functional specifications of primary sensory cortices. This suggests that the type of sensory activity and experience can play an important role in forging parts of the neuroarchitecture of the hosting cortex [1, 4, 105, 106].

2.3. Can Multisensory Integration Already Be Present in Normal Primary Sensory Cortices? The classical modality

exclusivity of primary sensory areas has recently been challenged. Observations in a variety of species suggest that each of these domains could already be subjected to influences from other senses in normally reared animals. The first evidence was found in the early 1970s where a study, contested at the time, showed that auditory stimuli could elicit neuronal activity in primary (area 17) and secondary (area 18) visual cortices of normal cats [107]. More recently, transitional multisensory zones of multimodal responsive

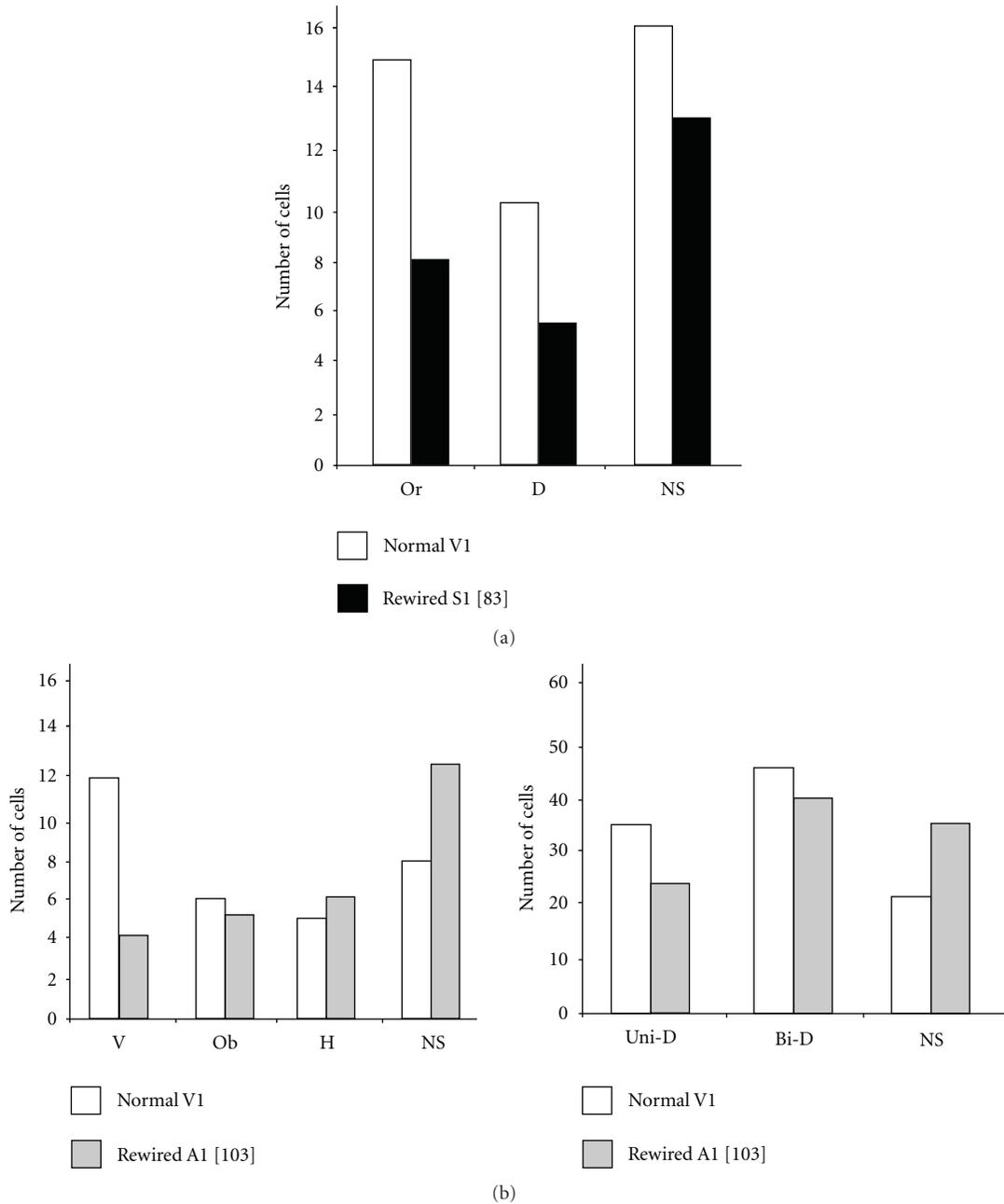


FIGURE 4: Visual properties of single neurons in auditory and somatosensory cortices of rewired hamsters. These cells that responded to visual stimuli showed orientation selectivity, motion and direction sensitivity with receptive field properties similar with those obtained from neurons in the visual cortex of normal hamsters. (a) Examples of visual responsive neurons in the somatosensory cortex of hamsters with new retinal projections in the somatosensory ventrobasal nucleus (VB) of the thalamus adapted from Metin and Frost [83]. (b) Receptive field properties of visual neurons found in the auditory cortex of hamsters with ectopic retinal terminals in the auditory medial geniculate nucleus (MG). Orientation (left panel) and direction (right panel) selectivity adapted from Frost and collaborators [103]. V, vertical orientation; Ob, oblique orientation; Or, orientation selective; H, horizontal orientation; D, direction selective; Uni-D, unidirectional; Bi-D, bidirectional; NS, non-selective neuron.

neurons have been reported at the border of the primary visual cortex in rats [108]. Other recent electrophysiological studies in cats, ferrets, and monkeys have highlighted a low-level influence of other sensory modalities on auditory areas including A1 [69, 109, 110]. These results suggest the possible existence of an important multisensory convergence,

occurring at low hierarchical levels, of sensory cortical areas involving feedback, feedforward and lateral cortico-cortical connections and also subcortical inputs. This way a sensory area processing one particular modality could have access, simultaneously, to other unimodal and polymodal sensory information [18, 111]. For example, it is possible that in

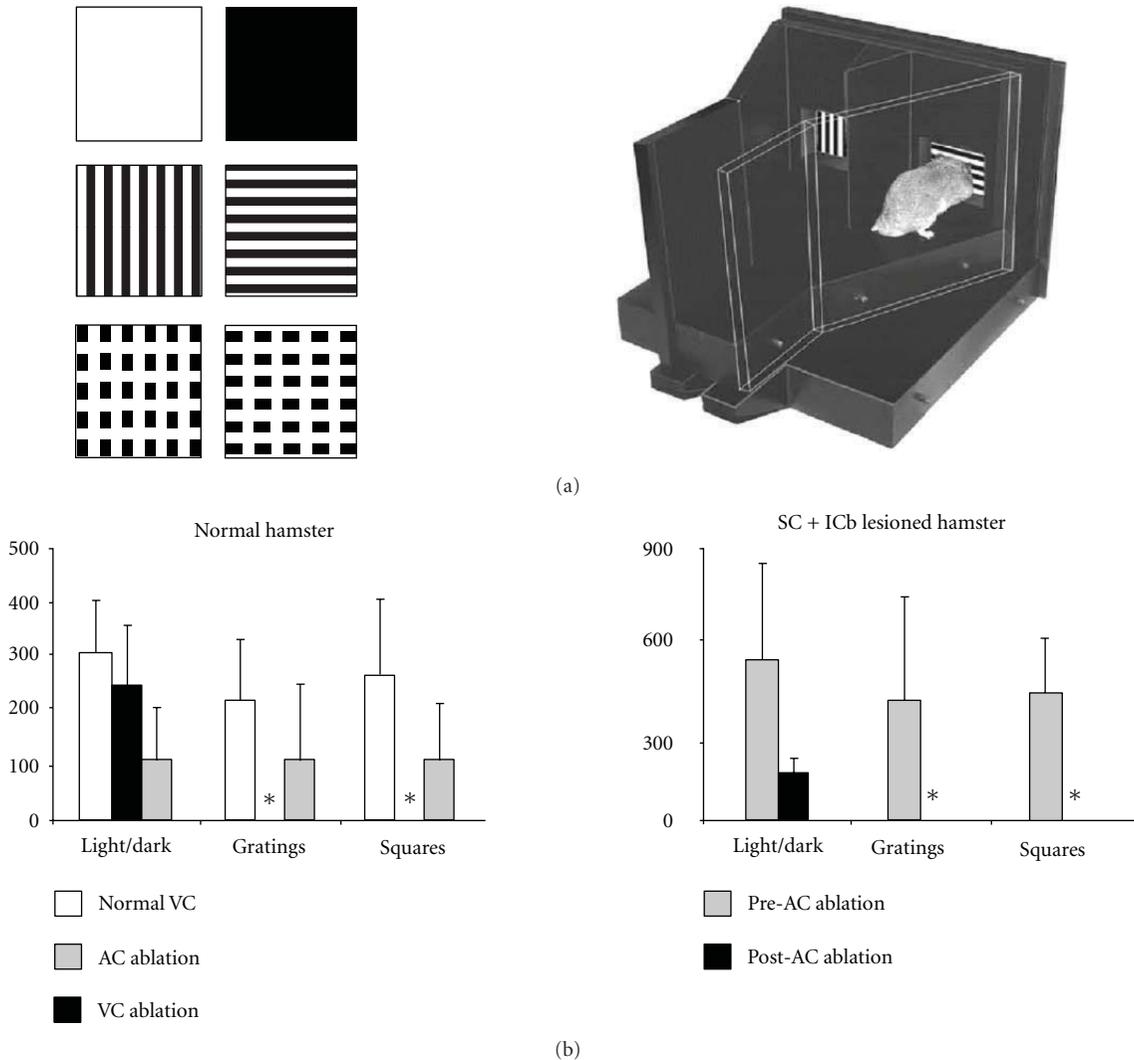


FIGURE 5: Visually guided behavior in SC + ICb lesioned (or rewired) hamsters. (a) Example of the experimental setup with visual stimuli and Y maze. (b) Histograms showing trials to criterion on the visual discrimination tasks in normal hamsters before and after ablation of visual (VC) and auditory (AC) cortices. (c) Behavior of rewired hamsters before and after AC lesions. At the behavioral level, rewired hamsters can learn visual discrimination tasks as well as normal ones and a lesion of the auditory cortex abolishes this function. In fact, SC + ICb lesioned hamsters with auditory cortex lesions exhibit cortical blindness (*) similar to normal hamsters with visual cortex lesions. These results provide strong evidence for sensory substitution where a given sensory modality acquires the functional properties of a missing one. Adapted from Frost et al. [103] and Ptito et al. [95].

visually deprived animals or blind humans these putative interactions are modified to permit a greater recruitment of the primary visual cortex by the spared modalities.

At this point we can assert that cross-modal plasticity that occurs following an early sensory function loss involves important rewiring and cross-wiring processes. However, the question of how thalamo- and cortico- cortical changes, as well as new multisensory integrations, are taking place remains unresolved. A possibly significant mechanism may involve the cortical inhibitory (or GABAergic) interneurons since they are important for visual cortex plasticity and for refinement of sensory information reaching the cortex.

3. The Importance of Cortical GABAergic Interneurons

The neocortex contains mainly two neuronal types, excitatory (glutamatergic) pyramidal cells and inhibitory non-pyramidal (GABAergic) neurons. These aspiny interneurons are widespread and represent only 15–30% of all neocortical neurons. Inhibitory interneurons include a vast array of subtypes that vary in morphological, physiological, and neurochemical characteristics (e.g., calcium-binding proteins, neuropeptides, ion channels, receptors, and transporters). Further, they target their synapses onto distinct subcellular

locations at the postsynaptic level [112–115]. In the cortex, different GABAergic interneuron subtypes were originally classified by expression of calcium-binding proteins parvalbumin (PV), calbindin (CB), or calretinin (CR). Recently, a more accurate classification through expression of several neuropeptides suggests that most inhibitory interneurons in the cortex can be classified in three largely independent populations expressing PV, CB/somatostatin (SS), and CR/vasointestinal peptide (VIP) [112, 113, 115, 116]. Exactly how many specific interneuron subtypes actually exist in the cortices of different species [112, 117–119] is still a matter of debate. Inhibition is critical to a wide range of brain processes specifically in network oscillations and synchronisation, synaptic plasticity, and in preventing runaway excitation. In addition, GABAergic interneurons also regulate nearly all key developmental steps in the cortex, from neuronal proliferation, migration, and differentiation to experience-dependent refinement of local cortical circuits. As a result, disturbances in inhibitory circuits have been involved in a number of neurological disorders, such as epilepsy, schizophrenia, autism, anxiety disorders, and Alzheimer's disease [116, 120, 121]. These GABAergic interneurons are present in all sensory modalities. In sensory cortices interneurons play an essential role in refining sensory receptive fields as well as in confining and modulating sensory afferent activity to sensory cortices. For example, in the visual cortex they shape spacing between cortical columns and strongly influence ocular dominance plasticity [20–22, 122–128].

The large diversity of interneurons suggests that individual inhibitory classes may have unique roles in arbitrating the balance between excitation and inhibition in cortical circuits and plasticity. Development of GABAergic circuits is a prolonged process that begins during midgestation and is complete only by the end of adolescence [114, 116]. PV-positive cells represent the largest subgroup of the GABAergic population in sensory neocortices. Amongst inhibitory neurons in the cortex, the PV subtype is the last to mature in rodents, human and nonhuman primates [129]. The prolonged development of interneurons may constitute a sensitive period where environmental changes can lead to permanent alterations in the inhibitory circuitry. Considering the numerous roles played by GABAergic interneurons in the development, function, and plasticity of cortical networks, combined with their late maturation in the postnatal life, it is reasonable to hypothesize that they could be a key component in gating cross-modal plasticity processes following early sensory deprivation. We will discuss several lines of evidence that stress their possible implication in this context.

3.1. Effects of Sensory Deprivation on the Expression of Calcium-Binding Proteins (CBPs) in Cortical GABAergic Interneurons. A large number of studies show that the expression of calcium-binding proteins (CBPs) in cortical GABAergic interneurons can be altered significantly in different sensory areas, olfactory bulb, and in the hippocampus following modifications to the afferent sensory input [130–138]. For example, deafferentation of the primary auditory

cortex in ferret at P14 (two weeks before auditory capability) by bilateral ablation of the cochlea causes a reduction in the density of neurons immunoreactive (IR) for GABA, PV, and CB [105]. Ibotenic acid lesions of thalamic afferents from the somatosensory ventroposterior nucleus delay the development of PV and CB interneurons in the rat barrel cortex [139]. More precisely the visual cortex has mostly been studied in rodents, carnivores, and nonhuman primates in that context. Dark rearing, retinal lesions, TTX intraocular injections, mutated loss of photoreceptors in the retinae during development, visual deprivation by eye lid sutures or enucleation have all been shown to modify the expression of the different CBPs (PV, CB, and CR) [140–146]. This was shown for the first time in rats where a monocular enucleation, performed before the critical period at P14, could induce a significant decrease in PV expression in interneurons in the contralateral binocular zone of the primary visual cortex (V1) [142, 147]. The authors had postulated an important role for PV-expressing interneurons in ocular dominance plasticity in this paper. Similar results were also found in the primary visual cortex of adult macaques, wherein following monocular inactivation by intraocular injection of TTX, both PV and CB but not CR were affected in ocular dominance columns associated with the deprived eye. More specifically, in these columns there was significant decrease in the number of neurons, as well as neuropil expressing PV and CB in cortical layers IV to VI and II/III, respectively [141]. Analogous changes were also observed following monocular enucleation or retinal lesions. The main finding was the varying density of perineuronal nets expressing these neurochemical markers between layers II/III and V [140, 143]. In experiments using dark-reared mice, a decrease in the expression of PV but not CB and CR mRNA levels was observed in V1, while monocular deprivation induced no changes [144]. Further studies done in cats show that following dark rearing from birth there is a significant decrease in the total number of cells expressing PV and CR but not CB in areas 17 and 18 [145]. Long-term monocular deprivation in rats decreases the expression of CB but not CR and PV in primary visual cortex, suggesting that the CBPs-IR neuronal subpopulations may be differently affected by the various types of visual deprivation paradigms [148]. More recently, distribution of PV-IR and CB-IR interneurons was studied in the primary visual cortex of CRX^{-/-} mice, where photoreceptors lack outer segments resulting in the complete absence of vision from birth as compared to C57Bl/6 controls [149]. In CRX mutants, there is significant decrease in PV in all layers and of CB only in layers II/III of the primary visual cortex. Developmental results in these mutant mice further suggest that PV expression requires visual activity in V1. In hamsters a lesion of the stratum opticum layer of the SC at birth leads to the death of ~70% of the primary retinal projections to the dLGN-V1 pathway and a fourfold increase in the remaining afferents to the lateral posterior nucleus (LP)-V2L route. Interestingly, in these animals we found changes in the number of PV- and CB- immunoreactive neurons in V1 and V2L as compared to intact hamsters. More precisely, these two populations of neurons were decreased in layer V of

V1, but PV interneurons were significantly increased in layer V of V2L in SC-lesioned animals [30] (Figure 6(c)). These results suggest that the decline in visual activity influences PV and CB expressions only in layer V of area V1 whereas the increase of PV-IR cells in layer V of V2L may be correlated with the nurturing presence of new ectopic retinal projections in LP.

Overall, these studies support the idea that different subpopulations of GABAergic neurons are differently influenced by sensory activity depending on their specific anatomical localisation (cortical areas, layers, and modules) and intrinsic cellular properties. It is therefore important to assess the precise function of each specific subtype of interneurons. However, in these experiments, it is not always clear whether the alterations are caused by reduced GABA, PV, CR, and CB immunoreactivity/expression in intact interneurons or by an overall reduction of the GABAergic interneuron population. Furthermore, it is still not clear so far about how the neurochemical identity of a specific interneuron relates to its function.

3.2. GABAergic Networks in Experience-Dependent Plasticity and Critical Period Formation. Several recent studies in the visual system suggest that an adequate development and function of GABAergic interneurons in the V1 are critical for controlling the onset and time course of critical periods and for the establishment of the cortical circuit architecture that is necessary for the occurrence of ocular dominance (OD) plasticity [128, 150–153]. This was demonstrated for the first time by Hensch et al. where knockout mice, lacking Glutamic Acid Decarboxylase 65 (GAD65, the synaptic isoform of GABA-producing enzyme), showed no closure of the critical period and thus no occurrence of OD plasticity. Nonetheless, this shortage was rescued by cortical infusion of the GABA agonist diazepam (DZ) [126]. Conversely, augmenting inhibitory signaling prematurely launches the critical period making the mice insensitive to monocular deprivation, as is normally the case in the adult mouse [150, 154, 155]. GABA transmission mediated by the $\alpha 1$ subunit containing GABA-A receptors has been shown to be mandatory for the induction of the critical period for OD plasticity [155, 156]. In parallel, the precocious development of inhibitory circuitry via exposure to brain derived neurotrophic factor (BDNF) in rodents accelerates the maturation of PV interneurons and triggers early onset of the critical period for visual plasticity [153, 157, 158]. Using kittens and the monocular eye lid suture paradigm, Stryker and collaborators have shown that muscimol-induced blockade in V1 causes inversion of OD plasticity, resulting in a consistent shift in the responsiveness of this cortex in favour of the less-active closed eye [159, 160]. More recently, in similar conditions but using normal sighted cats it was found that infusion of DZ on top of V1 resulted in a widening of ocular dominance column spacing, while reducing inhibition shortened the distance between columns [161]. However, once inhibition is mature, it can restrict cortical plasticity. In the adult visual system of the rat, ocular dominance plasticity is greatly reduced but can be restored to juvenile levels by

suppressing inhibition with the antidepressant fluoxetine [162]. Indeed, directly attenuating GABA release by a GAD inhibitor reinstates OD plasticity in adult rats [163]. This is also true for environmental enrichment in old age where diazepam infusion averts the reduction in OD plasticity [164].

More recently, it has been shown by Maffei and collaborators that, in layer IV of binocular V1 in rats, depression of inhibitory synapses on pyramidal neurons is induced when these animals are monocularly deprived for 2 days at the end of the third postnatal week (i.e., before the critical period), whereas potentiation is induced if the monocular deprivation is started in the fourth postnatal week (i.e., within the critical period). During development, these two forms of plasticity shift the balance between circuit excitation and inhibition while the excitatory synaptic drive remains unaffected. Thus, inhibitory plasticity seems to be fundamental in modulating cortical circuit refinement and might be one of the key mechanisms promoting ocular dominance shifts [165]. Such dynamic adjustment of the excitation-inhibition balance may allow the networks to maintain stable levels of activity in the face of variable sensory input. More electrophysiological evidence from this group [148, 166–169] and others [170] suggests that inhibition mediated specifically by fast-spiking (FS) basket-like (PV positive) and regular spiking nonpyramidal (RSNP) (CB or SS positive) interneurons is critically involved in plasticity following deprivation or deafferentation-induced degradation of visual function in V1 of rodents. These effects may however depend upon the age and nature of the visual deprivation and might be differentially regulated across specific cortical layers in the primary visual cortex. Taken as a whole, these studies suggest that maturation of specific subclasses of GABA interneurons is crucial in initiating critical period plasticity, shaping thalamo-cortical afferents, and modulating experience-dependent plasticity in the visual cortex.

3.3. Maturation of Inhibitory Networks and Dependence on Sensory Experience. Although great progress has been made towards understanding both the process of postnatal maturation of excitatory networks and the mechanisms underlying activity-dependent plasticity of excitatory synapses in principal neurons, an understanding of the maturation of inhibitory (GABAergic) circuits has emerged only recently. While the first steps of the development and migration of GABAergic interneurons are likely coordinated by genetic programs, the maturation of these neurons and their synapses are strongly modulated by afferent neuronal activity and experience in both visual and somatosensory cortices. For example, monocular enucleation or dark rearing exposures in rats have been shown to decrease the number of cells and terminals containing GABA and glutamic acid decarboxylase (GAD) [130, 171]. Similar groundbreaking results were also found in the somatosensory cortex, where the unilateral resection of whiskers one day after birth induces a 50% decrease in GABAergic neurons and synapses in layer IV, in the deprived barrels [172]. Conversely and even

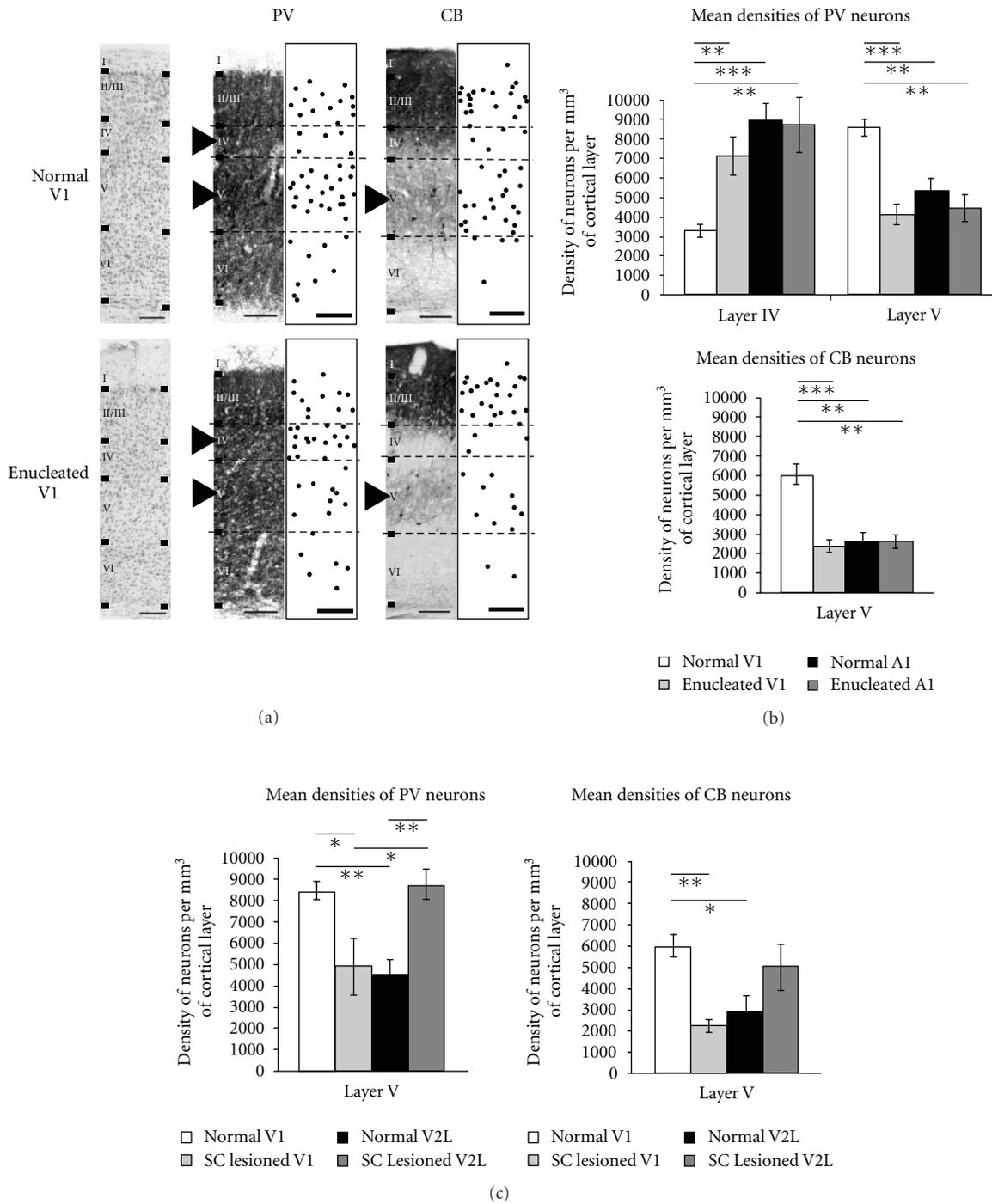


FIGURE 6: Laminar distribution of PV and CB expressing interneurons in the visual and auditory cortices of normal, enucleated and SC lesioned at birth hamsters. (a) Immunostaining patterns of the two CBP-IR neuronal subpopulations in V1 of normal and enucleated hamster. Left panels: cresyl violet staining with laminar boundaries; middle panels: photomicrographs of the distribution of PV and CB immunoreactivities; right panels: distribution of each CBP-IR neurons (black dots) plotted from three superimposed sampled sections. Black arrows indicate layer IV and V changes for PV-IR and CB-IR neurons between experimental groups. Pial surface of the cortices are at the top. Scale bars 100 μ m. (b) Changes in the distributions of PV and CB neurons in layer IV and V of V1 compared to A1 in normal versus enucleated hamsters. (c) Alterations in the distribution of PV and CB interneurons in V1 and V2L of SC lesioned versus normal hamsters. Histograms illustrate the mean density number of neurons per mm³ of cortical layer and error bars represent SEM. Significant differences are represented by stars * $P < 0.05$, ** $P < 0.01$ and *** $P < 0.001$. Adapted from Desgent et al. [29, 30].

in the adult brain, mice that undergo excessive stimulation of a single whisker for 24 hours have an increase of inhibitory synapses on dendrites of principal cells in the corresponding barrel [173]. In adult primates, eliminating retinal activity by an intraocular administration of Tetrodotoxin (TTX) reduces the immunoreactivity for GABA, GAD, and the GABA-A receptor in neurons in the areas corresponding to the injected eye in V1 [146, 174–176].

More recently, Morales et al. have shown that between the time at which the eyes first open and the end of the critical period for experience-dependent plasticity, the total GABAergic input converging onto pyramidal cells increases threefold in rats. A developmental increase in GABAergic input can be prevented in animals deprived of light since birth, but not in animals deprived of light after a period of normal experience. Thus, sensory experience appears to play a permissive role in the maturation of intracortical GABAergic circuits [177]. In the past decade genetic strategies based on interneuron cell type specific promoters and fluorescent protein reporters have allowed more efficient high-resolution labelling of specific GABAergic interneurons and associated morphology. By using these approaches direct experimental evidence was found linking structural and functional changes of specific inhibitory networks with their sensory experiences *in vivo* and *in vitro*. A significant study in this context from Chattopadhyaya et al. showed that sensory input deprivation using intraocular injections of TTX in mice, and in postnatal organotypic cultures, causes a reduction in the density of perisomatic synapses formed by basket GABAergic neurons in the visual cortex. This sensitivity was restricted to a critical time window during the third postnatal week in mice which correlates with the time course of the critical period for ocular dominance plasticity [178]. These results are consistent with studies done in the barrel cortex in mice that underwent whisker removal from the left mystacial pad at neonatal day 7, until day 15 [134, 138, 179]. These experiments using Glutamate Acid Decarboxylase 67 (GAD67)-Green fluorescent protein (GFP) (Δ neo) and wild-type mice showed specific structural anatomical changes, illustrated by a reduction in the number of presynaptic perisomatic inhibitory boutons, specifically from PV interneurons. These changes were associated again with a lack of sensory experience during the second and the third postnatal week. However, the total number of GFP-GAD67 cells (i.e., total number of GABAergic cells) remained unchanged indicating that these changes in PV expression from basket cells appeared to be the major effect of sensory deprivation. Moreover, these modifications were associated with a reduction in the amplitude of evoked intracortical inhibitory synaptic potentials in patch-clamp recordings in deprived versus spared cortices. These results indicate that perisomatic inhibition mediated by PV-positive basket cells was pruned by sensory deprivation. More recently, Jiao and collaborators, using a line of mutant mice that lack activity-dependent BDNF expression (*bdnf-KIV*), have shown that experience regulates the cortical GABAergic network via activity-driven BDNF expression of principal neurons [138]. Levels of endogenous BDNF protein in the barrel cortex are strongly regulated by sensory inputs from the whiskers.

Moreover, the mutant barrel cortex exhibits significantly reduced levels of GABA release only from the PV-expressing fast-spiking (FS) interneurons. Postnatal deprivation of sensory inputs markedly decreases perisomatic inhibition selectively from FS cells in wild-type but not *bdnf-KIV* mice. These results suggest that postnatal experience, through sensory-driven BDNF expression, controls cortical development by regulating FS cell-mediated perisomatic inhibition *in vivo*. This further highlights that PV (FS) networks can selectively be inhibited by sensory deprivation from the thalamo-cortical afferent pathway.

Together, these results suggest that the properties of local cortical inhibitory network are modified by sensory experience. Thus, postnatal sensory activity is necessary for transformation of immature inhibitory transmission to a mature functional phenotype. Nevertheless, precisely how activity and molecular-driven mechanisms work together to accomplish the remarkable specificity of GABAergic synapse maturation, localization, and formation is not fully understood but is emerging. Several molecular factors have also been implicated in the process such as BDNF, GABA itself, Otx2 homeoprotein, molecular components of the extracellular matrix, and cell adhesion molecules (e.g., chondroitin sulfate proteoglycans (CSPGs), polysialic acid (PSA), and the neural cell adhesion molecule (NCAM)). For example, in mouse visual cortex, PSA is downregulated following eye opening and this decrease has been shown to allow the maturation of GABAergic synapses and the opening, of the critical period for ocular dominance plasticity [180]. For more exhaustive reviews on this topic please refer to the following papers [22, 181–185].

4. Alterations of GABAergic Interneurons in Animal Models of Cross-Modal Plasticity

Very few studies have looked at the possible role of GABAergic interneurons in cross-modal plasticity. Alterations in inhibitory circuits were observed qualitatively for the first time in deaf and rewired cross-modal ferrets and concerned modifications in the morphology and proportion of interneurons containing PV and CB. Specifically, CB neurons in A1 of these animals showed an atypical and extended dendritic arborisation in the horizontal axis. However these changes were never studied further with quantitative validation [1, 105]. Interestingly, a recent study done in FVB (*GAD-GFP*) mice has shown that olfactory deprivation occurring at P12 can lower the number of GABAergic interneurons in the piriform cortex and at the same time increase their number in the barrel cortex, ipsilateral to the lesion, upregulating whisker tactile sensation [28]. This suggests that these neurons are important for cortical sensory compensation and substitution. Recent work carried out in our laboratory, on hamsters enucleated at birth (EH), follows the idea that observed cross-modal plasticity changes may be due to modifications in GABAergic interneurons that express calcium-binding proteins (CBPs) like PV and CB [29, 30]. Since the laminar distribution of these proteins is significantly different in the primary visual and auditory

cortices of normal hamsters [186], the induction of aberrant connectivity to these cortices should also be evident at the neurochemical level. Indeed, hamsters enucleated at birth show significant changes in the distribution of CBPs only in their primary visual cortex. Compared to intact hamsters, the density of PV-immunoreactive neurons is higher in layer IV and lower in layer V, whereas the density of CB-immunoreactive cells is significantly lower in layer V of V1 in the enucleated animals (see Figures 6(a) and 6(b)). These results suggest that the affected primary visual cortex may adopt the GABAergic chemical features of the auditory cortex through cross-modal rewiring.

Several possibilities exist. As described earlier, sensory deprivation generally reduces the expression of PV in primary sensory cortices. Normally, visual activity is an essential requirement in preventing a robust downregulation of PV expression, mainly in cortical layer IV. The functional implications of this decrease, following sensory deprivation in several animal models, have been associated with a reduced inhibition following loss of sight. One can therefore expect a general decrease in PV expression in the V1 of enucleated hamsters, with possible stronger effects in the thalamorecipient cortical layer IV. In CRX^{-/-} mutant mice, there is a significant decrease of PV-IR cells in all layers of the primary visual cortex [149]. This contrasts with the increase in the number of PV-IR cells in layer IV and the decrease observed in layer V of enucleated hamsters. The decrease in PV-IR in mutant mice suggests that parvalbumin expression requires visual activity in V1. There is clearly no visual activity during postnatal development in enucleated hamsters whereas in CRX^{-/-} mice, even with photoreceptors lacking outer segments, there remain waves of spontaneous retinal ganglion cell activity transmitted to the thalamus and cortex before P14. It could therefore be expected that, in enucleated hamsters, one would find an even greater decrease of PV-IR cells in all cortical layers of the V1 as with these mice, but except for layer V this was not the case.

Significant reduction of expression of both PV and CB in interneurons of layer V of V1 in enucleated hamsters may imply changes in an alternate pathway for cortico-cortical communication between the primary visual cortex and neighbouring-associated areas. Guillery and Sherman proposed that the driving of cortico-cortical projections is mediated by layer V pyramidal neurons that project to the pulvinar of the thalamus (or lateral posterior nucleus (LP) in rodents), which in turn provides the output to higher cortical areas (i.e., the feedforward corticothalamocortical pathway) [187, 188]. Dysfunction of inhibitory interneurons of layer V could play a pivotal role in gating this alternative process and corticocortical communications [26]. It is however unknown as to whether the observed changes in PV and CB expression in interneurons within layer V of EH are directly involved in cortico-cortical networks. In fact, changes in the number of PV-IR and CB-IR neurons in enucleated hamsters might reflect laminar-specific increase and/or decrease of the synaptic drive on these particular neurons in layer IV and V of V1 after enucleation at birth.

Changes in immunoreactive interneurons might reflect two phenomena: (1) a true change in the number/density

of PV or CB immunoreactive neurons (e.g., via apoptosis/neurogenesis, impaired migration, suppressed cell proliferation, etc.) or (2) altered immunocytochemical detection levels of the proteins PV or CB, respectively, whose cellular expression might be positively correlated to physiological activity levels [134, 138, 143, 145, 189–191]. Because we did not find any differences in the total population densities for these two proteins, we favour the interpretation that PV and CB expression (changes in synthesis or degradation) in layer IV and V of V1 is altered in EH and that the altered protein levels may be related to the activity of these inhibitory interneurons. However, due to limited knowledge of the physiological function of CBP proteins, the interpretation of the physiological consequences of this early enucleation is complicated. Furthermore, the causal relationship between our anatomical findings and the putative role of these cells in cross-modal plasticity in this animal model remains exploratory.

The observed changes in EH could be explained not only by the absence of postnatal visual input to the V1 but also by the presence of auditory information reaching V1 from new ectopic projections arising from the IC to the dLGN of the thalamus in this rewired model [47]. Another alternative involves cortico-cortical projections originating from the auditory cortex [60]. We hypothesize that these specific changes in the laminar distribution of mostly PV-IR but also CB-IR neurons in V1 could be responsible for shaping auditory response properties of V1 neurons previously observed in enucleated hamsters. The new auditory thalamic afferents into the visual system of the enucleated hamsters could explain the auditory cortex-like distribution pattern of PV-IR neurons in the primary visual cortex. Noteworthy is recent work by Sugiyama et al. which led to the discovery of a novel mechanism explaining how visual input is tied to the onset of ocular dominance plasticity in the visual cortex. This group has shown that a retinal-derived homeoprotein, Otx2, can be directly transferred into V1 through a visual-experience-dependent mechanism. Once Otx 2 has reached the visual cortex, it can nurture specific types of GABAergic interneurons (viz. PV neurons) and modulate critical period plasticity [22, 151, 185, 192, 193]. The study of target genes and proteins of Otx2 could reveal further insights into the machinery linking sensory experience, GABAergic circuit maturation, and plasticity. It is that, as yet unknown homologues of Otx2 might be delivered from other sensory receptor arrays and pathways to precise cortical areas, to promote local inhibitory circuit maturation depending on modality type. In hamsters enucleated at birth, for example, such a molecular factor coming from the auditory system but “redirected” to the primary visual cortex by the cochlea-IC-dLGN pathway could lead to modality-specific changes observed in circuits in absence of visually driven Otx2 but in presence of an auditory homologue. This might explain the altered auditory-like distribution of PV-IR neurons we observed therein [29, 30]. However, the absence of spontaneous electrical activity in the retinal afferents to the lateral geniculate nucleus and the lack of trophic influences of the retina on neurons in the dLGN and from there to area V1 could also be involved in the changes observed in our animal

model. It may also be that other sensory modalities, such as somatosensory inputs, could induce the same changes. Injections of HRP into the dorsal column nuclei of adult mice enucleated at birth have shown that ascending somatic sensory axons can be rerouted to the lateral geniculate nucleus [55]. Even if this type of connection has not yet been reported in early enucleated hamsters [47], any combination of these factors, in addition to abnormal auditory rewired inputs to V1 in these animals, could account for the present modifications in the anatomy and laminar circuitry of the V1.

5. Future Directions and Conclusion

In conclusion, several animal studies have revealed important properties of inhibitory network alterations in the neocortex following the early loss of a given sensory function. We are just beginning to acquire the necessary knowledge on response properties of GABAergic neurons, their maturation mechanisms, and how they influence sensory and cross-modal plasticity. Hence, we have uncovered only the tip of a very large iceberg in that context. Visual and other sensory cortical circuits are organized at multiple levels of complexity including cortical areas, layers and columns, and specific cell types within these modules. Making sense of the functions of these circuits, from an anatomical point of view, requires linking these circuits to function at each of these levels of complexity. Functional studies on cross-modal plasticity in animals have previously been limited to pharmacological approaches, electrophysiology, tracing or lesion researches that provide poor cell-type specificity and sometimes low-spatial or temporal resolutions. Nowadays, advancements in molecular techniques have made it possible to address questions that were unapproachable just a decade ago. As new methods for single cell two-photon imaging, voltage sensitive dyes for cortical optical imaging, transneuronal viral tracing, *in vivo* MRI spectroscopy (MRS), laser microdissection, transfection and genetic targeting of specific subpopulations of inhibitory interneurons are perfected, we will certainly one day be able to highlight and incorporate better the roles of subtype of GABAergic neurons into multifaceted animal models of cross-modal plasticity. More specifically, one interesting approach concerning this issue would be to use optogenetic in transgenic mouse strains for activation or inhibition of specific GABAergic subpopulations using a viral-mediated transfer of a Cre/loxP transgene controlling the expression of light-activated ion channels in these cells *in vivo* or *in vitro* (e.g., channelrhodopsin ChR and halorhodopsins eNpHR). For example, these would allow us to dissect the function of different neuronal class during awakening behavior or electrophysiology, sensory stimulation, and discrimination tasks with a millisecond resolution. Experiments using these techniques will help us to understand more clearly some important questions in the field: how specific GABAergic subpopulations participate in the rewiring/cross-wiring processes between cortical modalities, what would be the effects of shutting down one population type on cross-modal integration, how these

interneurons could integrate inputs from spared modalities, what are the possible multimodal versus unimodal receptive field properties in these cells in normal and sensory deprived cortices, what are the effects on neuronal plasticity as well as neighbouring neuronal networks, are there modality-specific biomarkers that could travel from the sensory receptor periphery to these cortical neurons via a modality experience-dependent-mechanism, and what are the cortical network dynamics between excitatory and inhibitory synapses *in vivo* following the early-life lost of sight? It is going to be a long ride, and experiments to tackle these issues will be very challenging technically. Nevertheless, the clarification of these underlying mechanisms may one day provide clues to develop new therapeutic advances aimed to increase adaptive circuit rewiring following insult to help sensory substitution and recovery.

Acknowledgments

Dr. Desgent is a former grantee from the Canadian Institutes of Health Research (CIHR)/Canadian National Institute for the Blind (CNIB) now enrolled in a postdoctoral fellow in the laboratory of Dr. Lionel Carmant at the CHU Sainte-Justine biomedical research center. His present work is supported by three national postdoctoral awards from Saint-Justine Hospital Foundation/Foundation of Stars, Epilepsy Canada and Savoy Foundation. The research was supported over the years by the Natural Sciences and Engineering Research Council of Canada (NSERC) to MP. The authors are grateful to Dr. Derek Robertson and Dr. Bidisha Chattopadhyaya for reading and editing the present paper.

References

- [1] S. L. Pallas, "Intrinsic and extrinsic factors that shape neocortical specification," *Trends in Neurosciences*, vol. 24, no. 7, pp. 417–423, 2001.
- [2] A. K. Majewska and M. Sur, "Plasticity and specificity of cortical processing networks," *Trends in Neurosciences*, vol. 29, no. 6, pp. 323–329, 2006.
- [3] D. D. O'Leary, S. J. Chou, and S. Sahara, "Area patterning of the mammalian cortex," *Neuron*, vol. 56, no. 2, pp. 252–269, 2007.
- [4] M. Ptito and S. Desgent, "Sensory input-based adaptation and brain architecture," in *Lifespan Development and the Brain. The Perspective of Biocultural Co-constructivism*, P. Baltes, P. Reuter-Lorenz, and F. Rosler, Eds., pp. 111–134, Cambridge University Press, Cambridge, UK, 2006.
- [5] L. Krubitzer, "The magnificent compromise: cortical field evolution in mammals," *Neuron*, vol. 56, no. 2, pp. 201–208, 2007.
- [6] L. B. Merabet and A. Pascual-Leone, "Neural reorganization following sensory loss: the opportunity of change," *Nature Reviews Neuroscience*, vol. 11, no. 1, pp. 44–52, 2010.
- [7] E. Ricciardi and P. Pietrini, "New light from the dark: what blindness can teach us about brain function," *Current Opinion in Neurology*, vol. 24, no. 4, pp. 357–363, 2011.
- [8] R. Kupers and M. Ptito, "Insights from darkness: what the study of blindness has taught us about brain structure and

- function,” *Progress in Brain Research*, vol. 192, pp. 17–31, 2011.
- [9] M. Ptito and R. Kupers, “Cross-modal plasticity in early blindness,” *Journal of Integrative Neuroscience*, vol. 4, no. 4, pp. 479–488, 2005.
- [10] R. Kupers, P. Pietrini, E. Ricciardi, and M. Ptito, “The nature of consciousness in the visually deprived brain,” *Frontiers in Psychology*, vol. 2, article 19, 2011.
- [11] U. Noppeney, “The effects of visual deprivation on functional and structural organization of the human brain,” *Neuroscience and Biobehavioral Reviews*, vol. 31, no. 8, pp. 1169–1180, 2007.
- [12] R. Kupers, D. R. Chebat, K. H. Madsen, O. B. Paulson, and M. Ptito, “Neural correlates of virtual route recognition in congenital blindness,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 107, no. 28, pp. 12716–12721, 2010.
- [13] A. Bubic, E. Striem-Amit, and A. Amedi, “Large-scale brain plasticity following blindness and the use of sensory substitution devices,” in *Multisensory Object Perception in the Primate Brain*, M. J. Naumer and J. Kaiser, Eds., pp. 351–380, Springer, 2010.
- [14] I. Matteau, R. Kupers, E. Ricciardi, P. Pietrini, and M. Ptito, “Beyond visual, aural and haptic movement perception: hMT+ is activated by electrotactile motion stimulation of the tongue in sighted and in congenitally blind individuals,” *Brain Research Bulletin*, vol. 82, no. 5–6, pp. 264–270, 2010.
- [15] O. Collignon, G. Vandewalle, P. Voss et al., “Functional specialization for auditory-spatial processing in the occipital cortex of congenitally blind humans,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 11, pp. 4435–4440, 2011.
- [16] R. Kupers, M. Beaulieu-Lefebvre, F. C. Schneider et al., “Neural correlates of olfactory processing in congenital blindness,” *Neuropsychologia*, vol. 49, no. 7, pp. 2037–2044, 2011.
- [17] D. Bavelier and H. J. Neville, “Cross-modal plasticity: where and how?” *Nature Reviews Neuroscience*, vol. 3, no. 6, pp. 443–452, 2002.
- [18] J. Driver and T. Noesselt, “Multisensory interplay reveals crossmodal influences on “sensory-specific” brain regions, neural responses, and judgments,” *Neuron*, vol. 57, no. 1, pp. 11–23, 2008.
- [19] R. C. Kadosh and V. Walsh, “Cognitive neuroscience: rewired or crosswired brains?” *Current Biology*, vol. 16, no. 22, pp. R962–R963, 2006.
- [20] Q. Q. Sun, J. R. Huguenard, and D. A. Prince, “Barrel cortex microcircuits: thalamocortical feedforward inhibition in spiny stellate cells is mediated by a small number of fast-spiking interneurons,” *Journal of Neuroscience*, vol. 26, no. 4, pp. 1219–1230, 2006.
- [21] H. A. Swadlow, “Fast-spike interneurons and feedforward inhibition in awake sensory neocortex,” *Cerebral Cortex*, vol. 13, no. 1, pp. 25–32, 2003.
- [22] S. Sugiyama, A. A. Di Nardo, S. Aizawa et al., “Experience-dependent transfer of Otx2 homeoprotein into the visual cortex activates postnatal plasticity,” *Cell*, vol. 134, no. 3, pp. 508–520, 2008.
- [23] Y. Gonchar and A. Burkhalter, “Differential subcellular localization of forward and feedback interareal inputs to parvalbumin expressing GABAergic neurons in rat visual cortex,” *Journal of Comparative Neurology*, vol. 406, no. 3, pp. 346–360, 1999.
- [24] R. R. Johnson and A. Burkhalter, “Microcircuitry of forward and feedback connections within rat visual cortex,” *Journal of Comparative Neurology*, vol. 368, no. 3, pp. 383–398, 1996.
- [25] A. Yamashita, K. Valkova, Y. Gonchar, and A. Burkhalter, “Rearrangement of synaptic connections with inhibitory neurons in developing mouse visual cortex,” *Journal of Comparative Neurology*, vol. 464, no. 4, pp. 426–437, 2003.
- [26] E. M. Callaway, “Feedforward, feedback and inhibitory connections in primate visual cortex,” *Neural Networks*, vol. 17, no. 5–6, pp. 625–632, 2004.
- [27] S. L. Pallas, P. Wenner, C. Gonzalez-Islas et al., “Developmental plasticity of inhibitory circuitry,” *Journal of Neuroscience*, vol. 26, no. 41, pp. 10358–10361, 2006.
- [28] H. Ni, L. Huang, N. Chen et al., “Upregulation of barrel GABAergic neurons is associated with cross-modal plasticity in olfactory deficit,” *PLoS ONE*, vol. 5, no. 10, Article ID e13736, 2010.
- [29] S. Desgent, D. Boire, and M. Ptito, “Altered expression of parvalbumin and calbindin in interneurons within the primary visual cortex of neonatal enucleated hamsters,” *Neuroscience*, vol. 171, no. 4, pp. 1326–1340, 2010.
- [30] S. Desgent, D. Boire, and M. Ptito, *Changes in Distribution of Calcium-Binding Proteins in the Visual and Auditory Cortices Following Neonatal Alterations of the Visual System in the Hamster*, vol. 4, abstract no. 054.13, Federation of European Neuroscience Societies (FENS), Geneva, Switzerland, 2008.
- [31] S. J. Karlen, D. M. Kahn, and L. Krubitzer, “Early blindness results in abnormal corticocortical and thalamocortical connections,” *Neuroscience*, vol. 142, no. 3, pp. 843–858, 2006.
- [32] D. D. Larsen, J. D. Luu, M. E. Burns, and L. Krubitzer, “What are the effects of severe visual impairment on the cortical organization and connectivity of primary visual cortex?” *Frontiers in Neuroanatomy*, vol. 3, article 30, 2009.
- [33] G. Rebillard, E. Carlier, M. Rebillard, and R. Pujol, “Enhancement of visual responses on the primary auditory cortex of the cat after an early destruction of cochlear receptors,” *Brain Research*, vol. 129, no. 1, pp. 162–164, 1977.
- [34] D. L. Hunt, E. N. Yamoah, and L. Krubitzer, “Multisensory plasticity in congenitally deaf mice: how are cortical areas functionally specified?” *Neuroscience*, vol. 139, no. 4, pp. 1507–1524, 2006.
- [35] H. Jiang, F. Lepore, M. Ptito, and J. P. Guillemot, “Sensory modality distribution in the anterior ectosylvian cortex (AEC) of cats,” *Experimental Brain Research*, vol. 97, no. 3, pp. 404–414, 1994.
- [36] J. P. Rauschecker and M. Korte, “Auditory compensation for early blindness in cat cerebral cortex,” *Journal of Neuroscience*, vol. 13, no. 10, pp. 4538–4548, 1993.
- [37] J. P. Rauschecker, “Compensatory plasticity and sensory substitution in the cerebral cortex,” *Trends in Neurosciences*, vol. 18, no. 1, pp. 36–43, 1995.
- [38] J. P. Rauschecker, “Cortical map plasticity in animals and humans,” *Progress in Brain Research*, vol. 138, pp. 73–88, 2002.
- [39] J. Toldi, T. Farkas, and B. Volgyi, “Neonatal enucleation induces cross-modal changes in the barrel cortex of rat. A behavioural and electrophysiological study,” *Neuroscience Letters*, vol. 167, no. 1–2, pp. 1–4, 1994.
- [40] J. Toldi, O. Feher, and J. R. Wolff, “Neuronal plasticity induced by neonatal monocular (and binocular) enucleation,” *Progress in Neurobiology*, vol. 48, no. 3, pp. 191–218, 1996.
- [41] G. Bronchti, N. Schonenberger, E. Welker, and H. Van der Loos, “Barrelfield expansion after neonatal eye removal in mice,” *NeuroReport*, vol. 3, no. 6, pp. 489–492, 1992.

- [42] J. R. Newton, R. W. Sikes, and A. A. Skavenski, "Cross-modal plasticity after monocular enucleation of the adult rabbit," *Experimental Brain Research*, vol. 144, no. 4, pp. 423–429, 2002.
- [43] J. P. Rauschecker, B. Tian, M. Korte, and U. Egert, "Cross-modal changes in the somatosensory vibrissa/barrel system of visually deprived animals," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 89, no. 11, pp. 5063–5067, 1992.
- [44] R. Yaka, U. Yinon, M. Rosner, and Z. Wollberg, "Pathological and experimentally induced blindness induces auditory activity in the cat primary visual cortex," *Experimental Brain Research*, vol. 131, no. 1, pp. 144–148, 2000.
- [45] R. Yaka, U. Yinon, and Z. Wollberg, "Auditory activation of cortical visual areas in cats after early visual deprivation," *European Journal of Neuroscience*, vol. 11, no. 4, pp. 1301–1312, 1999.
- [46] N. Chabot, S. Robert, R. Tremblay, D. Miceli, D. Boire, and G. Bronchti, "Audition differently activates the visual system in neonatally enucleated mice compared with anophthalmic mutants," *European Journal of Neuroscience*, vol. 26, no. 8, pp. 2334–2348, 2007.
- [47] R. Izraeli, G. Koay, M. Lamish et al., "Cross-modal neuroplasticity in neonatally enucleated hamsters: structure, electrophysiology and behaviour," *European Journal of Neuroscience*, vol. 15, no. 4, pp. 693–712, 2002.
- [48] D. M. Kahn and L. Krubitzer, "Massive cross-modal cortical plasticity and the emergence of a new cortical area in developmentally blind mammals," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 99, no. 17, pp. 11429–11434, 2002.
- [49] M. Piche, N. Chabot, G. Bronchti, D. Miceli, F. Lepore, and J. P. Guillemot, "Auditory responses in the visual cortex of neonatally enucleated rats," *Neuroscience*, vol. 145, no. 3, pp. 1144–1156, 2007.
- [50] N. Doron and Z. Wollberg, "Cross-modal neuroplasticity in the blind mole rat *Spalax ehrenbergi*: a WGA-HRP tracing study," *NeuroReport*, vol. 5, no. 18, pp. 2697–2701, 1994.
- [51] G. Bronchti, P. Heil, R. Sadka, A. Hess, H. Scheich, and Z. Wollberg, "Auditory activation of "visual" cortical areas in the blind mole rat (*Spalax ehrenbergi*)," *European Journal of Neuroscience*, vol. 16, no. 2, pp. 311–329, 2002.
- [52] G. Bronchti, P. Heil, H. Scheich, and Z. Wollberg, "Auditory pathway and auditory activation of primary visual targets in the blind mole rat (*Spalax ehrenbergi*): I. 2-deoxyglucose study of subcortical centers," *Journal of Comparative Neurology*, vol. 284, no. 2, pp. 253–274, 1989.
- [53] P. Heil, G. Bronchti, Z. Wollberg, and H. Scheich, "Invasion of visual cortex by the auditory system in the naturally blind mole rat," *NeuroReport*, vol. 2, no. 12, pp. 735–738, 1991.
- [54] I. R. Kaiserman-Abramof, A. M. Graybiel, and W. J. Nauta, "The thalamic projection to cortical area 17 in a congenitally anophthalmic mouse strain," *Neuroscience*, vol. 5, no. 1, pp. 41–52, 1980.
- [55] C. Asanuma and B. B. Stanfield, "Induction of somatic sensory inputs to the lateral geniculate nucleus in congenitally blind mice and in phenotypically normal mice," *Neuroscience*, vol. 39, no. 3, pp. 533–545, 1990.
- [56] N. Chabot, V. Charbonneau, M. E. Laramée, R. Tremblay, D. Boire, and G. Bronchti, "Subcortical auditory input to the primary visual cortex in anophthalmic mice," *Neuroscience Letters*, vol. 433, no. 2, pp. 129–134, 2008.
- [57] M. Piche, S. Robert, D. Miceli, and G. Bronchti, "Environmental enrichment enhances auditory takeover of the occipital cortex in anophthalmic mice," *European Journal of Neuroscience*, vol. 20, no. 12, pp. 3463–3472, 2004.
- [58] D. M. Kahn and L. Krubitzer, "Retinofugal projections in the short-tailed opossum (*Monodelphis domestica*)," *Journal of Comparative Neurology*, vol. 447, no. 2, pp. 114–127, 2002.
- [59] S. J. Karlen and L. Krubitzer, "Effects of bilateral enucleation on the size of visual and nonvisual areas of the brain," *Cerebral Cortex*, vol. 19, no. 6, pp. 1360–1371, 2009.
- [60] M. A. Kingsbury, N. A. Lettman, and B. L. Finlay, "Reduction of early thalamic input alters adult corticocortical connectivity," *Developmental Brain Research*, vol. 138, no. 1, pp. 35–43, 2002.
- [61] L. Negyessy, V. Gál, T. Farkas, and J. Toldi, "Cross-modal plasticity of the corticothalamic circuits in rats enucleated on the first postnatal day," *European Journal of Neuroscience*, vol. 12, no. 5, pp. 1654–1668, 2000.
- [62] K. S. Rockland and H. Ojima, "Multisensory convergence in calcarine visual areas in macaque monkey," *International Journal of Psychophysiology*, vol. 50, no. 1-2, pp. 19–26, 2003.
- [63] G. M. Innocenti, P. Berbel, and S. Clarke, "Development of projections from auditory to visual areas in the cat," *Journal of Comparative Neurology*, vol. 272, no. 2, pp. 242–259, 1988.
- [64] S. Clavagnier, A. Falchier, and H. Kennedy, "Long-distance feedback projections to area V1: implications for multisensory integration, spatial awareness, and visual consciousness," *Cognitive, Affective and Behavioral Neuroscience*, vol. 4, no. 2, pp. 117–126, 2004.
- [65] A. Falchier, S. Clavagnier, P. Barone, and H. Kennedy, "Anatomical evidence of multimodal integration in primate striate cortex," *Journal of Neuroscience*, vol. 22, no. 13, pp. 5749–5759, 2002.
- [66] A. J. Hall and S. G. Lomber, "Auditory cortex projections target the peripheral field representation of primary visual cortex," *Experimental Brain Research*, vol. 190, no. 4, pp. 413–430, 2008.
- [67] E. Vaudano, C. R. Legg, and M. Glickstein, "Afferent and efferent connections of temporal association cortex in the rat: a horseradish peroxidase study," *European Journal of Neuroscience*, vol. 3, no. 4, pp. 317–330, 1991.
- [68] E. Budinger, P. Heil, A. Hess, and H. Scheich, "Multisensory processing via early cortical stages: connections of the primary auditory cortical field with other sensory systems," *Neuroscience*, vol. 143, no. 4, pp. 1065–1083, 2006.
- [69] J. K. Bizley, F. R. Nodal, V. M. Bajo, I. Nelken, and A. J. King, "Physiological and anatomical evidence for multisensory interactions in auditory cortex," *Cerebral Cortex*, vol. 17, no. 9, pp. 2172–2189, 2007.
- [70] M. E. Laramée, T. Kurotani, K. S. Rockland, G. Bronchti, and D. Boire, "Indirect pathway between the primary auditory and visual cortices through layer V pyramidal neurons in V2L in mouse and the effects of bilateral enucleation," *European Journal of Neuroscience*, vol. 34, no. 1, pp. 65–78, 2011.
- [71] G. E. Schneider, "Early lesions of superior colliculus: factors affecting the formation of abnormal retinal projections," *Brain, Behavior and Evolution*, vol. 8, no. 1-2, pp. 73–109, 1973.
- [72] B. J. Crain and W. C. Hall, "The normal organization of the lateral posterior nucleus in the golden hamster and its reorganization after neonatal superior colliculus lesions," *Behavioural Brain Research*, vol. 3, no. 2, pp. 223–238, 1981.

- [73] C. Ling, S. Jhaveri, and G. E. Schneider, "Target- as well as source-derived factors direct the morphogenesis of anomalous retino-thalamic projections," *Journal of Comparative Neurology*, vol. 388, no. 3, pp. 454–466, 1997.
- [74] M. Sur, P. E. Garraghty, and A. W. Roe, "Experimentally induced visual projections into auditory thalamus and cortex," *Science*, vol. 242, no. 4884, pp. 1437–1441, 1988.
- [75] D. O. Frost, "Orderly anomalous retinal projections to the medial geniculate, ventrobasal, and lateral posterior nuclei of the hamster," *Journal of Comparative Neurology*, vol. 203, no. 2, pp. 227–256, 1981.
- [76] D. O. Frost, "Anomalous visual connections to somatosensory and auditory systems following brain lesions in early life," *Brain Research*, vol. 255, no. 4, pp. 627–635, 1982.
- [77] D. O. Frost, "Axonal growth and target selection during development: retinal projections to the ventrobasal complex and other "nonvisual" structures in neonatal syrian hamsters," *Journal of Comparative Neurology*, vol. 230, no. 4, pp. 576–592, 1984.
- [78] D. O. Frost, "Development of anomalous retinal projections to nonvisual thalamic nuclei in syrian hamsters: a quantitative study," *Journal of Comparative Neurology*, vol. 252, no. 1, pp. 95–105, 1986.
- [79] P. G. Bhide and D. O. Frost, "Axon substitution in the reorganization of developing neural connections," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 89, no. 24, pp. 11847–11851, 1992.
- [80] P. G. Bhide and D. O. Frost, "Intrinsic determinants of retinal axon collateralization and arborization patterns," *Journal of Comparative Neurology*, vol. 411, no. 1, pp. 119–129, 1999.
- [81] G. Campbell and D. O. Frost, "Target-controlled differentiation of axon terminals and synaptic organization," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 84, no. 19, pp. 6929–6933, 1987.
- [82] G. Campbell and D. O. Frost, "Synaptic organization of anomalous retinal projections to the somatosensory and auditory thalamus: target-controlled morphogenesis of axon terminals and synaptic glomeruli," *Journal of Comparative Neurology*, vol. 272, no. 3, pp. 383–408, 1988.
- [83] C. Metin and D. O. Frost, "Visual responses of neurons in somatosensory cortex of hamsters with experimentally induced retinal projections to somatosensory thalamus," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 86, no. 1, pp. 357–361, 1989.
- [84] D. O. Frost, "Functional organization of surgically created visual circuits," *Restorative Neurology and Neuroscience*, vol. 15, no. 2-3, pp. 107–113, 1999.
- [85] S. L. Pallas, J. Hahm, and M. Sur, "Morphology of retinal axons induced to arborize in a novel target, the medial geniculate nucleus. I. Comparison with arbors in normal targets," *Journal of Comparative Neurology*, vol. 349, no. 3, pp. 343–362, 1994.
- [86] S. L. Pallas and M. Sur, "Morphology of retinal axon arbors induced to arborize in a novel target, the medial geniculate nucleus. II. Comparison with axons from the inferior colliculus," *Journal of Comparative Neurology*, vol. 349, no. 3, pp. 363–376, 1994.
- [87] A. Angelucci, F. Clascá, E. Bricolo, K. S. Cramer, and M. Sur, "Experimentally induced retinal projections to the ferret auditory thalamus: development of clustered eye-specific patterns in a novel target," *Journal of Neuroscience*, vol. 17, no. 6, pp. 2040–2055, 1997.
- [88] A. W. Roe, P. E. Garraghty, M. Esguerra, and M. Sur, "Experimentally induced visual projections to the auditory thalamus in ferrets: evidence for a W cell pathway," *Journal of Comparative Neurology*, vol. 334, no. 2, pp. 263–280, 1993.
- [89] S. L. Pallas, A. W. Roe, and M. Sur, "Visual projections induced into the auditory pathway of ferrets. I. Novel inputs to primary auditory cortex (AI) from the LP/pulvinar complex and the topography of the MGN-AI projection," *Journal of Comparative Neurology*, vol. 298, no. 1, pp. 50–68, 1990.
- [90] S. L. Pallas and M. Sur, "Visual projections induced into the auditory pathway of ferrets: II. Corticocortical connections of primary auditory cortex," *Journal of Comparative Neurology*, vol. 337, no. 2, pp. 317–333, 1993.
- [91] W. J. Gao and S. L. Pallas, "Cross-modal reorganization of horizontal connectivity in auditory cortex without altering thalamocortical projections," *Journal of Neuroscience*, vol. 19, no. 18, pp. 7940–7950, 1999.
- [92] S. L. Pallas, T. Littman, and D. R. Moore, "Cross-modal reorganization of callosal connectivity without altering thalamocortical projections," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 96, no. 15, pp. 8751–8756, 1999.
- [93] D. O. Frost, "Sensory processing by novel, experimentally induced cross-modal circuits," *Annals of the New York Academy of Sciences*, vol. 608, pp. 92–109, 1990, discussion 109–112.
- [94] D. O. Frost and C. Metin, "Induction of functional retinal projections to the somatosensory system," *Nature*, vol. 317, no. 6033, pp. 162–164, 1985.
- [95] M. Ptito, J. F. Giguère, D. Boire, D. O. Frost, and C. Casanova, "When the auditory cortex turns visual," *Progress in Brain Research*, vol. 134, pp. 447–458, 2001.
- [96] A. W. Roe, S. L. Pallas, J. O. Hahm, and M. Sur, "A map of visual space induced in primary auditory cortex," *Science*, vol. 250, no. 4982, pp. 818–820, 1990.
- [97] A. W. Roe, S. L. Pallas, Y. H. Kwon, and M. Sur, "Visual projections routed to the auditory pathway in ferrets: receptive fields of visual neurons in primary auditory cortex," *Journal of Neuroscience*, vol. 12, no. 9, pp. 3651–3664, 1992.
- [98] J. Sharma, A. Angelucci, and M. Sur, "Induction of visual orientation modules in auditory cortex," *Nature*, vol. 404, no. 6780, pp. 841–847, 2000.
- [99] M. Sur, A. Angelucci, and J. Sharma, "Rewiring cortex: the role of patterned activity in development and plasticity of neocortical circuits," *Journal of Neurobiology*, vol. 41, no. 1, pp. 33–43, 1999.
- [100] M. Sur and C. A. Leamey, "Development and plasticity of cortical areas and networks," *Nature Reviews Neuroscience*, vol. 2, no. 4, pp. 251–262, 2001.
- [101] S. H. Horng and M. Sur, "Visual activity and cortical rewiring: activity-dependent plasticity of cortical networks," *Progress in Brain Research*, vol. 157, pp. 3–11, 2006.
- [102] A. W. Lyckman and M. Sur, "Role of afferent activity in the development of cortical specification," *Results and Problems in Cell Differentiation*, vol. 39, pp. 139–156, 2002.
- [103] D. O. Frost, D. Boire, G. Gingras, and M. Ptito, "Surgically created neural pathways mediate visual pattern discrimination," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 20, pp. 11068–11073, 2000.
- [104] L. von Melchner, S. L. Pallas, and M. Sur, "Visual behaviour mediated by retinal projections directed to the auditory pathway," *Nature*, vol. 404, no. 6780, pp. 871–876, 2000.
- [105] S. L. Pallas, "Cross-modal plasticity as a tool for understanding the ontogeny and phylogeny of cerebral cortex," in

- Cortical Areas: Unity and Diversity*, A. Schuz and R. Miller, Eds., pp. 245–271, Taylor and Francis, New York, NY, USA, 2002.
- [106] S. L. Pallas, M. Xu, and K. A. Razak, “Influence of thalamocortical activity on sensory cortical development and plasticity,” in *Development and Plasticity in Sensory Thalamus and Cortex*, R. Erzurumlu, W. Guido, and Z. Molnar, Eds., pp. 121–137, Springer, Singapore, 2006.
- [107] M. C. Fishman and C. R. Michael, “Integration of auditory information in the cat’s visual cortex,” *Vision Research*, vol. 13, no. 8, pp. 1415–1419, 1973.
- [108] M. T. Wallace, R. Ramachandran, and B. E. Stein, “A revised view of sensory cortical parcellation,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 101, no. 7, pp. 2167–2172, 2004.
- [109] M. Brosch, E. Selezneva, and H. Scheich, “Nonauditory events of a behavioral procedure activate auditory cortex of highly trained monkeys,” *Journal of Neuroscience*, vol. 25, no. 29, pp. 6797–6806, 2005.
- [110] A. A. Ghazanfar and C. E. Schroeder, “Is neocortex essentially multisensory?” *Trends in Cognitive Sciences*, vol. 10, no. 6, pp. 278–285, 2006.
- [111] C. Kayser and N. K. Logothetis, “Do early sensory cortices integrate cross-modal information?” *Brain Structure and Function*, vol. 212, no. 2, pp. 121–132, 2007.
- [112] G. A. Ascoli, L. Alonso-Nanclares, S. A. Anderson et al., “Petilla terminology: nomenclature of features of GABAergic interneurons of the cerebral cortex,” *Nature Reviews Neuroscience*, vol. 9, no. 7, pp. 557–568, 2008.
- [113] H. Markram, M. Toledo-Rodriguez, Y. Wang, A. Gupta, G. Silberberg, and C. Wu, “Interneurons of the neocortical inhibitory system,” *Nature Reviews Neuroscience*, vol. 5, no. 10, pp. 793–807, 2004.
- [114] W. Kilb, “Development of the GABAergic system from birth to adolescence,” submitted to *Neuroscientist*.
- [115] R. Druga, “Neocortical inhibitory system,” *Folia Biologica*, vol. 55, no. 6, pp. 201–217, 2009.
- [116] G. Di Cristo, “Development of cortical GABAergic circuits and its implications for neurodevelopmental disorders,” *Clinical Genetics*, vol. 72, no. 1, pp. 1–8, 2007.
- [117] J. DeFelipe, G. N. Elston, I. Fujita et al., “Neocortical circuits: evolutionary aspects and specificity versus non-specificity of synaptic connections. Remarks, main conclusions and general comments and discussion,” *Journal of Neurocytology*, vol. 31, no. 3–5, pp. 387–416, 2002.
- [118] P. R. Hof, I. I. Glezer, E. A. Nimchinsky, and J. M. Erwin, “Neurochemical and cellular specializations in the mammalian neocortex reflect phylogenetic relationships: evidence from primates, cetaceans, and artiodactyls,” *Brain, Behavior and Evolution*, vol. 55, no. 6, pp. 300–310, 2000.
- [119] J. DeFelipe, “Cortical interneurons: from Cajal to 2001,” *Progress in Brain Research*, vol. 136, pp. 215–238, 2002.
- [120] P. R. Moul, “Neuronal glutamate and GABA_A receptor function in health and disease,” *Biochemical Society Transactions*, vol. 37, part 6, pp. 1317–1322, 2009.
- [121] E. Rossignol, “Genetics and function of neocortical GABAergic interneurons in neurodevelopmental disorders,” *Neural Plasticity*, vol. 2011, Article ID 649325, 2011.
- [122] J. T. Porter, C. K. Johnson, and A. Agmon, “Diverse types of interneurons generate thalamus-evoked feedforward inhibition in the mouse barrel cortex,” *Journal of Neuroscience*, vol. 21, no. 8, pp. 2699–2710, 2001.
- [123] J. F. Staiger, K. Zilles, and T. F. Freund, “Distribution of GABAergic elements ppstsynaptic to ventroposteromedial thalamic projections in layer IV of rat barrel cortex,” *European Journal of Neuroscience*, vol. 8, no. 11, pp. 2273–2285, 1996.
- [124] M. Beierlein, J. R. Gibson, and B. W. Connors, “Two dynamically distinct inhibitory networks in layer 4 of the neocortex,” *Journal of Neurophysiology*, vol. 90, no. 5, pp. 2987–3000, 2003.
- [125] H. A. Swadlow and A. G. Gusev, “Receptive-field construction in cortical inhibitory interneurons,” *Nature Neuroscience*, vol. 5, no. 5, pp. 403–404, 2002.
- [126] T. K. Hensch, M. Fagiolini, N. Mataga, M. P. Stryker, S. Baekkeskov, and S. F. Kash, “Local GABA circuit control of experience-dependent plasticity in developing visual cortex,” *Science*, vol. 282, no. 5393, pp. 1504–1508, 1998.
- [127] R. Mao, J. Schummers, U. Knoblich et al., “Influence of a subtype of inhibitory interneuron on stimulus-specific responses in visual cortex,” *Cerebral Cortex*, vol. 22, no. 3, pp. 493–508, 2012.
- [128] J. A. Heimel, D. van Versendaal, and C. N. Levelt, “The role of GABAergic inhibition in ocular dominance plasticity,” *Neural Plasticity*, vol. 2011, Article ID 391763, 2011.
- [129] T. K. Hensch, “Critical period plasticity in local cortical circuits,” *Nature Reviews Neuroscience*, vol. 6, no. 11, pp. 877–888, 2005.
- [130] L. A. Benevento, B. W. Bakkum, and R. S. Cohen, “Gamma-aminobutyric acid and somatostatin immunoreactivity in the visual cortex of normal and dark-reared rats,” *Brain Research*, vol. 689, no. 2, pp. 172–182, 1995.
- [131] J. M. Blasco-Ibanez, F. J. Martinez-Guijarro, and C. Lopez-Garcia, “Changes in GABA and parvalbumin immunoreactivities in the cerebral cortex of lizards after narine occlusion,” *Brain Research*, vol. 652, no. 2, pp. 334–340, 1994.
- [132] S. Chaudhury, T. C. Nag, and S. Wadhwa, “Prenatal acoustic stimulation influences neuronal size and the expression of calcium-binding proteins (calbindin D-28K and parvalbumin) in chick hippocampus,” *Journal of Chemical Neuroanatomy*, vol. 32, no. 2–4, pp. 117–126, 2006.
- [133] S. Chaudhury, T. C. Nag, and S. Wadhwa, “Calbindin D-28K and parvalbumin expression in embryonic chick hippocampus is enhanced by prenatal auditory stimulation,” *Brain Research*, vol. 1191, pp. 96–106, 2008.
- [134] Y. Jiao, C. Zhang, Y. Yanagawa, and Q. Q. Sun, “Major effects of sensory experiences on the neocortical inhibitory circuits,” *Journal of Neuroscience*, vol. 26, no. 34, pp. 8691–8701, 2006.
- [135] B. D. Philpot, J. H. Lim, and P. C. Brunjes, “Activity-dependent regulation of calcium-binding proteins in the developing rat olfactory bulb,” *Journal of Comparative Neurology*, vol. 387, no. 1, pp. 12–26, 1997.
- [136] E. de Villiers-Sidani, K. L. Simpson, Y. F. Lu, R. C. S. Lin, and M. M. Merzenich, “Manipulating critical period closure across different sectors of the primary auditory cortex,” *Nature Neuroscience*, vol. 11, no. 8, pp. 957–965, 2008.
- [137] M. V. Barbado, J. G. Brión, E. Weruaga et al., “Changes in immunoreactivity to calcium-binding proteins in the anterior olfactory nucleus of the rat after neonatal olfactory deprivation,” *Experimental Neurology*, vol. 177, no. 1, pp. 133–150, 2002.
- [138] Y. Jiao, Z. Zhang, C. Zhang et al., “A key mechanism underlying sensory experience-dependent maturation of neocortical GABAergic circuits in vivo,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 29, pp. 12131–12136, 2011.

- [139] S. Alcantara, E. Soriano, and I. Ferrer, "Thalamic and basal forebrain afferents modulate the development of parvalbumin and calbindin D28k immunoreactivity in the barrel cortex of the rat," *European Journal of Neuroscience*, vol. 8, no. 7, pp. 1522–1534, 1996.
- [140] I. Blumcke, E. Weruaga, S. Kasas, A. E. Hendrickson, and M. R. Celio, "Discrete reduction patterns of parvalbumin and calbindin D-28k immunoreactivity in the dorsal lateral geniculate nucleus and the striate cortex of adult macaque monkeys after monocular enucleation," *Visual Neuroscience*, vol. 11, no. 1, pp. 1–11, 1994.
- [141] R. K. Carder, S. S. Leclerc, and S. H. Hendry, "Regulation of calcium-binding protein immunoreactivity in GABA neurons of macaque primary visual cortex," *Cerebral Cortex*, vol. 6, no. 2, pp. 271–287, 1996.
- [142] A. Cellerino, R. Siciliano, L. Domenici, and L. Maffei, "Parvalbumin immunoreactivity: a reliable marker for the effects of monocular deprivation in the rat visual cortex," *Neuroscience*, vol. 51, no. 4, pp. 749–753, 1992.
- [143] E. P. Botelho, J. Guimarães Martins Soares, S. da Silva Pereira, M. Fiorani, and R. Gattass, "Distribution of calbindin-28kD and parvalbumin in V1 in normal adult *Cebus apella* monkeys and in monkeys with retinal lesions," *Brain Research*, vol. 1117, no. 1, pp. 1–11, 2006.
- [144] D. Tropea, G. Kreiman, A. Lyckman et al., "Gene expression changes and molecular pathways mediating activity-dependent plasticity in visual cortex," *Nature Neuroscience*, vol. 9, no. 5, pp. 660–668, 2006.
- [145] M. V. Sanchez-Vives, L. G. Nowak, V. F. Descalzo, J. V. Garcia-Velasco, R. Gallego, and P. Berbel, "Crossmodal audio-visual interactions in the primary visual cortex of the visually deprived cat: a physiological and anatomical study," *Progress in Brain Research*, vol. 155, pp. 287–311, 2006.
- [146] E. G. Jones, "GABAergic neurons and their role in cortical plasticity in primates," *Cerebral Cortex*, vol. 3, no. 5, pp. 361–372, 1993.
- [147] N. Berardi, L. Domenici, V. Parisi, T. Pizzorusso, A. Cellerino, and L. Maffei, "Monocular deprivation effects in the rat visual cortex and lateral geniculate nucleus are prevented by nerve growth factor (NGF): I. Visual cortex," *Proceedings of the Royal Society B*, vol. 251, no. 1330, pp. 17–23, 1993.
- [148] M. Mainardi, S. Landi, N. Berardi, L. Maffei, and T. Pizzorusso, "Reduced responsiveness to long-term monocular deprivation of parvalbumin neurons assessed by c-Fos staining in rat visual cortex," *PLoS ONE*, vol. 4, no. 2, Article ID e4342, 2009.
- [149] Y. Goldshmit, S. Galley, D. Foo, E. Sernagor, and J. A. Bourne, "Anatomical changes in the primary visual cortex of the congenitally blind *Crx-/-* mouse," *Neuroscience*, vol. 166, no. 3, pp. 886–898, 2010.
- [150] M. Fagiolini and T. K. Hensch, "Inhibitory threshold for critical-period activation in primary visual cortex," *Nature*, vol. 404, no. 6774, pp. 183–186, 2000.
- [151] T. K. Hensch, "Critical period mechanisms in developing visual cortex," *Current Topics in Developmental Biology*, vol. 69, pp. 215–237, 2005.
- [152] T. K. Hensch and M. Fagiolini, "Excitatory-inhibitory balance and critical period plasticity in developing visual cortex," *Progress in Brain Research*, vol. 147, pp. 115–124, 2005.
- [153] Z. J. Huang, A. Kirkwood, T. Pizzorusso et al., "BDNF regulates the maturation of inhibition and the critical period of plasticity in mouse visual cortex," *Cell*, vol. 98, no. 6, pp. 739–755, 1999.
- [154] Y. Iwai, M. Fagiolini, K. Obata, and T. K. Hensch, "Rapid critical period induction by tonic inhibition in visual cortex," *Journal of Neuroscience*, vol. 23, no. 17, pp. 6695–6702, 2003.
- [155] M. Fagiolini, J. M. Fritschy, K. Löw, H. Möhler, U. Rudolph, and T. K. Hensch, "Specific GABA_A circuits for visual cortical plasticity," *Science*, vol. 303, no. 5664, pp. 1681–1683, 2004.
- [156] H. Katagiri, M. Fagiolini, and T. K. Hensch, "Optimization of somatic inhibition at critical period onset in mouse visual cortex," *Neuron*, vol. 53, no. 6, pp. 805–812, 2007.
- [157] J. L. Hanover, Z. J. Huang, S. Tonegawa, and M. P. Stryker, "Brain-derived neurotrophic factor overexpression induces precocious critical period in mouse visual cortex," *The Journal of Neuroscience*, vol. 19, no. 22, article RC40, 1999.
- [158] L. Gianfranceschi, R. Siciliano, J. Walls et al., "Visual cortex is rescued from the effects of dark rearing by overexpression of BDNF," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 100, no. 21, pp. 12486–12491, 2003.
- [159] Y. Hata, T. Tsumoto, and M. P. Stryker, "Selective pruning of more active afferents when cat visual cortex is pharmacologically inhibited," *Neuron*, vol. 22, no. 2, pp. 375–381, 1999.
- [160] H. O. Reiter and M. P. Stryker, "Neural plasticity without postsynaptic action potentials: less-active inputs become dominant when kitten visual cortical cells are pharmacologically inhibited," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 85, no. 10, pp. 3623–3627, 1988.
- [161] T. K. Hensch and M. P. Stryker, "Columnar architecture sculpted by GABA circuits developing cat visual cortex," *Science*, vol. 303, no. 5664, pp. 1678–1681, 2004.
- [162] J. F. M. Vetencourt, M. Vetencourt, A. Sale et al., "The antidepressant fluoxetine restores plasticity in the adult visual cortex," *Science*, vol. 320, no. 5874, pp. 385–388, 2008.
- [163] A. Harauzov, M. Spolidoro, G. DiCristo et al., "Reducing intracortical inhibition in the adult visual cortex promotes ocular dominance plasticity," *Journal of Neuroscience*, vol. 30, no. 1, pp. 361–371, 2010.
- [164] A. Sale, J. F. Maya Vetencourt, P. Medini et al., "Environmental enrichment in adulthood promotes amblyopia recovery through a reduction of intracortical inhibition," *Nature Neuroscience*, vol. 10, no. 6, pp. 679–681, 2007.
- [165] A. Maffei, M. E. Lambo, and G. G. Turrigiano, "Critical period for inhibitory plasticity in rodent binocular V1," *Journal of Neuroscience*, vol. 30, no. 9, pp. 3304–3309, 2010.
- [166] A. Maffei and G. G. Turrigiano, "Multiple modes of network homeostasis in visual cortical layer 2/3," *Journal of Neuroscience*, vol. 28, no. 17, pp. 4377–4384, 2008.
- [167] A. Maffei and G. Turrigiano, "The age of plasticity: developmental regulation of synaptic plasticity in neocortical microcircuits," *Progress in Brain Research*, vol. 169, pp. 211–223, 2008.
- [168] A. Maffei, K. Nataraj, S. B. Nelson, and G. G. Turrigiano, "Potentiation of cortical inhibition by visual deprivation," *Nature*, vol. 443, no. 7107, pp. 81–84, 2006.
- [169] A. Maffei, S. B. Nelson, and G. G. Turrigiano, "Selective reconfiguration of layer 4 visual cortical circuitry by visual deprivation," *Nature Neuroscience*, vol. 7, no. 12, pp. 1353–1359, 2004.
- [170] M. S. Lazarus and Z. J. Huang, "Distinct maturation profiles of perisomatic and dendritic targeting GABAergic interneurons in the mouse primary visual cortex during the critical

- period of ocular dominance plasticity," *Journal of Neurophysiology*, vol. 106, no. 2, pp. 775–787, 2011.
- [171] C. E. Ribak and R. T. Robertson, "Effects of neonatal monocular enucleation on the number of GAD-positive puncta in rat visual cortex," *Experimental Brain Research*, vol. 62, no. 1, pp. 203–206, 1986.
- [172] K. D. Micheva and C. Beaulieu, "Development and plasticity of the inhibitory neocortical circuitry with an emphasis on the rodent barrel field cortex: a review," *Canadian Journal of Physiology and Pharmacology*, vol. 75, no. 5, pp. 470–478, 1997.
- [173] G. W. Knott, C. Quairiaux, C. Genoud, and E. Welker, "Formation of dendritic spines with GABAergic synapses induced by whisker stimulation in adult mice," *Neuron*, vol. 34, no. 2, pp. 265–273, 2002.
- [174] S. H. Hendry and E. G. Jones, "Reduction in numbers of immunostained GABAergic neurones in deprived-eye dominance columns of monkey area 17," *Nature*, vol. 320, no. 6064, pp. 750–753, 1986.
- [175] S. H. Hendry and E. G. Jones, "Activity-dependent regulation of GABA expression in the visual cortex of adult monkeys," *Neuron*, vol. 1, no. 8, pp. 701–712, 1988.
- [176] S. H. Hendry and K. L. Miller, "Selective expression and rapid regulation of GABA_A receptor subunits in geniculocortical neurons of macaque dorsal lateral geniculate nucleus," *Visual Neuroscience*, vol. 13, no. 2, pp. 223–235, 1996.
- [177] B. Morales, S. Y. Choi, and A. Kirkwood, "Dark rearing alters the development of GABAergic transmission in visual cortex," *Journal of Neuroscience*, vol. 22, no. 18, pp. 8084–8090, 2002.
- [178] B. Chattopadhyaya, G. Di Cristo, H. Higashiyama et al., "Experience and activity-dependent maturation of perisomatic GABAergic innervation in primary visual cortex during a postnatal critical period," *Journal of Neuroscience*, vol. 24, no. 43, pp. 9598–9611, 2004.
- [179] Q. Q. Sun, "The missing piece in the "use it or lose it" puzzle: is inhibition regulated by activity or does it act on its own accord?" *Reviews in the Neurosciences*, vol. 18, no. 3-4, pp. 295–310, 2007.
- [180] G. Di Cristo, B. Chattopadhyaya, S. J. Kuhlman et al., "Activity-dependent PSA expression regulates inhibitory maturation and onset of critical period plasticity," *Nature Neuroscience*, vol. 10, no. 12, pp. 1569–1577, 2007.
- [181] B. Chattopadhyaya, "Molecular mechanisms underlying activity-dependent GABAergic synapse development and plasticity and its implications for neurodevelopmental disorders," *Neural Plasticity*, vol. 2011, Article ID 734231, 2011.
- [182] Z. J. Huang, "Activity-dependent development of inhibitory synapses and innervation pattern: role of GABA signalling and beyond," *Journal of Physiology*, vol. 587, part 9, pp. 1881–1888, 2009.
- [183] Z. J. Huang, G. Di Cristo, and F. Ango, "Development of GABA innervation in the cerebral and cerebellar cortices," *Nature Reviews Neuroscience*, vol. 8, no. 9, pp. 673–686, 2007.
- [184] T. Pizzorusso, P. Medini, S. Landi, S. Baldini, N. Berardi, and L. Maffei, "Structural and functional recovery from early monocular deprivation in adult rats," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 103, no. 22, pp. 8517–8522, 2006.
- [185] S. Sugiyama, A. Prochiantz, and T. K. Hensch, "From brain formation to plasticity: insights on Otx2 homeoprotein," *Development Growth and Differentiation*, vol. 51, no. 3, pp. 369–377, 2009.
- [186] S. Desgent, D. Boire, and M. Ptito, "Distribution of calcium binding proteins in visual and auditory cortices of hamsters," *Experimental Brain Research*, vol. 163, no. 2, pp. 159–172, 2005.
- [187] R. W. Guillery, "Anatomical evidence concerning the role of the thalamus in corticocortical communication: a brief review," *Journal of Anatomy*, vol. 187, part 3, pp. 583–592, 1995.
- [188] R. W. Guillery and S. M. Sherman, "Thalamic relay functions and their role in corticocortical communication: generalizations from the visual system," *Neuron*, vol. 33, no. 2, pp. 163–175, 2002.
- [189] O. Caillard, H. Moreno, B. Schwaller, I. Llano, M. R. Celio, and A. Marty, "Role of the calcium-binding protein parvalbumin in short-term synaptic plasticity," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 24, pp. 13372–13377, 2000.
- [190] S. Patz, J. Grabert, T. Gorba, M. J. Wirth, and P. Wahle, "Parvalbumin expression in visual cortical interneurons depends on neuronal activity and TrkB ligands during an early period of postnatal development," *Cerebral Cortex*, vol. 14, no. 3, pp. 342–351, 2004.
- [191] M. Vreugdenhil, J. G. R. Jefferys, M. R. Celio, and B. Schwaller, "Parvalbumin-deficiency facilitates repetitive IPSCs and gamma oscillations in the hippocampus," *Journal of Neurophysiology*, vol. 89, no. 3, pp. 1414–1422, 2003.
- [192] T. K. Hensch, "Recovery in the blink of an eye," *Neuron*, vol. 48, no. 2, pp. 166–168, 2005.
- [193] Z. J. Huang and G. Di Cristo, "Time to change: retina sends a messenger to promote plasticity in visual cortex," *Neuron*, vol. 59, no. 3, pp. 355–358, 2008.

Review Article

Studies of Olfactory System Neural Plasticity: The Contribution of the Unilateral Naris Occlusion Technique

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Received 16 December 2011; Revised 1 March 2012; Accepted 19 March 2012

Academic Editor: Ron Kupers

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Unilateral naris occlusion has long been the method of choice for effecting stimulus deprivation in studies of olfactory plasticity. A significant body of literature speaks to the myriad consequences of this manipulation on the ipsilateral olfactory pathway. Early experiments emphasized naris occlusion's deleterious and age-critical effects. More recent studies have focused on life-long vulnerability, particularly on neurogenesis, and compensatory responses to deprivation. Despite the abundance of empirical data, a theoretical framework in which to understand the many sequelae of naris occlusion on olfaction has been elusive. This paper focuses on recent data, new theories, and underappreciated caveats related to the use of this technique in studies of olfactory plasticity.

1. Introduction

Ideas concerning the influence of deprivation and enrichment on the quality of human relationships can be summed up in the following aphorisms: concerning deprivation it is said that “absence makes the heart grow fonder,” a sentiment that has been expressed at least since Roman times; concerning abundance, Chaucer tells us that “familiarity breeds contempt,” a perhaps cynical but resonant view of human nature. Enlightenment thinkers, too, were keenly interested in the effects of deprivation and enrichment—not on the heart—but on the organ of thought. In 1688 the Irish politician and scientist William Molyneux posed a question, in a letter to John Locke, later known as “Molyneux’s Problem,” concerning the role of experience in visual perception that was to occupy philosophers and scientists for the ensuing three centuries [1]. Later, Charles Darwin considered the heritable effects of deprivation and enrichment on the nervous system. He concluded in his 1868 opus entitled *The Variation of Animals and Plants Under Domestication*, after observing smaller crania in domesticated rabbits compared to their wild counterparts, “We thus see that the most important and complicated organ in the whole organization is subject to the law of decrease in size from disuse [2].” But it is the neuroanatomist and

psychiatrist, Bernhard von Gudden, working at the same time as Darwin, who should be credited with pioneering the neurobiological study of sensory manipulation on brain development [3]. Among his other innovations, Gudden developed the unilateral deprivation model. Brilliant in its simplicity, this paradigm affords a within-subject comparison of different amounts of sensory stimulation on brain development. A century before Hubel and Wiesel won their 1981 Nobel Prize, in part, for their studies of the effects of unilateral deprivation by lid suture on visual cortex, Gudden had already invented the method and described its effects on the visual system in his monograph of 1870 [4, 5]. In this same series of studies he also described, for the first time, the effects of unilateral nostril occlusion (UNO) on the olfactory system, the topic of this paper. Gudden established that occluding one nostril of newborn rabbits caused a pronounced reduction in the size of the olfactory bulb on the same side after six weeks (Figure 1). While the history of UNO studies is neither as bulging nor ballyhooed as that of unilateral eyelid suture, the technique has, nevertheless, formed the mainstay of olfactory neuroplasticity studies and remains in active use more than four generations after its invention. In 1994 Brunjes provided an excellent review of the literature, up to that time, on the effects of UNO on

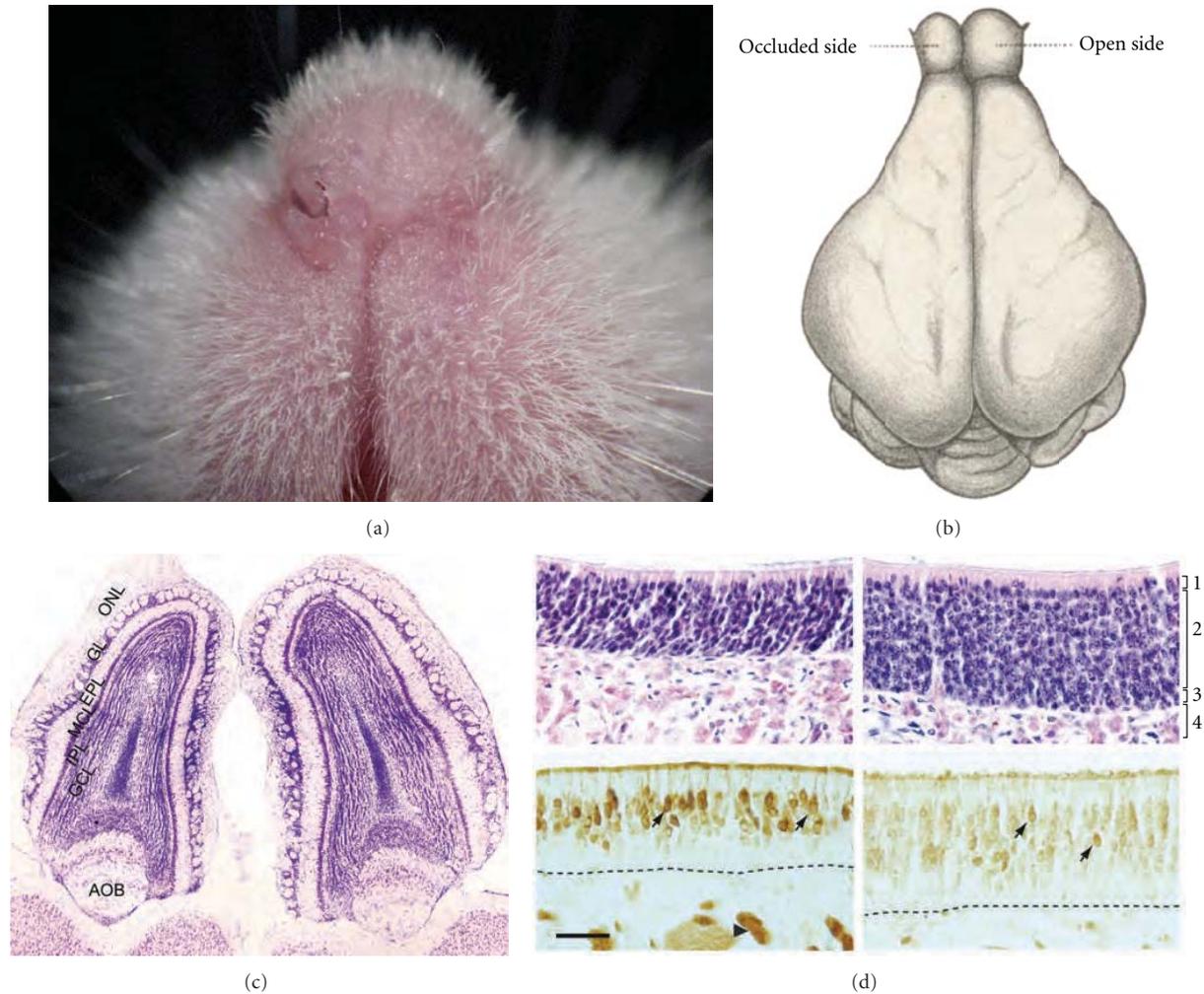


FIGURE 1: Anatomical and histological effects of UNO. (a) Adult mouse that underwent left UNO as a neonate. Note the normal right naris and apparently unaffected morphology around the location of the occluded naris. (b) Bernhard von Gudden's drawing of the brain from a young adult rabbit that had undergone UNO as a neonate ([4]; dorsal view reflected right/left to match other panels; occl: occluded). (c) Horizontal section through the olfactory bulbs of a young-adult mouse that had UNO as a neonate. Note all layers of the occluded bulb (left) are thinner than open-side bulb on the right (ONL: olfactory nerve layer; GL: glomerular layer; EPL: external plexiform layer; MCL: mitral cell layer; IPL: internal plexiform layer; GCL: granule cell layer; AOB: accessory olfactory bulb). (d) Histological sections through olfactory mucosa of young-adult mouse that had UNO as a neonate. Right column: open-side; left column: occluded side; top row: H&E stain; bottom row: OMP immunolabeling (arrows: mature OSN cell bodies) (layers: 1: sustentacular layer; 2: olfactory receptor cells; 3: basal cell layer; 4: lamina propria).

the olfactory system [6]. Thus, after a brief discussion of olfaction and UNO phenomenology, the current paper will focus on more recent findings, new interpretations of the older literature, and remaining questions.

2. Basic Phenomenology

The mammalian olfactory system consists of an olfactory mucosa, sequestered in the dorsocaudal part of the nasal cavity, from which olfactory sensory neurons (OSNs) project their axons to the olfactory bulbs, rostral extensions of the telencephalon. At the bulbs, OSNs form synapses with output neurons, the mitral and tufted cells, in neuropil structures known as glomeruli. The largest known mammalian

gene family codes for olfactory receptor (OR) proteins, a given OSN expressing but one of ~1000 genes in the mouse ([9]; Figure 2). All the OSNs expressing a given OR across the nasal cavity converge onto only a couple glomeruli, typically one medial and one lateral in each bulb. Odor information arriving at the bulb from the OSNs is processed by a highly laminar and complex set of direct and indirect pathways present in this well-studied structure [9]. Juxtglomerular cells which, as their name implies, are part of the glomerular circuit and granule cells residing deeper are the key inhibitory interneurons of the bulb and are particularly important to our story. From the Mitral and Tufted cells olfactory information is transmitted to a group of central targets collectively known as the primary olfactory

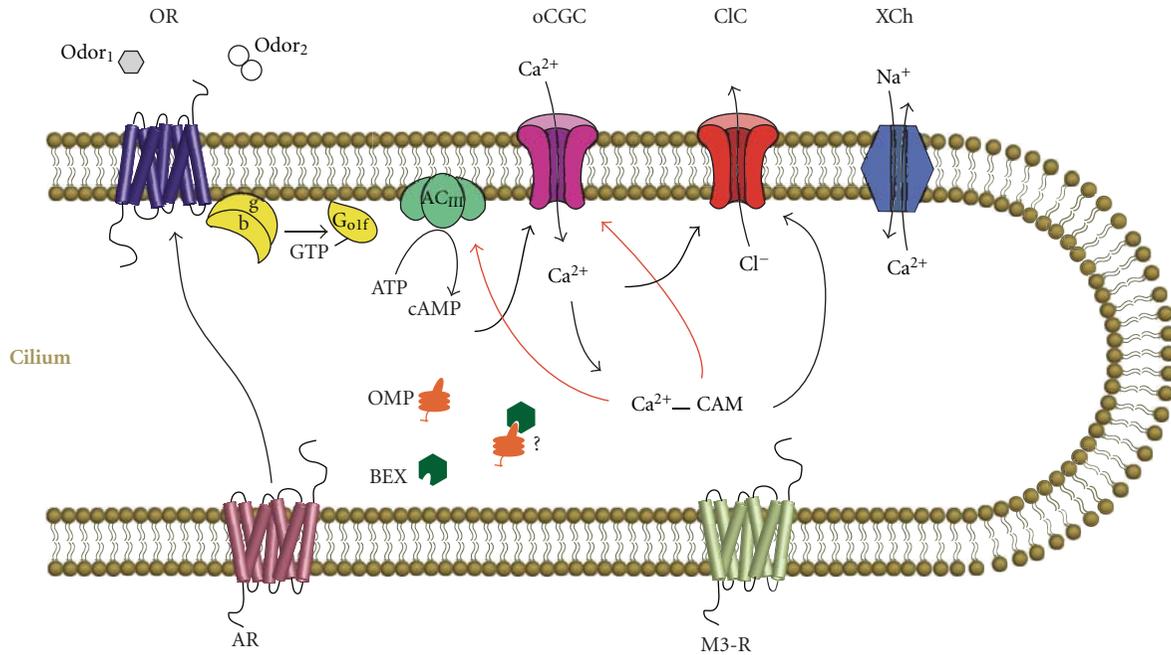


FIGURE 2: Simplified model of olfactory signal transduction within OSN cilium. AC_{III}: adenylyl cyclase; AR: adrenergic receptor; BEX: Brain-expressed X-linked protein; CIC: sodium/calcium exchanger; G_{olf}: olfactory g-protein; M3-R: muscarinic acetylcholine receptor; oCGC: olfactory cyclic nucleotide-gated channel; OMP: olfactory marker protein; OR: odorant receptor. The Na⁺/K⁺/Cl⁻ cotransporter is not shown. Black arrows: stimulation; red arrows: inhibition (after [7] cf. [8]).

cortex including accessory olfactory nucleus, the piriform cortex, the entorhinal cortex, and the amygdala. It is in these central areas where a smell is given its appropriate perceptual and emotion qualities [9]. The following discussion will briefly consider the major developmental effects of UNO on each of the three tiers of the olfactory system starting with the bulb since this structure has received the most attention.

2.1. Olfactory Bulb. Gudden's principal discovery that UNO performed in the neonate causes the ipsilateral olfactory bulb to fail to reach its normal adult size has been replicated repeatedly in a number of different species (e.g., [10–13]; see Figure 1). Like Gudden, modern investigators have logically assumed that UNO's effects are due to odor deprivation to the occluded nasal fossa, though, as we will see, this assumption has only rarely been tested given the difficulty of olfactory deprivation by other means. The diminution of the ipsilateral olfactory bulb following UNO is due, in part, to reduction in the external plexiform and glomerular layers [14]. Indeed, the size of glomeruli, as judged in transgenic P2-receptor reporter mice, is smaller in the occluded bulb compared to the nonoccluded bulb within weeks of early postnatal occlusion [15–17]. However, the most dramatic decline after UNO in the ipsilateral bulb, by far, is in the granule cell layer ([14]; Figure 1).

Earlier studies using tritiated thymidine and more recent studies using bromodeoxyuridine establish that the loss of granule cell layer volume from the occluded-side bulb is predominantly due to decreased cell survival not decreased neurogenesis [18, 19]. In contrast to these anatomical

sequelae of UNO, which take weeks to detect, metabolic effects can be quite rapid. In the occluded-side olfactory bulb decreased 2-deoxyglucose uptake and Krebs-cycle enzyme immunochemistry is apparent in a matter of days after UNO [20, 21]. Also a rapid decline in protein synthesis, as measured by radiolabeled amino acid uptake, and change in gene expression, as measured by *in situ* hybridization, have been reported ([6, 22]; see the following).

Concerning bulb neurochemistry, an early observation was that tyrosine hydroxylase, the rate limiting step of dopamine synthesis, is markedly reduced in the ipsilateral bulb within days of nostril occlusion, an effect that can be reversed by reopening the nostril of experimental animals [23–25]. Tyrosine hydroxylase and dopamine content of juxtglomerular cells, the predominant dopaminergic neurons of the bulb, decrease ipsilaterally after all of the following: UNO, olfactory nerve axotomy, or chemical lesion of the olfactory mucosa [24, 26, 27]. UNO causes a downregulation of β_1 and β_2 -adrenergic receptors [28] but may have no effect on norepinephrine receptors [29]. Glutamate receptors, as a family, are not known to be affected by UNO but GluR1-positive short-axon cells are much reduced ipsilaterally in the external plexiform layer [30].

Neurotrophic factors, neuromodulators, and their receptors in the bulb are affected by UNO. Nerve growth factor receptors are increased on the occluded side 19 and 60 days after neonatal occlusion in the rat [31]. Brain-derived neurotrophic factor is initially increased and then later decreases ipsilateral to occlusion [32]. Insulin receptor kinase is downregulated on the occluded side; interesting

since this receptor and its ligand are implicated in ion channel modulation as is BDNF [33–35]. The mitogen-activated protein kinase/extracellular signal-regulated kinase (MAPK/ERK) pathway, part of a key signaling-cascade, is also downregulated ipsilateral to occlusion [36].

Some attention has been paid to the physiological or circuit effects of UNO on the bulb. In rats, three weeks of occlusion, starting on the day after birth, causes enhanced inhibition of mitral/tufted (M/T) cells in response to paired-pulse stimulation of the lateral olfactory tract [37, 38], an effect which appears to be NMDA receptor mediated [39]. In adult rats, either short-term (1-2 months) or long-term (12 months) UNO increased the proportion of M/T cells from the ipsilateral bulb that respond to multiple odorants suggesting a decrease in discrimination [40]. And in young rats even 15 min of naris occlusion causes a decoupling of M/T responses from respiration [41]. In a recent study, early UNO slowed the morphological development of ipsilateral mitral cells and checked the changes in membrane conductance and coupling coefficients that are part of the normal maturational shift from electrical to chemical synapses in the bulb [42]. Despite these results, electrophysiological studies have, for the most part, failed to show differences in the circuit properties of the ipsilateral bulb after UNO that are commensurate with the profound structural changes reviewed previously [43].

2.2. Mucosa. Studies of the effects of UNO, as in the case with visual and auditory deprivation, have tended to focus on central effects such that relatively less is known about the changes in the periphery resulting from this procedure. Metabolism, as measured by succinate dehydrogenase histochemistry, is measurably reduced in the ipsilateral mucosa of newborn rats within two days of occlusion [44]. In the mouse, rat, and rabbit, UNO leads to a substantial decrease in the thickness of the respiratory and olfactory mucosa ipsilaterally ([10, 45, 46]; see Figure 1). In the rat, early UNO causes a decline in the rate of mitosis in the olfactory epithelium [45, 47], a condition that can be reversed in a matter of days by reopening the closed naris [48]. Despite the difference in mucosal thickness, the number of mature olfactory sensory neurons is apparently unaffected by nostril occlusion in the mouse and rat ([10, 45] but see [15, 49]) though there appears to be a decrease in the rabbit, an inconsistency which has been attributed to the lack of a nasopharyngeal canal in the latter species [6, 46]. More recently, a histological study of rats occluded unilaterally from near birth to 60 days of age reported reorganization of tissue types in both the ipsilateral and contralateral mucosa compared to controls, most notably an expansion of olfactory mucosa on the occluded side [50]. In this vein, turbinate shape and positioning in three- to four-week-old mice, which had undergone UNO on the day after birth, are also affected such that the occluded-side turbinates take on a fine filigree-like appearance, especially rostrally, compared to their more robust partners on the open side [51]. Finally, a rapidly growing line of research is implicating OSN activity in the expression of axon guidance molecules. In these studies, UNO is often used to compare the abundance of

guidance molecules in open and occluded-side mucosa as evidence for their activity dependence (e.g., [52–54]).

2.3. Central Pathways. The potentially important topic of the effects of early UNO on the development of central pathways has unfortunately garnered rather little attention judging by the literature. In one study, the thickness of piriform cortex layer 1b and the size of semilunar cell dendrites were reduced ipsilaterally in postnatal day (PND) 30 rats occluded on PND1 [55]. In a recent study, the expression of the NMDA receptor NR2B and the phosphorylated form of the regulator element CREB were downregulated in the piriform five days after naris occlusion, an effect which could be fully reversed ten days after reopening of the naris [56]. And early postnatal UNO in rats delays the normal developmental increase in the ratio of AMPA receptors to NMDA receptors at primary sensory synapses but not associational synapses on pyramidal neurons in piriform cortex slice preparations [57]. In a previous related study, field potential recordings from intact anterior piriform cortex establish an ipsilateral depression of responses evoked by stimulation of cortical afferents in early (PND1) but not late (PND 30) occlusion [58]. However, in this study evoked potentials in intracortical association fiber were enhanced ipsilaterally in both early and late-onset UNO rats.

Concerning other central olfactory structures, UNO from PND 1–20 caused a decrease in 2-deoxyglucose uptake in the rostral anterior olfactory nucleus [59]. However, published reports on the effects of UNO on other central olfactory structures such as the amygdala and entorhinal cortex are lacking.

Collectively, these rather modest effects ipsilateral to UNO in higher brain centers have been attributed to the bilateral inputs of these structures [6]. Nevertheless, given the current paucity of studies more work in this area would be of benefit.

2.4. Human Studies. Olfactory bulb size measured by MRI has been positively correlated with olfactory psychophysical test scores in clinical populations recovering from head-trauma as well as in normal adults and in young people [60–62]. That this relationship is causally linked, and more importantly, plastic over the lifetime, is suggested by research showing that patients with the most severe chronic rhinosinusitis (CRS) tended to have the smallest olfactory bulb volumes and poorest olfactory performance [63]. Moreover, a longitudinal study of CRS patients under a standard treatment regimen showed an increase in bulb size accompanied by a decrease in odor thresholds after treatment [64]. Together, these clinical findings suggest that levels of peripheral input in humans may affect cell survival in the olfactory bulb and other olfactory processes as they do in rodents (see the following). More to the point, there has been at least one study of short-term (one-week) UNO in humans accompanied by a similar duration recovery period [65]. Psychophysical testing and fMRI analysis of subjects after deprivation and again after recovery provide modest evidence that odor deprivation induced a reversible

increase in odor detection with a concomitant decrease in the specificity of odor coding in the piriform cortex.

3. A Method in Search of a Theory

While admittedly the aforementioned literature review focuses on positive results, it would appear that wherever one looks, and whatever the endpoint—anatomical, biochemical, or physiological—UNO has marked effects on the ipsilateral olfactory system. However, a theoretical framework in which to place the myriad effects of naris occlusion has been more elusive. One obvious solution, contemplated by Meisami [12] upon his modern rediscovery of Gudden's method, was to place these findings in the now massive corpus establishing the indispensable role of activity in normal neural development [66, 67]. In this formulation, neural activity is believed, through Hebbian mechanisms, to strengthen appropriate functional connections and weaken, ultimately weeding out, inappropriate connections [6]. It is perhaps ironic that arguably the best understood example of this process comes from studies of the effects of monocular lid suture (Gudden's technique) on ocular dominance column formation in the visual cortex [68]. But there are reasons to question whether Hebb's postulate and the unavoidable comparisons to ocular dominance plasticity are really appropriate to UNO phenomenology. For example, the notion that sensory experience might play an instructive role in the layout of the olfactory map in the bulb has collapsed under the weight of contrary evidence. Modern genetic approaches, which have allowed the creation of mouse strains lacking essential components of the transducing cascade, establish that the proper guidance of sensory cell axons to bulbar targets does not require sensory activity or even functional synaptic release, though spontaneous activity (i.e., nonsensory driven and presumably uncorrelated) seems to be necessary ([69] reviewed in [9]). Given the problem that developmental linguists have colorfully referred to as the "poverty of the stimulus", it was probably never a tenable proposition that >1000 types of OSNs could sort their axons in a matter of a few days of development, based on sensory-driven correlated activity [69]. Thus, Hebb's postulate finds little succor in what we have recently learned about the development of the bulb odor map; however, intrabulbar connections may be a different matter [15]. But there are additional problems to consider with the analogies between experience-dependent plasticity in other sensory systems and the effects of UNO on the olfactory system.

3.1. Critical Period. One line of evidence in favor of the paradigm described previously comes from early evidence that the effects of UNO have a "critical period" like the sharply defined boundaries of ocular dominance plasticity. In rats, it was shown that UNO from PND 1–30 caused the typical 25% reduction in ipsilateral bulb size, while occlusion from PND 30–60 or PND 60–90 had little effect [70]. However, in subsequent studies many of the effects on the ipsilateral bulb after early postnatal UNO have been shown to accrue after adult occlusion including reductions in size [71–73], neurogenesis (e.g., [72]), and granule cell branching

[74]. Adult UNO also causes a rapid bilateral increase in cell death within layer I/II of the piriform [75]. Moreover, unlike the case for ocular dominance column plasticity, UNO effects are reversible, for an apparently indefinite period, upon naris reopening [76]. Thus, the bulk of the available data does not support a "critical" or sensitive period for UNO effects on the olfactory bulb.

3.2. Firing and Wiring. At an even more fundamental level, however, the analogy between activity-dependent processes in other sensory systems and the phenomenology of UNO seem inapt, particularly concerning its effects on the bulb. The profound reduction in bulb size that accompanies UNO has no obvious precedent in other sensory systems deprived of their appropriate stimulus. In the visual system, even total stimulus deprivation by dark-rearing does not cause anything like the size change and cell loss seen in the olfactory bulb after UNO [68]. For example, in the retina, the structure in the visual pathway perhaps most analogous to the olfactory bulb, dark-rearing causes a decreased pruning of retinal ganglion cell dendrites not a net loss of cells [77]. Even the celebrated decrease in deprived-eye ocular dominance columns in visual cortex after monocular deprivation is due to the competitive environment of ingrowing binocular inputs [68]. No analogous competition exists in the exclusively unilateral afferents to the olfactory bulb. Indeed, dark-rearing has rather modest effects on the size and wiring of visual pathways provided that it does not persist too long into young adulthood [60]. Importantly, it is the *pattern* not the total amount of activity that seems to be the critical determinant of experience-dependent plasticity in the visual system [68, 77]. These aspects of the effects of stimulus deprivation on the visual system also appear to apply to the somatosensory system [77].

3.3. Persistent Neurogenesis. Admitting that the effects of UNO on the olfactory bulb are unique among sensory systems, one has to seek unique explanations. A singular and striking feature of the olfactory system is its continuous turnover of OSNs and supply of new interneurons to the olfactory bulb from the rostral migratory stream (RMS).

In the olfactory mucosa precursor cells near the basal lamina (Figure 1) divide to become new OSNs sending dendrites toward the mucosal surface and axons that make their way to the olfactory bulb where they gain functional connection in glomeruli (reviewed by [78]). This process, which occurs throughout life, underlies the replacement of dying mature neurons in a cycle with a period of a few months [78].

The bulb's continuous supply of new RMS neurons differentiates predominantly into juxtglomerular cells and granule cells, both inhibitory interneurons. A spate of recent studies establishes that some of these adult-born neurons survive and become functionally integrated in an activity-dependent manner (reviewed in [79, 80]). As noted previously, UNO decreases the survival of adult-born neurons in the bulb and, amazingly, olfactory "enrichment" increases their survival [72, 80]. Behavioral studies suggest that the incorporation of adult-born neurons into the olfactory bulb

may actually play important functional roles in certain types of olfactory learning and memory [80]. While a complete review of the burgeoning literature on olfactory bulb adult-born neurons is beyond the scope of this paper, it is mentioned here as a potential explanation for the dramatic effects of UNO on bulb size and other parameter. Given that most of the decrease in ipsilateral bulb size after UNO is related to granule cells loss and stimulus-driven activity is necessary for granule cell survival, it follows that UNO, which surely reduces stimulus driven activity, would lead to a decline in bulb size in adults and failure to reach full size in young animals. Of course this analysis merely begs the question of why the visual, auditory, and somatosensory systems manage to function efficiently without ongoing neurogenesis. One suggestion is that bulbar neurogenesis can be understood in the context of a neural circuit in which the inputs—the OSNs—are undergoing continuous turnover for the purpose of adjusting to an ever-changing odor environment [80]. An important caveat to this line of reasoning emerges from recent studies of the RMS in humans [81]. While infants less than 18 months have an extensive population of newborn migrating neurons forming a substantial RMS, this germinal activity subsides thereafter and is virtually nil in adulthood. At least on its face, this result seems at odds with the view that adult-born neurons from the RMS play an essential role in ongoing olfactory function, especially given the very respectable olfactory capabilities in the average adult human [82].

Apart from the recent human data, the literature on olfactory neurogenesis is perplexing. As we have seen UNO causes a reduction in neurogenesis in the olfactory mucosa of young and adult animals and a decrease in granule cell survival in the olfactory bulb [45, 47, 72]. Interestingly, UNO also causes decreased neurogenesis within the non-sensory respiratory epithelium leading to the suggestion that something in respired air other than odors, such as toxins, microorganisms, or other foreign particles, might be driving mitogenesis [45]. Evidence in support of this idea came from an early study showing that many OSNs could live for up to a year in mice (the species' entire normal life expectancy) provided that the animals were raised in a laminar flow hood to prevent nasal infections [83]. Add to these findings a recent study showing that odor exposure, in bulbectomized mice, rescues OSNs from the apoptosis that usually accompanies this manipulation [84]. Whether odors or other factors in respired air govern the death and replacement of OSN in the olfactory mucosa and how this, in turn, effects death and replacement of interneurons in the bulb remain open questions, as does the evolutionary significance of this unprecedented plasticity.

Thus, it must be concluded that, despite the existence of so many excellent empirical studies, a satisfactory theoretical framework in which to understand the effects of UNO on the olfactory system has eluded us so far!

4. Problematic Paradigm

The cliché “nothing succeeds like success” surely applies to the UNO technique. As already noted, the number of

sequelae of this manipulation within the olfactory system is large and growing. It remains the method of choice for stimulus deprivation available to any investigator with a caution. However, the assumptions underlying its application have rarely been tested.

4.1. Deprivation. Unlike the case with dark-rearing or lid-suture (contrast deprivation) in vision studies, nothing like complete deprivation is achieved by UNO. Electrophysiological recordings in rats [41] and Fos immunohistochemistry in ferrets [85] reveal odor-driven activity in the ipsilateral bulb even after acute UNO. Using either acute or long-term UNO paradigms, rats that have undergone contralateral bulbectomy can detect odors at extremely low concentrations [86, 87]. And similarly prepared newborn mice can use odor cues to find their mother's nipple and navigate back to the nest [88]. Finally, adult mice that received UNO as newborns and contralateral bulbectomy as adults can perform *better* than controls (unilateral bulbectomy *without* contralateral UNO) in both odor habituation/cross-habituation and operant testing ([89]; see the following). One explanation for these results is that in rodents and some other mammals there is a surprisingly large nasopharyngeal canal that could allow odor mixing between the open and occluded nasal fossa [86]. Also, odors undoubtedly gain access to the occluded fossa by a retronasal route. Indeed the negative pressure that must be created in the occluded fossa with each inhalation *guarantees* a substantial exchange of air between the occluded fossa and the outside world by internasal and retronasal passage.

One last point on the deprivation achieved by UNO concerns the question of *what* the nasal cavity is actually being deprived of. The profound and comprehensive effect that UNO has on the interneuron population of the ipsilateral bulb stands in stark contrast to the subtotal, potentially regional, and environmentally dependent deprivation that occurs upon occluding a naris. Already noted are the substantially preserved olfactory capabilities of rodents forced to smell with only their occluded-side olfactory system intact, a feat requiring the rerouting of odor entry to the nasal fault. These considerations suggest that the interneuron population in some regions of the olfactory bulb should be spared by UNO. Even more fundamentally, the odor environment of the average laboratory or animal facility must be impoverished compared to a natural environment. Given this situation it seems likely that most of the 1000s of different types of OSNs (based on the olfactory receptor they express) are deprived of their specific ligand most of the time in the laboratory environment. In this light, the global effects of UNO on the bulb are all the more surprising. Is not deprivation in an impoverished environment of less moment than deprivation in an enriched environment? Considering these facts it is interesting that OSNs, in addition to responding to odor ligands, are exquisite mechanoreceptors [90]. There can be little doubt that UNO causes marked and global decreases in mechanical force in the occluded fossa that would normally accompany respiratory airflow. Thus, it could be speculated that mechanical force deprivation may explain the global effects of UNO on the bulb provided that

one also posits a role for OSN mechanical transduction in this activity dependence process.

In search of an effect of nostril occlusion commensurate with the global effects on the interneuron population of the bulb it is worth noting, as mentioned previously, that other factors in air: irritants, microbes, toxic substances, and the like, that the occluded-side nasal fossa is partially protected from, should be given more attention as potential causes of mucosal and bulbar changes following UNO. It is relevant in this regard that trigeminal sensory fibers richly innervate the olfactory mucosa sending collaterals to the olfactory bulb where they are thought to have a modulator influence [91]. Given the existence of this circuit, it seems possible that some of the effects of UNO on the ipsilateral bulb may be related to interference with the normal interplay between the trigeminal and olfactory systems.

4.2. Specificity. Another implicit assumption of the UNO technique is that its effects are systemically benign and limited to olfaction. However, investigators have repeatedly shown that animals grow at a slower rate after UNO compared to controls (e.g., [12, 88]). In contrast to deprivation directed at the eye or the ear, the nasal cavity has a number of functions besides olfaction not least respiration. Local reductions in oxygen, increases in carbon dioxide, and the aforementioned protection from drying, irritants, and microbes could all be factors underlying some of the effects of UNO. To this point and as noted previously, turbinate morphology is abnormal on the occluded side of young adult mice after early postnatal UNO, an effect unlikely to be related to odor deprivation [51]. Finally, we have recently compared the transcriptomes of ipsilateral and contralateral UNO mice to those of untreated mice ([92]; Figure 3). A number of genes, seemingly unrelated to olfaction, are regulated in the occluded-side mucosa and bulb, casting further doubt on the specificity assumption.

4.3. Contralateral Control. One of the most egregious suspensions of the basic tenants of experimental design occurs routinely in experiments using the UNO technique. Perhaps because some of the early investigators, dating back to Gudden, assumed that the contralateral side was “normal” and if true that this would afford a powerful within-subjects experimental design, many modern UNO aficionados use few or no control subjects in their studies. Yet, common sense and experimental evidence suggest that the contralateral side of UNO subjects is not normal. While on the occluded side the airflow is dramatically reduced, especially rostral to the nasopharyngeal canal, the open side is forced to carry a larger-than-normal volume of air (presumably twice the amount). Also, UNO abrogates alternating cycles of breathing, forcing constant duty on the open side. Not surprisingly this leads to detectable histological and physiological changes in the contralateral mucosa. Most dramatic is the profound loss of OSNs in the rostral end of the nasal cavity after long-term (\geq six weeks) UNO in mice [93, 94]. Similarly, a suite of histological abnormalities has been noted in the contralateral mucosa of rabbits [95] and rats [50] after UNO. In mice, there is hypertrophy of turbinates from the

contralateral nasal fossa compared to those from untreated subjects, within 18 days of early postnatal UNO [51]. Finally, one finds several differentially regulated genes comparing the transcriptomes of control and contralateral mucosa and bulb in young-adult mice that received UNO as newborns ([92]; Figure 3(a)). Some of these genes are not yet annotated, and others seemingly are unrelated to olfaction; however, they stand as existential proof that the contralateral side of UNO subjects is not “normal.”

4.4. Compensatory Processes. In contrast to the Hebbian view of olfactory development discussed previously, evidence has recently accumulated for the opposite proposition. Indeed many of the changes in the olfactory system following UNO appear to be compensatory in that they cause changes in the system that work to preserve olfactory function in the face of sensory deprivation [96]. For example, olfactory bulb neurotransmitter systems seem to follow this pattern in that the decrease in ipsilateral bulb dopamine following UNO is compensated for by a $>30\%$ increase in dopamine D₂ receptors that cannot be ascribed to shrinkage of lamina [97]. Analogously, the increase in ipsilateral bulb norepinephrine is compensated, in part, by a decrease in norepinephrine receptors [96]. Also, while UNO may cause an ipsilateral decrease in the extent of glomerular neuropil and the dendritic arbor of mitral cells, it also causes a more uniform distribution of a synaptic protein synaptophysin, a response that may be viewed as compensatory [96]. Finally, in a recent study, the depletion of ipsilateral granule cells following UNO appears to be compensated for by an increased excitability among the remnant granule cell population [19]. All of these examples may help explain how the ipsilateral olfactory bulb of animals subjected to long-term UNO appear to function normally, as far as we know, despite its abnormal morphology.

Evidence of compensation also abounds at the first synapses of the olfactory system and in the periphery. Tyler et al. published among the first detailed studies of the effects of UNO on primary and secondary synapses in the olfactory system [98]. Using the whole-cell voltage-clamp technique in a rat slice preparation, they showed that two weeks of olfactory deprivation, beginning on PND2, increases the probability and quantal content of neurotransmitter release at primary olfactory synapses in the ipsilateral bulb. This effect of UNO could be demonstrated as early as three days after the onset of naris occlusion in young adult rats. Furthermore, immunolabeling of the vesicular glutamate transporter and two glutamate receptor subunits demonstrated that UNO caused an upregulation of these components at ipsilateral primary olfactory synapses. Voltage-clamp recordings of spontaneous and olfactory-nerve-evoked activity in the predominant second-order neurons of the bulb, including mitral cells, revealed that UNO also strengthens synapses in down-stream components of the olfactory circuit. This latter finding may explain earlier observations that the size and intensity of odor-induced 2-deoxyglucose foci are increased in the ipsilateral-bulb glomerular-layer of UNO rats after reopening the occluded naris [43]. In this earlier study it was observed that

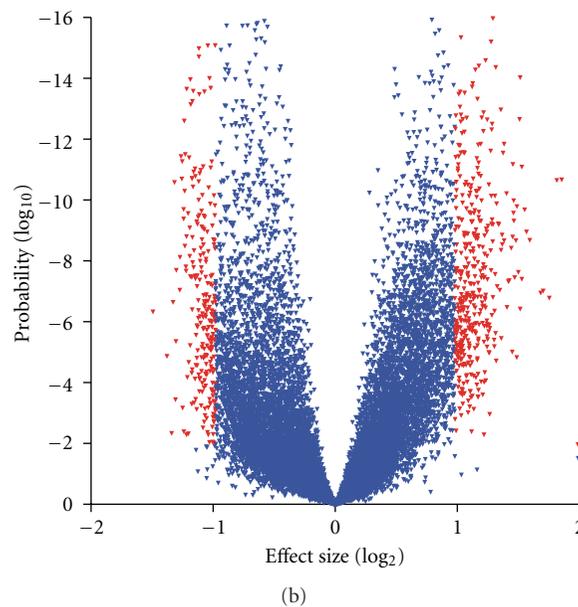
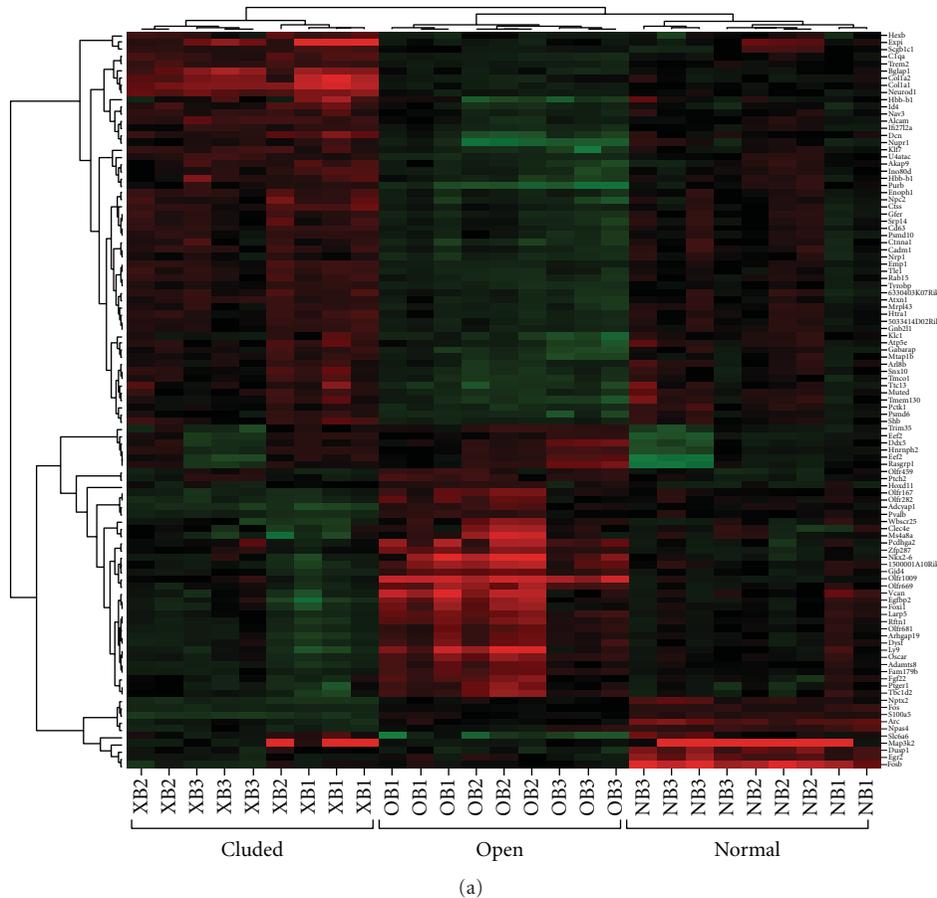


FIGURE 3: Microarray analysis of the effects of early postnatal UNO on olfactory bulb transcriptome of young adult mice. (a) Expression profile of 103 genes from the >20,000 total on the chip that met arbitrary significance criteria (2.25-fold up, or 2.25-fold down, with $P < 0.01$). Tissue source is shown on bottom axis. Note that there were three technical replicates within each of three biological replicates (subscript numbers). Color represents expression value with red, upregulation and green, downregulation. Dendrograms based on expression values show clustering genes (left) and samples (right), respectively. Note large number of up- and downregulated genes in both occluded and open (nonoccluded) bulb with normal bulb showing intermediate expression of most genes. (b) Volcano plot of 16,456 genes detected by the array for the comparison of occluded versus open olfactory bulb in UNO mice. Transcript abundance (\log_2) is plotted on the abscissa. Statistical significance (\log_{10}) is plotted on the ordinate. Genes shown in red meet a 2-fold and $P < 0.01$ criterion. Note that there are more upregulated genes (+) than downregulated genes (-) on occluded side.

more ipsilateral than contralateral mitral cells respond to a given odorant. Collectively, these studies reveal a previously unknown compensatory response, namely, that primary and secondary olfactory synapses are strengthened ipsilaterally after UNO. Such strengthening of primary and secondary synapses following deprivation is also hard to square with a Hebbian process being more consistent with the notion of homeostatic plasticity [98, 99].

What about the most peripheral components of olfaction—the OSNs? One clue that compensatory processes may exist in these sentinels of smell came from the still unexplained observation that olfactory marker protein (OMP) immunolabeling is more intense in the ipsilateral than contralateral (or control) mucosa of mice subjected to UNO ([100]; Figure 1(d)). Given that *less* stimulation led to *more* OMP in this study, some sort of compensatory process was suggested, especially given this protein's suspected function in olfactory transduction [101]. In a series of follow-up experiments, it was shown that adenylate cyclase type III (AC_{III}), a major component of the olfactory transducing cascade, and a nonciliary phosphodiesterase, which has been shown to be involved in transducing modulation, were also upregulated in OSNs in response to nostril occlusion [102]. At least for the AC_{III} result the implication was clear: decreased olfactory stimulation leads to an increase in this enzyme whose product cAMP ultimately causes OSNs to reach threshold for action potential initiation. Thus, stimulus deprivation could be setting in motion a biochemical cascade leading to an increase in “gain” in the OSN transducing cascade (Figure 2). Microarray analysis has recently been used to confirm and extend the previously findings based on immunolabeling [92]. The transcriptomes of adult olfactory mucosa from control mice were compared to those from the ipsilateral and contralateral sides of mice subjected to UNO as neonates. Transcripts of key genes involved in olfactory reception, transduction, and transmission including many olfactory receptors, the olfactory G-protein, the olfactory cyclic nucleotide gated channel, the olfactory calcium-activated chloride channel, and AC_{III}, were upregulated in deprived-side olfactory mucosa, with opposite effects in nondeprived-side mucosa, compared to controls. Thus, these microarray results support the hypothesis that the odor environment can trigger a previously unknown homeostatic control mechanism in OSNs.

Of course, if these observations at the gene and protein level have any functional significance, they should be measurable electrophysiologically. To address this issue EOG recordings were collected from matched locations on the olfactory mucosa from the ipsilateral and contralateral nasal cavity of UNO mice [103]. The stimulus set included a log-dilutions series for a number of odorants common in olfactory research. Consistent with the gene and protein data, EOG amplitudes from recording sites on the deprived mucosa of UNO mice were greater for a given odorant and concentration of stimulus than those from the open side. For some subjects the magnitude of the EOG on the occluded side was as much as double that on the open side. Given that the EOG is thought to be derived from the summed generator potential of OSNs in the vicinity of the recording

electrode, these results imply that OSNs on the occluded side have larger generator potentials or that more cells are recruited by a given odor, or both. These electrophysiological results, as for the protein and RNA data, are consistent with the hypothesis that OSNs respond to deprivation in a compensatory manner. This process would seemingly oppose any Hebbian pruning of synaptic connections in the bulb.

However, the fact still remains that UNO causes reduction in the survival of proliferating granule cells and other interneurons in the bulb [19, 30]! Given that granule cells are inhibitory on mitral cells, the major output neurons in the bulb, and are thought to participate in lateral inhibition that may sharpen odor discrimination, it is tempting to suggest that a homeostatic process in the olfactory circuit underlies their loss after UNO. Whatever else the function of the persistent supply of granule cells to the bulb (see the aforementioned part) their decline with deprivation would allow the system to increase odor detection, perhaps at the price of odor discrimination [40]. It is interesting in light of these considerations that mice whose bulbs are infused with a drug that limits neurogenesis have surprisingly normal olfactory capabilities [104]. Moreover, as noted previously, mice forced to rely only on their occluded olfactory system by removing their contralateral bulb perform better in behavioral tests of detection than control mice [89].

5. Residual Puzzles

Some of the outwardly conflicting results of UNO on the olfactory system discussed so far in this paper may not turn out to be incompatible. Hopefully additional research will make clear what now seems inconsistent. Nevertheless, there may be worth noting some additional lines of evidence concerning the effects of odor experience on the olfactory system that must be considered in any comprehensive theory of olfactory plasticity.

5.1. Olfactory Induction. The specific anosmia to androstenedione, which occurs in roughly half of the human population, can be reversed upon repeated exposure to this odorous steroid [105]. This result has been replicated in certain strains of mice, as has induced sensitivity to certain other odorants [106]. Based on EOG recordings, the locus of this effect has been shown to be the olfactory mucosa, at least in part [106, 107]. Consistent with these results, rats trained in an odorant detection task showed heightened responses and altered mucosal response patterns to the trained odors compared to those in age-matched controls [108]. Perhaps most surprisingly such olfactory induction can occur *trans-uteri*, as rabbits whose mothers have been fed juniper berries show heightened EOG responses to juniper odor postnatally [109]. No mechanism for olfactory induction has been established but the previously discussed evidence that odor exposure, in bulbectomized mice, rescues OSNs from the apoptosis that usually accompanies this manipulation may be pertinent [84]. Perhaps odor exposure changes the population make-up of OSN types in the olfactory mucosa.

5.2. Olfactory Perceptual Learning. This is a phenomenon that in some ways is reminiscent of olfactory induction (reviewed by [110]). After a period of passive exposure to certain binary mixtures of pure odorants, rats begin to discriminate components that had not been discriminated prior to the exposure period [111]. However, unlike induction, this phenomenon is thought to have a bulbar origin because blocking neurogenesis in the bulb with drug infusion before and during the odorant exposure period prevents the improvement in discrimination [111].

5.3. Compensation Redux. From an evolutionary perspective the compensatory processes discussed previously, which appear to be implemented at various levels of the olfactory system, seem quite logical. Given their finite dynamic range, nature has designed sufficient plasticity into sensory systems to continuously adjust their output to maximize the useful information transferred by them about the environment to the brain [112]. This is why sensory systems modulate in order to report *changes* in the environment rather than static levels of a stimulus [113]. Adaptation is a short-term example of this mechanism that has been examined extensively, both empirically and theoretically, in many sensory systems (e.g., [114]). The effects of longer-term deprivation on the olfactory system, such as those seen following UNO, can be understood in the same light as adaptation, though their cellular mechanisms, time course, and reversibility may be quite different. From this viewpoint, animals exposed to “noisy” or “enriched” odor environments might be expected to show changes opposite to those reported for the deprived state. This is exactly the kind of push-pull mechanism that was seen in the microarray studies discussed previously, with control transcript levels intermediate between ipsilateral and contralateral values for the most important olfactory transducing elements [92]. Consistent with such a compensation mechanism, a recent study has shown that transgenic mice with a gene-targeted potassium-channel deletion that renders mitral cells hyperexcitable actually lose many OSNs [93].

Olfactory induction and perceptual learning are more difficult to understand from an ethological viewpoint. Absent any behavioral relevance, why should the nervous system ramp up detection and discrimination of odors that it is merely exposed to that may have no survival value whatsoever? In any event, the empirical results showing that UNO causes upregulation of olfactory transducing elements ipsilaterally (deprived) and downregulation contralaterally (enriched) appear, at least on their face, to be at odds with the prediction of olfactory induction and perceptual learning. The preserved olfactory competence measured at the behavioral level seen in animals with long-term UNO coupled with contralateral bulb ablation also seems inconsistent with these predictions [89]. Notably, human subjects that are chronically exposed to a particular odor for a few weeks show *increases* in threshold that are odor specific, another finding that seems incompatible with the phenomena of induction and olfactory perceptual learning but is completely compatible with the predictions of compensation [115].

As a heuristic exercise, the predicted effects of deprivation or enrichment on the olfactory system at the levels of the

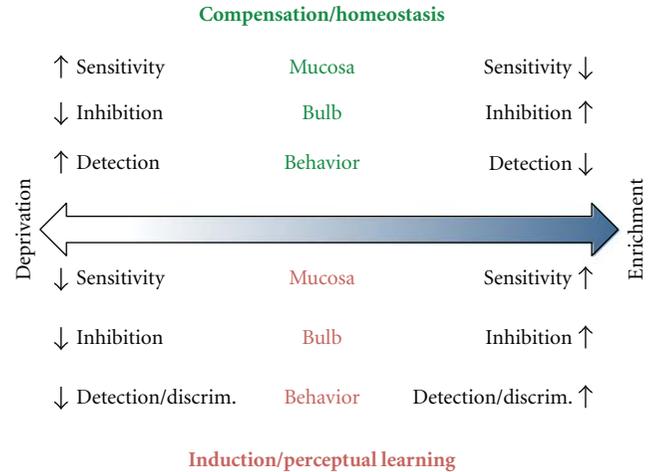


FIGURE 4: Diagram of the predicted effects of odor deprivation (left) and enrichment (right) according to the compensation/homeostasis hypothesis and induction/perceptual learning hypothesis on the olfactory mucosa, bulb, and behavior. Compensation makes no specific prediction about odor discrimination but this may necessarily be opposite to the predicted effects on detection based on bulbar neural circuits. Induction predicts greater detection under enrichment. Perceptual learning predicts greater discrimination under enrichment (see text for further explanation).

mucosa, bulb, and behavior can be contrasted for the induction, perceptual learning, and compensation paradigms (Figure 4). Notably, in some circumstances the predicted effects of these processes are congruent and in others they are in conflict.

6. Conclusions

Gudden, mentor of Emil Kraepelin and Franz Nissl among other notables, was arguably the very first neurobiologist. He pioneered the technique, which still bears his name, of using secondary degeneration to study interrelationships between cortical and subcortical structures and as a psychiatrist he helped humanize the treatment of the insane [3]. Ironically, he most certainly died at the hands of his psychiatric patient “mad King Ludwig II” of Bavaria, though the details of their simultaneous drowning are shrouded in mystery. It is interesting to ponder what Gudden would think of the progress that has been made in the intervening century since he invented his unilateral deprivation techniques. He might not be surprised to find out that the role sensory activity plays in the development and maintenance of the nervous system turns out to be an incredibly intractable problem. For example, some complex computed sensory modules, like orientation maps in visual cortex, develop normally without the benefit of sensory input, while other modules that coexist in the same cortical volume, such as visual direction domains, have an absolute requirement for visual experience [104, 116, 117].

The UNO technique will undoubtedly remain an indispensable tool in the armamentarium of olfactory neuroscientists despite the shortcomings of the procedure and the

conflicting results and hypotheses it has engendered. Key among the matters remaining to be resolved is the correct theoretical framework in which to understand the effects of deprivation on olfaction. Romantics will continue to debate the influence of a lover's presence or absence on the ardor of the human heart. Likewise for the olfactory system we would like to know if absence make the nose grow fonder? Or is it a case of out of smell, out of mind?

Acknowledgments

The author is grateful for research support from the National Science Foundation, Jeffress Memorial Trust, Chenery Endowment, and Craigie Endowment. He also would like to express his appreciation to James Cherry and the Boston University Department of Psychology for their hospitality and provision of office space during the writing of this paper.

References

- [1] R. Held, Y. Ostrovsky, B. Degelder et al., "The newly sighted fail to match seen with felt," *Nature Neuroscience*, vol. 14, no. 5, pp. 551–553, 2011.
- [2] C. Darwin, *The Variation of Animals and Plants Under Domestication*, vol. 1, John Murray, London, UK, 1868.
- [3] L. Sarikcioglu, "Johann Bernhard Aloys von Gudden: an outstanding scientist," *Journal of Neurology, Neurosurgery & Psychiatry*, vol. 78, no. 2, p. 195, 2007.
- [4] Gudden, "Experimentaluntersuchungen über das peripherische und centrale Nervensystem," *Archiv für Psychiatrie und Nervenkrankheiten*, vol. 2, no. 3, pp. 693–723, 1870.
- [5] T. N. Wiesel, "Postnatal development of the visual cortex and the influence of environment," *Nature*, vol. 299, no. 5884, pp. 583–591, 1982.
- [6] P. C. Brunjes, "Unilateral naris closure and olfactory system development," *Brain Research Reviews*, vol. 19, no. 1, pp. 146–160, 1994.
- [7] G. V. Ronnett and C. Moon, "G proteins and olfactory signal transduction," *Annual Review of Physiology*, vol. 64, pp. 189–222, 2002.
- [8] T. Hengl, H. Kaneko, K. Dauner, K. Vocke, S. Frings, and F. Möhrlein, "Molecular components of signal amplification in olfactory sensory cilia," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 107, no. 13, pp. 6052–6057, 2010.
- [9] D. J. Zou, A. Chesler, and S. Firestein, "How the olfactory bulb got its glomeruli: a just so story?" *Nature Reviews Neuroscience*, vol. 10, no. 8, pp. 611–618, 2009.
- [10] T. E. Benson, D. K. Ryugo, and J. W. Hinds, "Effects of sensory deprivation on the developing mouse olfactory system: a light and electron microscopic, morphometric analysis," *Journal of Neuroscience*, vol. 4, no. 3, pp. 638–653, 1984.
- [11] D. M. Cummings, B. R. Knab, and P. C. Brunjes, "Effects of unilateral olfactory deprivation in the developing opossum, *Monodelphis domestica*," *Journal of Neurobiology*, vol. 33, no. 4, pp. 429–438, 1997.
- [12] E. Meisami, "Effects of olfactory deprivation on postnatal growth of the rat olfactory bulb utilizing a new method for production of neonatal unilateral anosmia," *Brain Research*, vol. 107, no. 2, pp. 437–444, 1976.
- [13] E. Meisami and L. Safari, "A quantitative study of the effects of early unilateral olfactory deprivation on the number and distribution of mitral and tufted cells and of glomeruli in the rat olfactory bulb," *Brain Research*, vol. 221, no. 1, pp. 81–107, 1981.
- [14] L. L. Frazier and P. C. Brunjes, "Unilateral odor deprivation: early postnatal changes in olfactory bulb cell density and number," *Journal of Comparative Neurology*, vol. 269, no. 3, pp. 355–370, 1988.
- [15] D. M. Cummings and L. Belluscio, "Continuous neural plasticity in the olfactory intrabulbar circuitry," *Journal of Neuroscience*, vol. 30, no. 27, pp. 9172–9180, 2010.
- [16] A. M. Oliva, K. R. Jones, and D. Restrepo, "Sensory-dependent asymmetry for a urine-responsive olfactory bulb glomerulus," *Journal of Comparative Neurology*, vol. 510, no. 5, pp. 475–483, 2008.
- [17] A. M. Oliva, E. Salcedo, J. L. Hellier et al., "Toward a mouse neuroethology in the laboratory environment," *PLoS ONE*, vol. 5, no. 6, Article ID e11359, 2010.
- [18] L. Frazier-Cierpial and P. C. Brunjes, "Early postnatal cellular proliferation and survival in the olfactory bulb and rostral migratory stream of normal and unilaterally odor-deprived rats," *Journal of Comparative Neurology*, vol. 289, no. 3, pp. 481–492, 1989.
- [19] A. Saghatelian, P. Roux, M. Migliore et al., "Activity-dependent adjustments of the inhibitory network in the olfactory bulb following early postnatal deprivation," *Neuron*, vol. 46, no. 1, pp. 103–116, 2005.
- [20] D. L. Korol and P. C. Brunjes, "Rapid changes in 2-deoxyglucose uptake and amino acid incorporation following unilateral odor deprivation: a laminar analysis," *Developmental Brain Research*, vol. 52, no. 1-2, pp. 75–84, 1990.
- [21] P. E. Pedersen, G. M. Shephard, and C. A. Greer, "Cytochrome oxidase staining in the olfactory epithelium and bulb of normal and odor-deprived neonatal rats," *Annals of the New York Academy of Sciences*, vol. 510, pp. 544–546, 1987.
- [22] E. Meisami and R. Mousavi, "Lasting effects of early olfactory deprivation on the growth, DNA, RNA and protein content, and Na-K-ATPase and AChE activity of the rat olfactory bulb," *Developmental Brain Research*, vol. 2, no. 2, pp. 217–229, 1981.
- [23] H. Baker, "Unilateral, neonatal olfactory deprivation alters tyrosine hydroxylase expression but not aromatic amino acid decarboxylase or GABA immunoreactivity," *Neuroscience*, vol. 36, no. 3, pp. 761–771, 1990.
- [24] H. Baker, K. Morel, D. M. Stone, and J. A. Maruniak, "Adult naris closure profoundly reduces tyrosine hydroxylase expression in mouse olfactory bulb," *Brain Research*, vol. 614, no. 1-2, pp. 109–116, 1993.
- [25] T. Kosaka, K. Kosaka, and K. Hama, "Differential effect of functional olfactory deprivation on the GABAergic and catecholaminergic traits in the rat main olfactory bulb," *Brain Research*, vol. 413, no. 1, pp. 197–203, 1987.
- [26] H. Baker, T. Kawano, F. L. Margolis, and T. H. Joh, "Transneuronal regulation of tyrosine hydroxylase expression in olfactory bulb of mouse and rat," *Journal of Neuroscience*, vol. 3, no. 1, pp. 69–78, 1983.
- [27] N. S. Nadi, R. Head, M. Grillo, J. Hempstead, N. Grannot-Reisfeld, and F. L. Margolis, "Chemical deafferentation of the olfactory bulb: plasticity of the levels of tyrosine hydroxylase, dopamine and norepinephrine," *Brain Research*, vol. 213, no. 2, pp. 365–377, 1981.

- [28] C. C. Woo and M. Leon, "Distribution and development of beta-adrenergic receptors in the rat olfactory bulb," *Journal of Comparative Neurology*, vol. 352, no. 1, pp. 1–10, 1995.
- [29] P. C. Brunjes, L. K. Smith-Crafts, and R. McCarty, "Unilateral odor deprivation: effects on the development of olfactory bulb catecholamines and behavior," *Brain Research*, vol. 354, no. 1, pp. 1–6, 1985.
- [30] K. A. Hamilton and D. M. Coppola, "Distribution of GluR1 is altered in the olfactory bulb following neonatal naris occlusion," *Journal of Neurobiology*, vol. 54, no. 2, pp. 326–336, 2003.
- [31] F. Gomez-Pinilla, K. M. Guthrie, M. Leon, and M. Nieto-Sampedro, "NGF receptor increase in the olfactory bulb of the rat after early odor deprivation," *Developmental Brain Research*, vol. 48, no. 2, pp. 161–165, 1989.
- [32] J. H. McLean, A. Darby-King, and W. S. Bonnell, "Neonatal olfactory sensory deprivation decreases BDNF in the olfactory bulb of the rat," *Developmental Brain Research*, vol. 128, no. 1, pp. 17–24, 2001.
- [33] K. C. Biju, T. G. Mast, and D. A. Fadool, "Olfactory sensory deprivation increases the number of proBDNF-immunoreactive mitral cells in the olfactory bulb of mice," *Neuroscience Letters*, vol. 447, no. 1, pp. 42–47, 2008.
- [34] D. A. Fadool, K. Tucker, J. J. Phillips, and J. A. Simmen, "Brain insulin receptor causes activity-dependent current suppression in the olfactory bulb through multiple phosphorylation of Kv1.3," *Journal of Neurophysiology*, vol. 83, no. 4, pp. 2332–2348, 2000.
- [35] K. Tucker and D. A. Fadool, "Neurotrophin modulation of voltage-gated potassium channels in rat through TrkB receptors is time and sensory experience dependent," *Journal of Physiology*, vol. 542, no. 2, pp. 413–429, 2002.
- [36] J. M. Mirich, K. R. Illig, and P. C. Brunjes, "Experience-dependent activation of Extracellular Signal-Related Kinase (ERK) in the olfactory bulb," *Journal of Comparative Neurology*, vol. 479, no. 2, pp. 234–241, 2004.
- [37] D. A. Wilson, K. M. Guthrie, and M. Leon, "Modification of olfactory bulb synaptic inhibition by early unilateral olfactory deprivation," *Neuroscience Letters*, vol. 116, no. 3, pp. 250–256, 1990.
- [38] D. A. Wilson and J. G. Wood, "Functional consequences of unilateral olfactory deprivation: time-course and age sensitivity," *Neuroscience*, vol. 49, no. 1, pp. 183–192, 1992.
- [39] D. A. Wilson, "NMDA receptors mediate expression of one form of functional plasticity induced by olfactory deprivation," *Brain Research*, vol. 677, no. 2, pp. 238–242, 1995.
- [40] D. A. Wilson and R. M. Sullivan, "The D2 antagonist spiperone mimics the effects of olfactory deprivation on mitral/tufted cell odor response patterns," *Journal of Neuroscience*, vol. 15, no. 8, pp. 5574–5581, 1995.
- [41] B. D. Philpot, T. C. Foster, and P. C. Brunjes, "Mitral/tufted cell activity is attenuated and becomes uncoupled from respiration following naris closure," *Journal of Neurobiology*, vol. 33, no. 4, pp. 374–386, 1997.
- [42] B. J. Maher, M. J. McGinley, and G. L. Westbrook, "Experience-dependent maturation of the glomerular microcircuit," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 39, pp. 16865–16870, 2009.
- [43] K. M. Guthrie, D. A. Wilson, and M. Leon, "Early unilateral deprivation modifies olfactory bulb function," *Journal of Neuroscience*, vol. 10, no. 10, pp. 3402–3412, 1990.
- [44] P. C. Brunjes, A. O. Caggiano, D. L. Korol, and J. S. Stewart, "Unilateral olfactory deprivation: effects on succinate dehydrogenase histochemistry and [3H]leucine incorporation in the olfactory mucosa," *Developmental Brain Research*, vol. 62, no. 2, pp. 239–244, 1991.
- [45] A. I. Farbman, P. C. Brunjes, L. Rentfro, J. Michas, and S. Ritz, "The effect of unilateral naris occlusion on cell dynamics in the developing rat olfactory epithelium," *Journal of Neuroscience*, vol. 8, no. 9, pp. 3290–3295, 1988.
- [46] B. Stahl, H. Distel, and R. Hudson, "Effects of reversible nare occlusion on the development of the olfactory epithelium in the rabbit nasal septum," *Cell and Tissue Research*, vol. 259, no. 2, pp. 275–281, 1990.
- [47] D. M. Cummings and P. C. Brunjes, "Changes in cell proliferation in the developing olfactory epithelium following neonatal unilateral naris occlusion," *Experimental Neurology*, vol. 128, no. 1, pp. 124–128, 1994.
- [48] J. M. Mirich and P. C. Brunjes, "Activity modulates neuronal proliferation in the developing olfactory epithelium," *Developmental Brain Research*, vol. 127, no. 1, pp. 77–80, 2001.
- [49] M. A. Cavallin, K. Powell, K. C. Biju, and D. A. Fadool, "State-dependent sculpting of olfactory sensory neurons is attributed to sensory enrichment, odor deprivation, and aging," *Neuroscience Letters*, vol. 483, no. 2, pp. 90–95, 2010.
- [50] J. A. Cámara, M. Garrosa, and M. J. Gayoso, "Histological changes in rat nasal epithelia after unilateral neonatal naris occlusion," *Microscopy Research and Technique*, vol. 69, no. 7, pp. 585–594, 2006.
- [51] E. Weiler, J. Seeger, J. Cherry, and D. Coppola, *The Effect of Perinatal Naris Occlusion on Nasal Turbinate Development: Does Emmetropization Occur in the Nose?* European Chemoreception Research Organization, Manchester UK, 2011.
- [52] T. Kaneko-Goto, S. I. Yoshihara, H. Miyazaki, and Y. Yoshihara, "BIG-2 Mediates Olfactory Axon Convergence to Target Glomeruli," *Neuron*, vol. 57, no. 6, pp. 834–846, 2008.
- [53] H. Oztokatli, M. Hörnberg, A. Berghard, and S. Bohm, "Retinoic acid receptor and CNGA2 channel signaling are part of a regulatory feedback loop controlling axonal convergence and survival of olfactory sensory neurons," *The FASEB Journal*, vol. 26, no. 2, pp. 617–627, 2012.
- [54] E. O. Williams, H. M. Sickles, A. L. Dooley, S. Palumbos, A. J. Bisogni, and D. M. Lin, "Delta Protocadherin 10 is regulated by activity in the mouse main olfactory system," *Frontiers in Neural Circuits*. In press.
- [55] D. A. Wilson, A. R. Best, and P. C. Brunjes, "Trans-neuronal modification of anterior piriform cortical circuitry in the rat," *Brain Research*, vol. 853, no. 2, pp. 317–322, 2000.
- [56] H. H. Kim, A. C. Puche, and F. L. Margolis, "Odorant deprivation reversibly modulates transsynaptic changes in the NR2B-mediated CREB pathway in mouse piriform cortex," *Journal of Neuroscience*, vol. 26, no. 37, pp. 9548–9559, 2006.
- [57] K. M. Franks and J. S. Isaacson, "Synapse-specific downregulation of NMDA receptors by early experience: a critical period for plasticity of sensory input to olfactory cortex," *Neuron*, vol. 47, no. 1, pp. 101–114, 2005.
- [58] A. R. Best and D. A. Wilson, "A postnatal sensitive period for plasticity of cortical afferents but not cortical association fibers in rat piriform cortex," *Brain Research*, vol. 961, no. 1, pp. 81–87, 2003.
- [59] J. L. Brown and P. C. Brunjes, "Development of the anterior olfactory nucleus in normal and unilaterally odor deprived

- rats," *Journal of Comparative Neurology*, vol. 301, no. 1, pp. 15–22, 1990.
- [60] D. M. Yousem, R. J. Geckle, W. B. Bilker, H. Kroger, and R. L. Doty, "Posttraumatic smell loss: relationship of psychophysical tests and volumes of the olfactory bulbs and tracts and the temporal lobes," *Academic Radiology*, vol. 6, no. 5, pp. 264–272, 1999.
- [61] D. Buschhüter, M. Smitka, S. Puschmann et al., "Correlation between olfactory bulb volume and olfactory function," *NeuroImage*, vol. 42, no. 2, pp. 498–502, 2008.
- [62] T. Hummel, M. Smitka, S. Puschmann, J. C. Gerber, B. Schaal, and D. Buschhüter, "Correlations between olfactory bulb volume and olfactory function in children and adolescents," *Experimental Brain Research*, vol. 214, pp. 285–291, 2011.
- [63] P. Rombaux, H. Potier, B. Bertrand, T. Duprez, and T. Hummel, "Olfactory bulb volume in patients with sinonasal disease," *American Journal of Rhinology*, vol. 22, no. 6, pp. 598–601, 2008.
- [64] V. Gudziol, D. Buschhüter, N. Abolmaali, J. Gerber, P. Rombaux, and T. Hummel, "Increasing olfactory bulb volume due to treatment of chronic rhinosinusitis—a longitudinal study," *Brain*, vol. 132, no. 11, pp. 3096–3101, 2009.
- [65] K. Wu, J. D. Howard, K. M. Phillips, D. Conley, and J. A. Gottfried, *Odor Deprivation in the Human Brain*, Society for Neuroscience Abstract, San Diego, Calif, USA, 2010.
- [66] L. C. Katz and C. J. Shatz, "Synaptic activity and the construction of cortical circuits," *Science*, vol. 274, no. 5290, pp. 1133–1138, 1996.
- [67] L. C. Katz and M. Rubin, *Keep your Brain Alive*, Workman Publishing, New York, NY, USA, 1999.
- [68] N. W. Daw, *Visual Development*, Springer, New York, NY, USA, 2006.
- [69] C. R. Yu, J. Power, G. Barnea et al., "Spontaneous neural activity is required for the establishment and maintenance of the olfactory sensory map," *Neuron*, vol. 42, no. 4, pp. 553–566, 2004.
- [70] P. C. Brunjes and M. J. Borror, "Unilateral odor deprivation: differential effects due to time of treatment," *Brain Research Bulletin*, vol. 11, no. 5, pp. 501–503, 1983.
- [71] J. R. Henegar and J. A. Maruniak, "Quantification of the effects of long-term unilateral naris closure on the olfactory bulbs of adult mice," *Brain Research*, vol. 568, no. 1-2, pp. 230–234, 1991.
- [72] N. Mandairon, J. Sacquet, F. Jourdan, and A. Didier, "Long-term fate and distribution of newborn cells in the adult mouse olfactory bulb: influences of olfactory deprivation," *Neuroscience*, vol. 141, no. 1, pp. 443–451, 2006.
- [73] X. M. Zhang, K. Xiong, Y. Cai et al., "Functional deprivation promotes amyloid plaque pathogenesis in Tg2576 mouse olfactory bulb and piriform cortex," *European Journal of Neuroscience*, vol. 31, no. 4, pp. 710–721, 2010.
- [74] J. E. Dahlen, D. A. Jimenez, R. C. Gerkin, and N. N. Urban, "Morphological analysis of activity-reduced adult-born neurons in the mouse olfactory bulb," *Frontiers in Neural Circuits*. In press.
- [75] C. H. W. Leung and D. A. Wilson, "Trans-neuronal regulation of cortical apoptosis in the adult rat olfactory system," *Brain Research*, vol. 984, no. 1-2, pp. 182–188, 2003.
- [76] D. M. Cummings, H. E. Henning, and P. C. Brunjes, "Olfactory bulb recovery after early sensory deprivation," *Journal of Neuroscience*, vol. 17, no. 19, pp. 7433–7440, 1997.
- [77] K. Fox and R. O. L. Wong, "A comparison of experience-dependent plasticity in the visual and somatosensory systems," *Neuron*, vol. 48, no. 3, pp. 465–477, 2005.
- [78] K. K. Gokoffski, S. Kawachi, H. H. Wu et al., "Feedback regulation of neurogenesis in the mammalian olfactory epithelium: new insights from genetics and systems biology," in *The Neurobiology of Olfaction*, A. Menini, Ed., chapter 10, CRC Press, Boca Raton, Fla, USA, 2010.
- [79] F. Lazarini and P. M. Lledo, "Is adult neurogenesis essential for olfaction?" *Trends in Neurosciences*, vol. 34, no. 1, pp. 20–30, 2011.
- [80] P. M. Lledo and A. Saghatelian, "Integrating new neurons into the adult olfactory bulb: joining the network, life-death decisions, and the effects of sensory experience," *Trends in Neurosciences*, vol. 28, no. 5, pp. 248–254, 2005.
- [81] N. Sanai, T. Nguyen, R. A. Ihrie et al., "Corridors of migrating neurons in the human brain and their decline during infancy," *Nature*, vol. 478, no. 7369, pp. 382–386, 2011.
- [82] D. Krestel, D. Passe, J. C. Smith, and L. Jonsson, "Behavioral determination of olfactory thresholds to amyl acetate in dogs," *Neuroscience and Biobehavioral Reviews*, vol. 8, no. 2, pp. 169–174, 1984.
- [83] J. W. Hinds, P. L. Hinds, and N. A. McNelly, "An autoradiographic study of the mouse olfactory epithelium: evidence for long-lived receptors," *Anatomical Record*, vol. 210, no. 2, pp. 375–383, 1984.
- [84] W. C. Watt, H. Sakano, Z. Y. Lee, J. E. Reusch, K. Trinh, and D. R. Storm, "Odorant stimulation enhances survival of olfactory sensory neurons via MAPK and CREB," *Neuron*, vol. 41, no. 6, pp. 955–967, 2004.
- [85] M. N. Batterton, D. Robarts, S. K. Woodley, and M. J. Baum, "Comparison of odor and mating-induced glomerular activation in the main olfactory bulb of estrous female ferrets," *Neuroscience Letters*, vol. 400, no. 3, pp. 224–229, 2006.
- [86] B. M. Slotnick and A. J. Pazos, "Rats with one olfactory bulb removed and the contralateral naris closed can detect odors," *Physiology & Behavior*, vol. 48, no. 1, pp. 37–40, 1990.
- [87] N. L. Hunt and B. M. Slotnick, "Functional capacity of the rat olfactory bulb after neonatal naris occlusion," *Chemical Senses*, vol. 16, no. 2, pp. 131–142, 1991.
- [88] D. M. Coppola, J. A. Coltrane, and I. Arsov, "Retronasal or internasal olfaction can mediate odor-guided behaviors in newborn mice," *Physiology & Behavior*, vol. 56, no. 4, pp. 729–736, 1994.
- [89] C. J. Angely and D. M. Coppola, "How does long-term odor deprivation affect the olfactory capacity of adult mice?" *Behavioral and Brain Functions*, vol. 6, article 26, pp. i–ij, 2010.
- [90] X. Grosmaître, L. C. Santarelli, J. Tan, M. Luo, and M. Ma, "Dual functions of mammalian olfactory sensory neurons as odor detectors and mechanical sensors," *Nature Neuroscience*, vol. 10, no. 3, pp. 348–354, 2007.
- [91] M. L. Schaefer, B. Böttger, W. L. Silver, and T. E. Finger, "Trigeminal collaterals in the nasal epithelium and olfactory bulb: a potential route for direct modulation of olfactory information by trigeminal stimuli," *Journal of Comparative Neurology*, vol. 444, no. 3, pp. 221–226, 2002.
- [92] D. M. Coppola and C. Waggner, "The effects of unilateral naris occlusion on gene expression profiles in mouse olfactory mucosa," *Journal of Molecular Neuroscience*.
- [93] J. A. Maruniak, P. J. Lin, and J. R. Henegar, "Effects of unilateral naris closure on the olfactory epithelia of adult mice," *Brain Research*, vol. 490, no. 2, pp. 212–218, 1989.

- [94] J. A. Maruniak, J. R. Henegar, and T. P. Sweeney, "Effects of long-term unilateral naris closure on the olfactory epithelia of adult mice," *Brain Research*, vol. 526, no. 1, pp. 65–72, 1990.
- [95] M. Tos and C. Mogensen, "Changes of the nasal mucosa in altered airflow illustrated by blind quantitative histology," *Journal of Laryngology and Otology*, vol. 92, no. 8, pp. 667–681, 1978.
- [96] M. Leon, "Compensatory responses to early olfactory restriction," *Annals of the New York Academy of Sciences*, vol. 855, pp. 104–108, 1998.
- [97] K. M. Guthrie, J. M. Pullara, J. F. Marshall, and M. Leon, "Olfactory deprivation increases dopamine D2 receptor density in the rat olfactory bulb," *Synapse*, vol. 8, no. 1, pp. 61–70, 1991.
- [98] W. J. Tyler, G. C. Petzold, S. K. Pal, and V. N. Murthy, "Experience-dependent modification of primary sensory synapses in the mammalian olfactory bulb," *Journal of Neuroscience*, vol. 27, no. 35, pp. 9427–9438, 2007.
- [99] G. G. Turrigiano and S. B. Nelson, "Homeostatic plasticity in the developing nervous system," *Nature Reviews Neuroscience*, vol. 5, no. 2, pp. 97–107, 2004.
- [100] A. Waguespack, M. Reems, M. L. Butman, J. Cherry, and D. M. Coppola, "Olfactory receptor neurons have enhanced olfactory marker protein immunoreactivity in the nasal cavity ipsilateral to naris occlusion," *Brain Research*, vol. 1044, no. 1, pp. 1–7, 2005.
- [101] S. L. Youngentob and F. L. Margolis, "OMP gene deletion causes an elevation in behavioral threshold sensitivity," *NeuroReport*, vol. 10, no. 1, pp. 15–19, 1999.
- [102] D. M. Coppola, A. M. Waguespack, M. R. Reems, M. L. Butman, and J. A. Cherry, "Naris occlusion alters transducing protein immunoreactivity in olfactory epithelium," *Histology and Histopathology*, vol. 21, no. 4–6, pp. 487–501, 2006.
- [103] C. T. Waggener and D. M. Coppola, "Naris occlusion alters the electro-olfactogram: evidence for compensatory plasticity in the olfactory system," *Neuroscience Letters*, vol. 427, no. 2, pp. 112–116, 2007.
- [104] A. Mouret, G. Lepousez, J. Gras, M. M. Gabellec, and P. M. Lledo, "Turnover of newborn olfactory bulb neurons optimizes olfaction," *Journal of Neuroscience*, vol. 29, no. 39, pp. 12302–12314, 2009.
- [105] C. J. Wysocki, K. M. Dorries, and G. K. Beachamp, "Ability to perceive androstenone can be acquired by ostensibly anosmic people," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 86, no. 20, pp. 7976–7978, 1989.
- [106] H. W. Wang, C. J. Wysocki, and G. H. Gold, "Induction of olfactory receptor sensitivity in mice," *Science*, vol. 260, no. 5110, pp. 998–1000, 1993.
- [107] K. K. Yee and C. J. Wysocki, "Odorant exposure increases olfactory sensitivity: olfactory epithelium is implicated," *Physiology & Behavior*, vol. 72, no. 5, pp. 705–711, 2001.
- [108] S. L. Youngentob and P. F. Kent, "Enhancement of odorant-induced mucosal activity patterns in rats trained on an odorant identification task," *Brain Research*, vol. 670, no. 1, pp. 82–88, 1995.
- [109] R. Hudson and H. Distel, "Induced peripheral sensitivity in the developing vertebrate olfactory system," *Annals of the New York Academy of Sciences*, vol. 855, pp. 109–115, 1998.
- [110] D. A. Wilson and R. J. Stevenson, "Olfactory perceptual learning: the critical role of memory in odor discrimination," *Neuroscience and Biobehavioral Reviews*, vol. 27, no. 4, pp. 307–328, 2003.
- [111] M. M. Moreno, C. Linster, O. Escanilla, J. Sacquet, A. Didier, and N. Mandairon, "Olfactory perceptual learning requires adult neurogenesis," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 42, pp. 17980–17985, 2009.
- [112] M. Stemmler and C. Koch, "How voltage-dependent conductances can adapt to maximize the information encoded by neuronal firing rate," *Nature Neuroscience*, vol. 2, no. 6, pp. 521–527, 1999.
- [113] D. Coppola and D. Purves, "The extraordinarily rapid disappearance of entoptic images," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 93, no. 15, pp. 8001–8004, 1996.
- [114] S. B. Laughlin, "The role of sensory adaptation in the retina," *Journal of Experimental Biology*, vol. 146, pp. 39–62, 1989.
- [115] P. Dalton and C. J. Wysocki, "The nature and duration of adaptation following long-term odor exposure," *Perception and Psychophysics*, vol. 58, no. 5, pp. 781–792, 1996.
- [116] D. M. Coppola and L. E. White, "Visual experience promotes the isotropic representation of orientation preference," *Visual Neuroscience*, vol. 21, no. 1, pp. 39–51, 2004.
- [117] L. E. White and D. Fitzpatrick, "Vision and cortical map development," *Neuron*, vol. 56, no. 2, pp. 327–338, 2007.