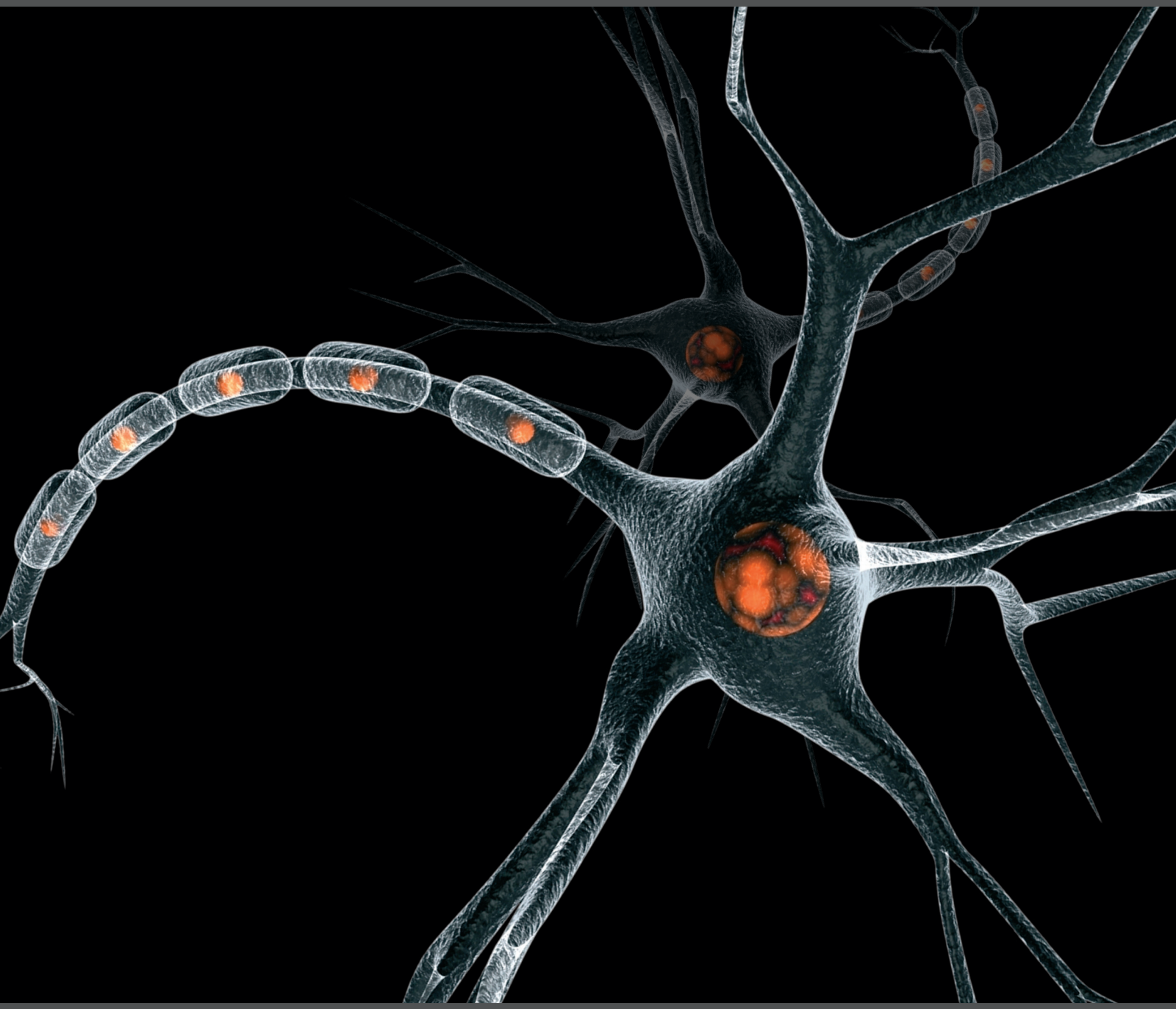
Neural Plasticity

The Role of Dentate Gyrus Neurogenesis in Neuropsychiatric Disorders

Guest Editors: M. Julia García-Fuster, Justin S. Rhodes, and Chitra D. Mandyam



The Role of Dentate Gyrus Neurogenesis in Neuropsychiatric Disorders

Neural Plasticity

The Role of Dentate Gyrus Neurogenesis in Neuropsychiatric Disorders

Guest Editors: M. Julia García-Fuster, Justin S. Rhodes, and Chitra D. Mandyam

Editorial Board

Contents

Hindawi Publishing Corporation
Neural Plasticity
Volume 2013, Article ID 584382, 2 pages
http://dx.doi.org/10.1155/2013/584382

Editorial

The Role of Dentate Gyrus Neurogenesis in Neuropsychiatric Disorders

M. Julia García-Fuster,[1] Justin S. Rhodes,[2] and Chitra D. Mandyam[3]

[1] *University Research Institute on Health Sciences (IUNICS), University of the Balearic Islands, 07122 Palma de Mallorca, Spain*
[2] *Department of Psychology, University of Illinois, Champaign, IL 61820, USA*
[3] *Committee on the Neurobiology of Addictive Disorders, The Scripps Research Institute, La Jolla, CA 92037, USA*

Correspondence should be addressed to Chitra D. Mandyam; cmandyam@scripps.edu

Received 15 May 2013; Accepted 15 May 2013

The special issue reviews the most recent developments in dentate gyrus (DG) neurogenesis with regard to neuropsychiatric disorders. A few laboratories worldwide have contributed to this special issue, and their contributions have showcased the current efforts in understanding the functional significance of DG neurogenesis in depression, psychosis, and addiction. Additionally, a promising therapeutic strategy for treating radiation therapy disorders is reviewed. The review and research articles in this special issue indicate that DG neurogenesis can be conceptualized as a form of plasticity and that the alterations in DG neurogenesis can predict a spectrum of disorders associated with the hippocampus. Highlights from the papers in the special issue are summarized below.

Introduction. The hippocampus is tactically situated in the brain such that it has reciprocal connections to several other regions involved in emotional behaviors, such as stress responses and positive and negative reinforcement. Alterations in these emotional behaviors can be considered precursors for the development of neuropsychiatric disorders, such as depression, schizophrenia, and drug addiction. Adult neurogenesis in the DG of the hippocampus is a new undisputed form of spontaneous plasticity, and most of the emotional behaviors implicated in neuropsychiatric disorders are known to inhibit DG neurogenesis. Therefore, the normalization of impaired DG neurogenesis during recovery or abstinence may be required to initiate normal neuroplasticity in the hippocampus, which could be critical for recovery. Contributions to this special issue include six review papers and one research paper that focus on the role of DG hippocampal neurogenesis in the etiology and treatment of three major neuropsychiatric disorders—depression, psychosis, and addiction—in human or animal models. Each paper was reviewed by at least two reviewers and revised according to the reviewers' comments.

Summary. Major depression is among the most prevalent psychiatric disorders, constituting the first leading cause of years lived with disability. In addition to the well-known monoamine neurotransmitter hypothesis of depression, many parallel processes exist that play a role in depression, such as alterations in neurotrophic factors (e.g., brain-derived neurotrophic factor (BDNF)) and the regulation of adult hippocampal neurogenesis. Three papers in this special issue discuss the neurogenesis hypothesis of depression, which postulates that the generation of new neurons in the adult DG is involved in the etiology and treatment of depression. The paper by H. Jun et al. reviews the literature on the effects of animal models of depression on adult hippocampal neurogenesis. They conclude that decreased neurogenesis is not associated with risk in the development of depressive behavior and speculate that newborn cells may be a major contributor to normalizing or ameliorating the disease state rather than being causally involved in the etiology of depression. The paper reviews possible therapeutic interventions to ameliorate depression symptoms that involve an increase in DG neurogenesis regulation. Nonclassic antidepressant methods (e.g., electroconvulsive therapy and deep brain stimulation) and chronic treatment with

classic antidepressants and exercise are shown to stimulate neurogenesis in the DG. The molecular targets that participate in or mediate the antidepressant-induced increase in DG neurogenesis are also reviewed in this paper, such as the involvement of BDNF and vascular endothelial growth factor (VEGF), neurotrophic factors that regulate the *microniche* of adult neurogenesis. The paper by F. Pilar-Cuéllar et al. reviews the neurotrophic/plasticity hypothesis of depression by discussing basic and clinical studies that ascertained the role of intracellular signaling cascades in the regulation of neural proliferation and plasticity. They focus their review on hippocampal modulation by different types and subtypes of serotonergic receptors and trophic factors (e.g., BDNF and VEGF) through intracellular signaling pathways (e.g., cAMP, Wnt/β-catenin, and mTOR). The paper by S. R. Wainwright and L. A. M. Galea reviews the neural plasticity theory of depression by focusing on the potential role of DG neurogenesis and a cell adhesion molecule, the polysialylated form of neural cell adhesion molecule (PSA-NCAM). PSA-NCAM appears to be an interesting target for future studies because it plays a fundamental role in mediating the broad effects of antidepressant treatment across multiple forms of neural plasticity and appears to function at the confluence of prevailing theories of depression.

Schizophrenia is a complex psychiatric disorder with an onset during early adulthood or adolescence. To clarify the mechanisms that underlie the onset of the illness, clinicians are studying the early phases of psychosis that might transition to schizophrenia spectrum disorder. The literature on neurogenesis-relevant research on postmortem human tissues from schizophrenia patients and animal models shows diminished cell proliferation or altered molecular markers involved in the regulation of neurogenesis. Typical and atypical antipsychotics have been described to differentially regulate cell proliferation/neurogenesis, reflected by their actions, mechanisms, primary effects, and side effects. The paper by G. Keilhoff et al. reviews the effects of typical and atypical antipsychotic drugs on neurogenesis in animal models that reflect neurodevelopmental aspects of schizophrenia. A different approach to the study of the role of DG neurogenesis in schizophrenia is described in the paper by H. Hagihara et al., which reviews the literature that describes a phenomenon defined as an "immature dentate gyrus" (iDG). This phenomenon is characterized by arresting almost all neurons in the DG at a pseudoimmature state at both the molecular and electrophysiological levels and was identified in several strains of genetically engineered mutant mice with abnormal behaviors that are characteristic of neuropsychiatric disorders. It was first discovered in heterozygous knockout mice with a null mutation in a synaptic plasticity molecule (CaMKII) relevant to schizophrenia. The iDG mouse model has been associated with abnormal behaviors related to schizophrenia and other neuropsychiatric disorders, suggesting the importance of the adequate maturation and integration of adult-generated neurons into the hippocampal circuit for normal cognitive and emotional development. Interestingly, iDG-like phenomena have been observed in postmortem brains in patients with schizophrenia and bipolar disorder, and it has been proposed as a potential brain endophenotype that may be useful for classifying these disorders.

Drug addiction is a chronic relapsing disorder characterized by compulsive drug-seeking and -taking behavior and a loss of the ability to control drug intake. Hippocampal plasticity likely plays an important role in maintaining addictive behavior and/or relapse. The role of DG neurogenesis in drug addiction is discussed in two papers in this special issue. The paper by H. Jun et al. reviews selected prototype animal studies that investigated the effects of drug addiction (i.e., cocaine and alcohol) on adult hippocampal neurogenesis. Abused drugs are potent negative regulators of adult DG neurogenesis and as a result may impair cognitive function. The research article by M. P. Hicks et al. investigates the effects of extinction of heroin-seeking behavior on DG immature neurons. The authors show that extinction training following heroin self-administration increases the number of doublecortin-positive immature neurons selectively in the dorsal regions of the DG. They also present evidence that inhibition of cell proliferation in the DG increases responding during extinction, which is indicative of impaired extinction learning. These findings support the hypothesis that immature neurons in the DG may play a functional role in certain learning and memory processes, including the extinction of drug-seeking behavior. Diminished neurogenesis in the DG by drugs of abuse may play a causal role in certain cognitive deficits frequently observed in drug addicts

Hippocampal structure and function have been closely studied with regard to cognitive and mental function. The paper by S. P. Rodgers et al. reviews the role of exercise in ameliorating the suppression of cell proliferation and cognitive impairments observed in adulthood following pediatric radiotherapy. Ionizing radiation has been shown to disrupt brain development and suppress cell proliferation by killing the brain's actively dividing neural stem cells. This disrupted brain plasticity is believed to contribute to cognitive impairments. Therefore, the authors discuss the benefit of exercise as one potential treatment that is able to enhance both cell proliferation and neural plasticity needed for optimal cognitive performance.

Acknowledgment

We hope that this special issue will bring new insights into the underlying role of the DG in neuropsychiatric disorders and attract attention of other researchers in the field. We would like to express our appreciation to all the authors and reviewers for their great support in making this special issue possible.

M. Julia García-Fuster
Justin S. Rhodes
Chitra D. Mandyam

Hindawi Publishing Corporation
Neural Plasticity
Volume 2013, Article ID 537265, 21 pages
http://dx.doi.org/10.1155/2013/537265

Review Article

Neural Plasticity and Proliferation in the Generation of Antidepressant Effects: Hippocampal Implication

Fuencisla Pilar-Cuéllar,[1,2] Rebeca Vidal,[1,2] Alvaro Díaz,[1,2] Elena Castro,[1,2] Severiano dos Anjos,[1,3] Jesús Pascual-Brazo,[1,2,4] Raquel Linge,[1,2] Veronica Vargas,[1,2] Helena Blanco,[1] Beatriz Martínez-Villayandre,[1] Ángel Pazos,[1,2] and Elsa M. Valdizán[1,2,5]

[1] *Departamento de Fisiología y Farmacología, Instituto de Biomedicina y Biotecnología de Cantabria (IBBTEC), Universidad de Cantabria-CSIC-IDICAN, Santander, Cantabria, Spain*
[2] *Centro de Investigación Biomédica en Red de SaludMental (CIBERSAM), Instituto de Salud Carlos III, Santander, Cantabria, Spain*
[3] *Stem Center, Clínica Palmaplanas, Camí dels Reis 308, Palma de Mallorca, Spain*
[4] *The Research Group for Neurobiology and Gene Therapy, KU Leuven, Leuven, Belgium*
[5] *Department of Physiology and Pharmacology, School of Medicine, Cardenal Herrera Oria s/n, University of Cantabria 39011 Santander, Spain*

Correspondence should be addressed to Elsa M. Valdizán; valdizae@unican.es

Received 4 March 2013; Revised 1 May 2013; Accepted 8 May 2013

Academic Editor: Chitra D. Mandyam

It is widely accepted that changes underlying depression and antidepressant-like effects involve not only alterations in the levels of neurotransmitters as monoamines and their receptors in the brain, but also structural and functional changes far beyond. During the last two decades, emerging theories are providing new explanations about the neurobiology of depression and the mechanism of action of antidepressant strategies based on cellular changes at the CNS level. The neurotrophic/plasticity hypothesis of depression, proposed more than a decade ago, is now supported by multiple basic and clinical studies focused on the role of intracellular-signalling cascades that govern neural proliferation and plasticity. Herein, we review the state-of-the-art of the changes in these signalling pathways which appear to underlie both depressive disorders and antidepressant actions. We will especially focus on the hippocampal cellularity and plasticity modulation by serotonin, trophic factors as brain-derived neurotrophic factor (BDNF), and vascular endothelial growth factor (VEGF) through intracellular signalling pathways—cAMP, Wnt/β-catenin, and mTOR. Connecting the classic monoaminergic hypothesis with proliferation/neuroplasticity-related evidence is an appealing and comprehensive attempt for improving our knowledge about the neurobiological events leading to depression and associated to antidepressant therapies.

1. Introduction

Major depressive disorder (MDD) constitutes the first leading cause of years lived with disability [1], and its incidence is on the rise globally. Yet, until recently, little was known about its pathogenesis, as these conditions are not associated with relevant brain alterations or clear animal models for spontaneous recurrent mood episodes.

The clinical phenomenology of major depression implicates brain neurotransmitter systems involved in the regulation of mood, anxiety, fear, reward processing, attention, motivation, stress responses, social interaction, and neurovegetative function [2]. MDD is associated with blunted reactivity to both positively and negatively stimuli [3]; thus, the decline in hedonic responses may be related to generalized affective insensitivity, instead of deficits in the capacity to feel pleasure at the level of basic sensory experience [4]. From the middle of the last century, a great effort has been made to elucidate the brain areas involved in emotion control and in the pathophysiology of mood disorders. Animal and human studies have indicated the involvement of the limbic system—including the hippocampal formation, cingulate gyrus, and

anterior thalamus—the amygdala and different cortical structures as well as the hypothalamus in these processes [5, 6]. These structures are connected in two main networks: the "orbital" and the "medial prefrontal networks." The orbital network appears to function both as a system for integration of multimodal stimuli and as a system for assessment of the value of those stimuli, and, probably, the support of abstract assessment of reward. The medial network is probably more significant for mood disorders [7].

In depressed subjects, the structures of the medial prefrontal network have been shown to contain alterations in gray matter volume, cellular elements, neurophysiological activity, receptor pharmacology, and gene expression. Dysfunction within this system underlies the disturbances in emotional behavior and other cognitive aspects of the major depressive disorder. Treatments for depression, involving pharmacological, neurosurgical, and deep brain stimulation methods, appear to suppress pathological activity within the components of medial prefrontal network such as the subgenual anterior cingulate cortex, ventromedial frontal cortex, striatum, and amygdala [6].

Although the causes of MDD are not yet completely known, genetic factors appear to play an important role although other factors deal with acute or chronic stress, childhood trauma, viral infections, and others [12, 13]. Regarding genetic causes, certain polymorphisms in genes related to the serotonergic system as the serotonin transporter, the brain-derived neurotrophic factor, the monoamine oxidase A, or the tryptophan hydroxylase 1, may increase the risk for depression or the vulnerability to stress [14]. Not all the studies published to date have found gene-environment interactions; however, the combination of both factors seems to predict more accurately a person's risk to suffer from major depressive disorder better than genes or environment alone.

The discovery that some drugs as iproniazid and imipramine exert an antidepressant effect dates back to the 1950s [15]. In 1965, it was shown that these drugs act through the monoaminergic system by increasing the brain levels of those monoamines [16]. These observations led to the development of the classical "monoaminergic hypothesis of depression," which proposes that low monoamine brain levels in depressed individuals are responsible for this pathology. The classic antidepressants that increase monoamine neurotransmitters in the synaptic cleft are generally used for first-line treatment. However, the clinical benefit of these treatments is not immediate, taking 3-4 weeks to obtain a full response. Other therapeutic problems of currently used antidepressant drugs include relapses, drug side effects, incomplete resolution, residual symptoms, and drug resistance.

Traditionally, research in the neurobiology of major depressive disorder has been focused on monoamines. However, several lines of evidence [17] have led to the conclusion that the abnormalities associated to depression go beyond monoaminergic neurotransmission: thus, the development of better antidepressants will surely depend on the discovery and understanding of new cellular targets. In this regard, in the late 90's a new hypothesis has tried to explain major depression based on molecular mechanisms of neuroplasticity [18].

The "neuroplasticity hypothesis" was postulated based on several findings: first, stress decrease hippocampal neurogenesis and synaptic plasticity in prefrontal cortex (PFCx) [19–22]. Moreover, most known antidepressant therapies stimulate the proliferation of hippocampal progenitor cells, which constitutes the first stage of adult hippocampal neurogenesis [23]. However, the contribution of hippocampal neurogenesis to the pathogenesis of depression is far from being fully understood. Second, hippocampal morphologic analyses in depressed patients reveal volume loss and gray matter alterations. While some studies suggest that decreased adult neurogenesis could be responsible for such fluctuating changes, others show that the hippocampal volumetric reductions could be due to changes in neuropil, glial number, and/or dendritic complexity, and not necessarily to a cell proliferation decrease [24]. Third, different neuroplasticity- and proliferation-related intracellular pathways appear to be involved in the antidepressants' action as brain-derived neurotrophic factor (BDNF) [25], β-catenin [10, 26], or the mammalian target of rapamycin (mTOR) [27].

2. Cell Proliferation and Plasticity Role in Mood Disorders and Antidepressant Treatments

Dentate gyrus proliferation is decreased by stress [19–22], and in several animal models of depression as unpredictable stress, chronic administration of corticosterone, olfactory bulbectomy, or maternal deprivation [22, 28–31]. This loss in cell proliferation is correlated with a decreased hippocampal volume [32–34]. Hippocampal proliferation decrease is also observed in other disease models such as diabetic mice, which present a high incidence of depression reported in individuals with that primary illness [35]. In these animals, the reduced hippocampal proliferation is reversed with chronic antidepressant treatments [35]. In animals subjected to acute or chronic stress, a period of at least 24 h or 3 weeks, respectively, is required to get a recovery of the cellular proliferation [29]. However, although all these changes have been extensively studied, major depression is not generally considered as "hippocampal disorder."

It is unlikely that impaired adult hippocampal neurogenesis alone may fully explain the neuropathology of major depression. In this sense, other studies have addressed cellular proliferation in anatomical structures quite relevant to depressive disorders, such as prefrontal cortex and amygdala, by using animal models of depression. Thus, medial frontal cortex presents a reduction in cell proliferation [36], downregulation of genes implicated in cell proliferation [37, 38], decreased cell growth and survival, and apoptosis inhibition [38]. Structures such as amygdala present an opposite pattern, with an increase in neuronal dendrite length in stress models [39]. Chronic administration of antidepressants leads to an increased proliferation in prefrontal cortex [36, 40] although the fate of the new generated cells goes toward the formation of glia rather than neurons, in contrast to hippocampus

[36]. No data are available regarding antidepressant effects on amygdalar cell proliferation; however, this structure has been involved in the negative control of the hippocampal cell survival induced by antidepressant treatments, based on the increased cell survival observed in hippocampus, after the basolateral complex of the amygdale (BLA) lesion [41]. It is interesting to note that amygdala—implicated in fear-related learning that impairs the memory processing of the Hp-PFCx memory—shows an enhancement of LTP under stress situations, which is not reverted by antidepressants. Thus, antidepressants as tianeptine are able to restore the normal functionality of Hp and PFCx under stress situations, while the amygdala retains the ability to increase its activity in the same stress conditions [42].

The disturbed adult hippocampal neurogenesis cannot fully explain major depression. It could only be the most conspicuous feature of a more fundamental type of cellular plasticity, which could also govern the prefrontal cortex and other regions. It has also been proposed that, in addition to neural proliferation, changes in synaptic plasticity would also be involved in the biological basis of depression [19], being modulated by antidepressant treatments [43, 44]. PFCx is also a region sensitive to stress-induced effects, with a reduction in the number and length of spines [45] in apical dendrites of the pyramidal cells in the medial prefrontal cortex area [46, 47], as well as changes in the number, morphology, metabolism, and function of glial cells, that produce changes in the glutamatergic transmission, resulting in memory impairments [48–51], and reduced synaptic plasticity in the Hp-PFCx neuronal pathway [52]. The increase in extracellular glutamate could be one of the reasons underlying the molecular changes associated to stress [51]. However, while frontal cortex and hippocampus are reduced and hypofunctional in major depression, structures such as amygdala present hypertrophy and hyperactivity. Changes in synaptic plasticity will reflect a vulnerability to suffer a depression episode [39].

An increased apoptosis has also been related to a higher risk of suffering major depression since increased cell death in areas as dentate gyrus (DG), CA1, and CA4 areas of the hippocampus, entorhinal cortex, and subiculum are reported in studies using human postmortem brain samples though this phenomenon does not seem to account for the hippocampal volume reduction [53]. Animal studies also report that acute stress increase hippocampal apoptosis [29], while chronic stress induces no changes [29], increased apoptosis in cortex [54, 55], or hippocampus [55]. Antidepressant treatment decreases cell death by different mechanisms, as the activation of the expression of trophic factors (BDNF and its receptor TrkB) which results in increased cell survival [56, 57] or directly reducing cellular apoptosis in animal stress models as reported for fluoxetine [28, 58].

It has been suggested that an increase in serotonin levels mediates the raise in cell proliferation, while the depletion of this neurotransmitter does not lead to an immediate effect over the hippocampal cell division [59]. In line with that, the direct or indirect modulation leads to an increase in proliferation. Treatments exerting a direct action over the serotonergic system include chronic but not acute administration with drugs such as tricyclic antidepressants, monoamine oxidase inhibitors (MAOI), serotonin-selective reuptake inhibitors (SSRI), serotonin and noradrenaline reuptake inhibitors (SNRI), and $5HT_4$ agonists [8, 19, 20, 60–65]. A nonpharmacological intervention such as the silencing of the serotonin transporter (SERT) by RNAi in dorsal raphe serotonergic neurons also leads to increased hippocampal proliferation [66]. The administration of other drugs, such as lithium in combination with antidepressants as desipramine, produces an increase in hippocampal proliferation [67] and a decrease in apoptosis of hippocampal progenitor cells in irradiated animals [68]. Treatments with antidepressants that increase serotonin levels in brain act by targeting different progenitor cell populations. Thus, chronic administration of the SSRI fluoxetine [69] or subchronic treatment with a $5\text{-}HT_4$ agonist [8] increases cell proliferation and neurogenesis-targeting amplifying neural progenitors (ANPs) (Figure 1) while chronic electroconvulsive seizure (ECS) produces a fast-acting effect targeting both quiescent neural progenitors (QNPs) and ANPs [70]. An increased hippocampal proliferation as a consequence of chronic antidepressant treatment has been proven necessary for some [71–73], but not all the antidepressant-like effects in animals. The antidepressant-like effects have been related to the increased hippocampal proliferation [8, 71, 74], dendritic arborization, maturation, and functional integration of newborn neurons [75]. However, other drugs with potential antidepressant action do not mediate their effect through the activation of progenitor cells division, since the complete elimination of hippocampal proliferation by direct irradiation of this structure does not block the antidepressant response promoted by the blockade of drugs acting on other neurotransmitter systems as the corticotrophin-releasing factor receptor (CRF) or arginine vasopressin 1b (V1b) receptors [76].

3. Pathways Leading to Proliferation and Neural Plasticity Changes That Exert Antidepressant-Like Effect

Classically, the modulation of different neurotransmitter systems has been implicated in the mediation of the antidepressant effects, and, for some of them, a link with proliferative or plastic changes has been reported. The traditionally involved neurotransmitter systems include the serotonergic, adrenergic, and dopaminergic ones, while others, such as the glutamatergic and cannabinoid systems and the corticotropin-releasing factor (CRF) system implicated in the secretion of ACTH are acquiring increasing importance in the last years. Here we will focus on the serotonergic receptors most relevant to modulating neural proliferation and synaptic plasticity processes.

3.1. Serotonergic Receptors. Serotonin has a positive role in the regulation of hippocampal neurogenesis. The partial lesion of dorsal and medial raphe nuclei, which results in a decrease of serotonergic neurons that innervate the dentate gyrus of the hippocampus and other projection areas as cortex and amygdala, decreases the proliferation in the subgranular zone of the dentate gyrus [132]. Several serotonin

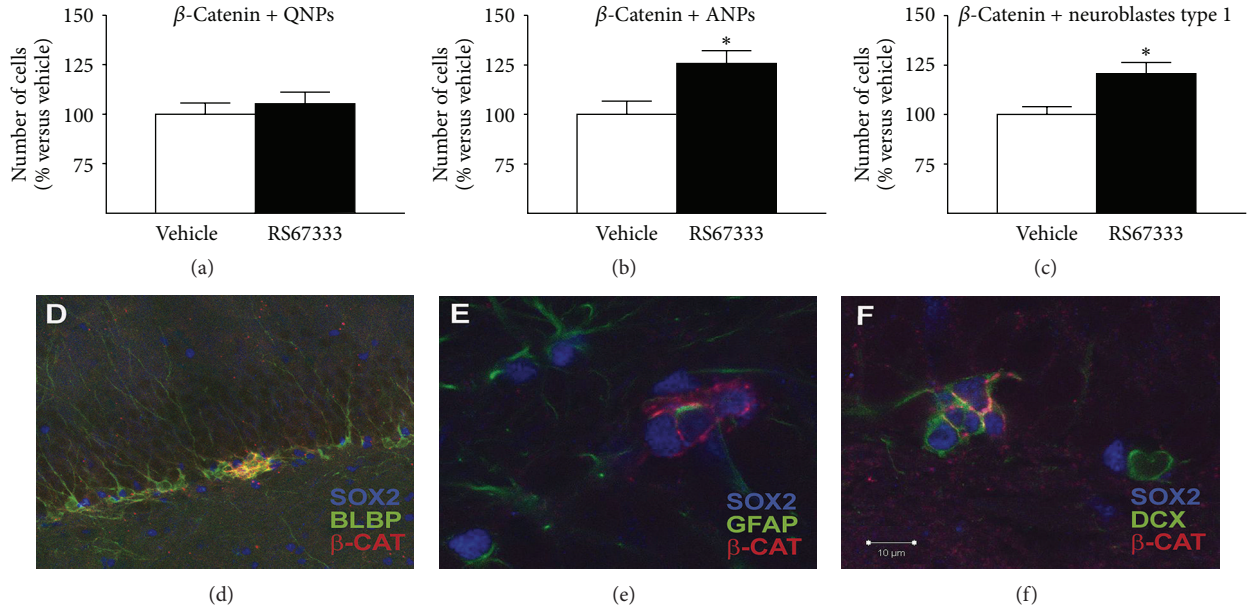


Figure 1: RS67333 increases the number of ANPs (b) and neuroblasts type 1 (c) that express β-catenin, but not total number of QNPs cells (a). Photomicrographs illustrating β-catenin expression in neural progenitors (d), ANPs (e), and neuroblasts (f). The results are the Mean ± S.E.M. $^{*}P < 0.05$ versus vehicle. Bar: 10 μM, modified from Pascual-Brazo et al., 2012 [8].

receptors have been involved in the antidepressant-induced increase of cell proliferation in the hippocampus, together with neurite outgrowth and cell survival in cells expressing these receptors [80]. However, other authors report a lack of changes in proliferation and/or neurotrophic factors expression after chronic treatment with the antidepressant fluoxetine, questioning the importance of the serotonergic system in hippocampal proliferation [133–135].

3.1.1. 5-HT$_{1A}$ Receptors. The importance of this serotonergic subtype in the effects of antidepressants has been shown in studies in vivo using 3 day treatment with the 5-HT$_{1A}$ agonist 8-OH-DPAT [77], and chronic administration of this drug [74, 79, 80] that produce an increase in proliferation in the subgranular zone (SGZ) of the hippocampus that depends on the 5-HT$_{1A}$ postsynaptic receptors. In studies using hippocampal neural progenitors, the serotonin-mediated increase in proliferation is blocked by the 5-HT$_{1A}$ antagonist WAY100,635 [78]. On the contrary, the acute administration of 5-HT$_{1A}$ antagonists produces a decrease of hippocampal proliferation [136], or no changes after 14 days [79]. Knock out animals for the 5-HT$_{1A}$ receptor subtype present no changes in basal proliferation compared to wild type animals [9, 71], but present a decreased hippocampal cell survival [9]. The 5-HT$_{1A}$ receptor subtype has been proven necessary for the hippocampal proliferative effect of some antidepressants as fluoxetine [71], although other drugs as imipramine, acting on other neurotransmitter systems, increases hippocampal proliferation in a 5-HT$_{1A}$-independent manner [71] (Table 1).

3.1.2. 5-HT$_{2A/C}$ Receptors. The role of the 5-HT$_{2A/C}$ receptors in the regulation of neurogenesis is less clear. The chronic administration of 5-HT$_{2A}$ antagonists as ketanserin [81], and 5-HT$_{2C}$ antagonists as SB243,213 and S32006 [82], produce the increase in hippocampal proliferation, while the acute treatment with 5-HT$_{2A/C}$ agonists or antagonists produce no changes or a decrease in proliferation, respectively [74, 81]. Interestingly, the subchronic treatment with ketanserine in combination with the SSRI fluoxetine increases a series of synaptic plasticity markers as β-catenin and N-cadherin present in the membrane fraction, together with BDNF gene expression, however, hippocampal proliferation is not significantly modified [83]. The increased proliferation or synaptic plasticity parallel the antidepressant-like effect observed for the treatments with antagonists [83, 137, 138], while the administration of 5-HT$_{2A}$ agonist counteracts the effect of SSRIs [137]. The blockade of 5-HT$_{2A}$ receptor subtype located in GABAergic interneurons produces the activation of hippocampal pyramidal neurons [139] that modulates dendritic activation and synaptic plasticity [140] (Table 1).

3.1.3. 5-HT$_4$ Receptors. In the last years, the 5-HT$_4$ receptor subtype has been proven to have an outstanding role on the depressive pathology. This receptor subtype density and signaling cascade through cAMP are up-regulated in the frontal cortex and caudate-putamen of depressed humans [141]. Chronic treatments with classical antidepressants produce a desensitization of this subtype in structures as hippocampus [97, 98]. In the last years, it has been described a short-term antidepressant-like response mediated by 5-HT$_4$ partial agonists [8, 64, 65], or when coadministered with classical antidepressants [63]. The antidepressant effect of 5-HT$_4$ agonist is mediated by an increase in hippocampal proliferation in vivo [8, 64], together with other proliferative and plasticity

Table 1: Role of the activation and blockade of the different serotonin receptor subtypes in neural proliferation and synaptic plasticity.

Serotonin receptor subtypes	Effect of pharmacological manipulation on hippocampal proliferation				ko	
	Agonist		Antagonist			
	Change	Reference	Change	Reference	Change	Reference
5-HT_{1A}	↑ proliferation (subchronic)	[77]	↓ proliferation (acute) = proliferation (chronic)	[78] [79]	= proliferation	[9, 71]
	↑ proliferation (chronic)	[74, 79, 80]	blocks 5-HT-induced proliferation	[78]	↓ cell survival	[9]
5-$HT_{2A/2C}$	= proliferation (+SSRI)	[74]	↑ proliferation (chronic) = proliferation (acute) ↑ plasticity markers and BDNF (+SSRI, subchronic)	[81, 82] [74, 81] [83]	*No data*	
5-HT_4	↑ proliferation (subchronic) ↑ plasticity markers and BDNF (subchronic)	[8, 64] [8, 63–65]	Blocks 5-HT-induced proliferation	[78]	*No data*	
5-HT_6	*No data*		= proliferation ↑ plasticity markers (PSA-NCAM)	[84] [84]	*No data*	
5-HT_7	*No data*		↑ proliferation (subchronic)	[85]	= proliferation	[86]

↑ : increase; ↓ : decrease; ko: knock-out mice.

markers as β-catenin, Akt [8], BDNF [8, 65], phosphorylated cAMP response element binding (CREB) protein [8, 63–65]. The 5-HT_4 implication in serotonin-induced hippocampal proliferation has been observed blocking this receptor with the 5-HT_4 antagonist DAU 6285 in primary hippocampal progenitor cell cultures [78] (Table 1).

3.1.4. 5-HT_6 Receptors. The role of the 5-HT_6 receptor subtype in depression is not clear, but tricyclic antidepressants as amitriptyline and atypical antidepressants as mianserin have high affinity for this serotonin receptor subtype, acting as antagonists [142]. Moreover, the expression of the 5-HT_6 receptors is regulated by glucocorticoid levels [142]. This receptor subtype is present postsynaptically in brain areas as cortex and hippocampus and is implicated in learning and memory [142]. The action over this receptor to date is contradictory since it has been published that both antagonists and agonists exert antidepressant and anxiolytic effects alone [142, 143] or enhance the beneficial effect when combined with antidepressant drugs [144]. When locally administered on hippocampus, 5-HT_6 antagonists produce antidepressant-like effect [145]. However, this effect is not mediated by increased neurogenesis but for an increase in neural cell adhesion molecule polysialylation (PSA-NCAM) that may mediate memory consolidation [84] through long-term changes in synaptic plasticity [146] (Table 1).

3.1.5. 5-HT_7 Receptors. The 5-HT_7 receptor subtype is also involved in the antidepressant effect. Recent studies have shown that the blockade of the 5-HT_7 receptor subtype produces antidepressant-like behaviour [147, 148]. This is supported by studies in animal depression models as the olfactory bulbectomy [85], the antidepressant-like behaviour of knock-out mice for the 5-HT_7 receptor subtype [149], and clinical data using the antagonist Lu AA21004 [150]. Moreover, a 7-day treatment with the 5-HT_7 antagonist SB-269970 produces an increase in proliferation in the subgranular zone of the hippocampus [85] although changes in the number of dividing cells do not appear in 5-HT_7 knock-out animals [86] (Table 1).

3.2. Neurotrophic Factors. In an attempt to explain those brain changes implicated on depression and/or antidepressant effect that could not be included in the initial monoaminergic hypothesis of depression, it was postulated the so-called "Neurotrophic hypothesis of Depression" that later was revised to a "new" "hypothesis of neuroplasticity" [18]. This hypothesis links the changes in depression models to a decrease of brain-derived neurotrophic factor (BDNF) and the antidepressant effect to an increase in BDNF in hippocampus [18, 19, 151, 152]. Moreover, the decreased BDNF observed in heterozygous knock-out mice ($BDNF^{+/-}$) is related to a depression-like phenotype [18]. These changes in brain BDNF expression are paralleled by serum levels, so that it has been proposed as a biomarker for depression disease, positive or negative response of the individuals to the antidepressant treatment [153–156], and even a marker of suicidal depression [157]. However, the role of BDNF is still not clear in the depressive pathology since some authors describe a lack of changes on the BDNF levels associated to stress animal models [57, 158–160].

The infusion of BDNF in brain [161, 162] or more specifically in hippocampus [163, 164] produces antidepressant-like effects. Moreover, within the hippocampus, the infusion of BDNF in the DG but not in the CA1 region produces an antidepressant-like effect [163], which is supported by the lack of antidepressant action in mice selectively knocked out for the BDNF gene in the DG and not in the CA1 [160]. Even peripherally administered BDNF is able to display antidepressant-like actions [165], resembling the increased serum BDNF observed after antidepressant treatments [166].

Chronic administration of antidepressants produces an increase in hippocampal BDNF mRNA expression and BDNF protein levels (Figure 2) [8, 65, 167]. The blockade of 5-HT_{2A} receptor reverses the effect of stress-induced downregulation of BDNF mRNA expression in hippocampus [168]. Also, the subchronic treatment with SSRI and 5-HT_{2A} antagonists is able to increase BDNF expression in the dentate gyrus of the hippocampus; however, the protein level is not yet modified in subchronic treatments (Figure 2) [83].

The main role of BDNF regarding adult neurogenesis is not linked to proliferation, but to the increase in cell survival, as described using BDNF and its receptor TrkB knock-out animals which present a reduced BDNF expression [61, 169]. BDNF is implicated in synaptic plasticity, and proteins as neuritin that are induced by BDNF are decreased in stress-induced animal models of depression [170] and increased after chronic antidepressant treatment, contributing to the BDNF antidepressant effect [170, 171].

The existence of a single-nucleotide polymorphism (SNP) in the human BDNF gene, BDNF (Val66Met) is associated to reduced BDNF secretion [172], and to an increased incidence of neuropsychiatric disorders [173, 174]. In animals BDNF (Val66Met) predisposes to a depression-like behaviour after stress situations that recover normal values after the administration of antidepressants [175]. This polymorphism is also associated to nonresponders after antidepressant treatment [176].

Other important trophic factor is the vascular endothelial growth factor (VEGF) implicated in the "vascular niche hypothesis of adult neurogenesis." This theory proposes the need of vascular recruitment associated to active sites of neurogenesis formed by proliferative cells that present an endothelial phenotype in 37% of the cases [177]. VEGF expression is reduced in hippocampal dentate gyrus after irradiation [178], and in stress models [179] although other authors do not show changes associated to stressed animal models [180]. From studies using irradiated rats, it was proposed that the decrease of progenitor cells responsible for the expression of VEGF would underlie the decrease of this factor [178].

Some antidepressant treatments, as the electroconvulsive therapy (ECS) [178, 181, 182], approache with antidepressant-like effect as exercise [180], or mood stabilizers as lamotrigine [183], result in the upregulation of VEGF expression. Moreover, the local administration of this trophic factor produces an increase in hippocampal proliferation [178]. In addition, the silencing of hippocampal VEGF [184] or the use of antagonists for its receptor Flk-1 [180] blocks its antidepressant-like effect and decreases markers of newborn neurons as doublecortin (DCX).

Even though these data indicate the importance of VEGF brain levels in the depressive disorder, preliminary reports do not show a clear correlation between peripheral VEGF and depressive disorders, not allowing for the use of this molecule as a marker of depression and/or antidepressant response [185, 186].

The activation of receptor tyrosine kinases by neurotrophic factors promotes the activation of the PI3K/Akt pathway that is linked to the Wnt/β-catenin through the inhibition of GSK-3β and to the mTOR pathway through the phosphorylation of mTOR protein [187] that are discussed below. The PI3K/Akt pathway per se has an outstanding role in promoting adult hippocampal proliferation and the inhibition of cell differentiation [188]. Antidepressant treatments also produce increases in Akt levels in structures as hippocampus [8, 10] and frontal cortex [27].

3.3. Intracellular Pathways

3.3.1. Cyclic Adenosine Monophosphate (cAMP) Cascade. The upstream and downstream components of the cAMP signaling pathway have been extensively involved in the pathophysiology of mood disorders as well as in the actions of antidepressant drugs. Alterations in several elements of this pathway, such as G proteins (Gs or Gi), adenylate cyclase (AC), cAMP levels, cAMP-dependent protein kinase (PKA), and the cAMP response element-binding protein (CREB) transcription factor, have been described in peripheral cells and the postmortem brain of patients with affective disorders, both untreated or after antidepressant therapy [11, 100, 189, 190]. Various elements along this pathway have been identified as potential targets for antidepressant drugs (Table 2).

In peripheral cells and postmortem brains of patient with mayor depression, there is a reduction of the adenylyl cyclase (AC) activity in response to forskolin [87], β_2-adrenergic agonists [88–93], and α2-adrenoceptor agonists [94]. Chronic treatment with antidepressant drugs produces the increase in cAMP levels in rat hippocampus, cortex, and striatum, as well as in postmortem human frontal cortex samples from depressed patients (Figure 3(a); personal observation). This effect has been attributed to both enhanced coupling of Gs proteins to adenylyl cyclase and increased adenylyl cyclase activity [95, 96]. The direct injection of cAMP or inhibition of cAMP degradation by rolipram produces antidepressant-like effect in animals [99]. Chronic antidepressant treatment in rat desensitizes cAMP response to serotonergic receptor as 5-HT_{1A} receptor (Figure 3(b)) and 5-HT_4 receptor [8, 97, 98] and increases the CB1-mediated inhibition of adenylyl cyclase (AC) in prefrontal cortex, an effect that is modulated by 5-HT_{1A} receptors [100].

The next step in this signaling pathway is the activation of cAMP-dependent protein kinase (PKA) by cAMP, so that PKA activity is increased after chronic antidepressant administration [101]. Active PKA phosphorylates proteins as CREB, a transcription factor that regulates the expression of

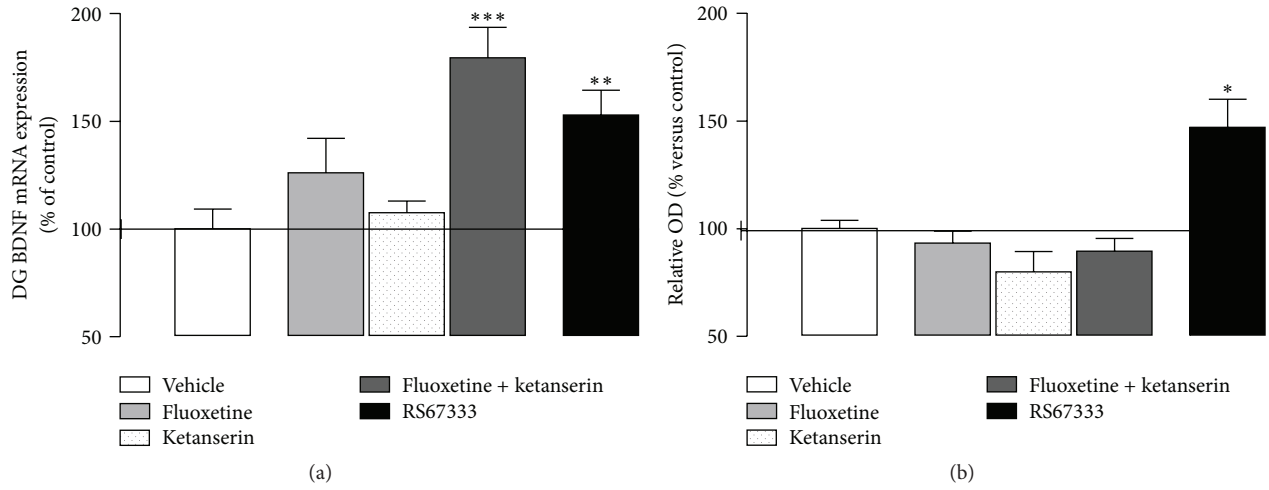


Figure 2: BDNF mRNA expression (a) and protein level (b) in the dentate gyrus of the hippocampus (DG) or total hippocampus, respectively, after 7-day treatment with the 5-HT_4 partial agonist RS67333 (1.5 mg/kg/day) (modified from [8]) and 7-day coadministration of the SSRI fluoxetine (5 mg/kg/day) and the 5-HT_{2A} antagonist ketanserin (0.1 mg/kg/day). $^{*}P < 0.05$ versus vehicle. Modified from Pascual-Brazo et al., 2012 [8], and Pilar-Cuéllar et al., 2012 [83].

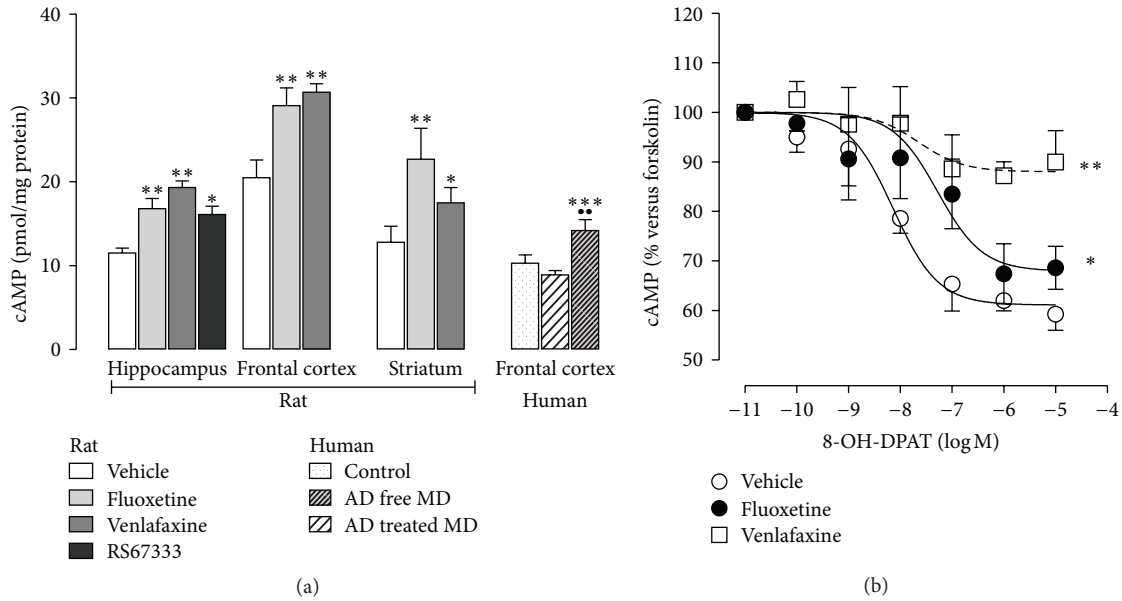


Figure 3: Antidepressant treatment increases basal cAMP in both rat and human. (a) Increase of basal cAMP levels in rat hippocampus, frontal cortex and striatum after chronic (14 days) antidepressant treatment with fluoxetine (10 mg/kg/day) and venlafaxine (40 mg/kg/day), and subchronic treatment (3 days) with the $5HT_4$ agonist RS67333 (1.5 mg/kg/day) and in postmortem frontal cortex samples from control, antidepressant free-depressed subjects (AD-free MD) and antidepressant-treated depressed subjects (AD-treated MD). cAMP is expressed in pmoles/mg protein. $^{*}P < 0.05$; $^{**}P < 0.01$ and $^{***}P < 0.001$ versus vehicle or control subjects; $^{\bullet\bullet}P < 0.01$ versus antidepressant-free depressed subjects. (b) Modulation of 5-HT_{1A} receptor subtype-mediated inhibition of cAMP accumulation by antidepressant drugs. Chronic antidepressant treatment with fluoxetine (10 mg/kg/day) and venlafaxine (40 mg/kg/day) for 14 days downregulates 8-OH-DPAT inhibition of forskolin-induced cAMP accumulation. (a) Modified from Mostany et al., 2008 [10], Pascual-Brazo et al., 2012 [8], and unpublished results and (b) unpublished results.

several genes involved in neuroplasticity, cell survival, and cognition [191–195].

CREB has been widely involved in the pathophysiology of depression and both behavioural and cellular responses to antidepressant treatments [11, 190]. Hippocampal expression of CREB is reduced in response to stress exposure [102, 103]. In contrast, chronic but not acute antidepressant therapy and electroconvulsive shock (ECS) increase levels of CREB mRNA, CREB protein (Figure 4), and CREB activity—promoting the phosphorylation of this protein—effects that

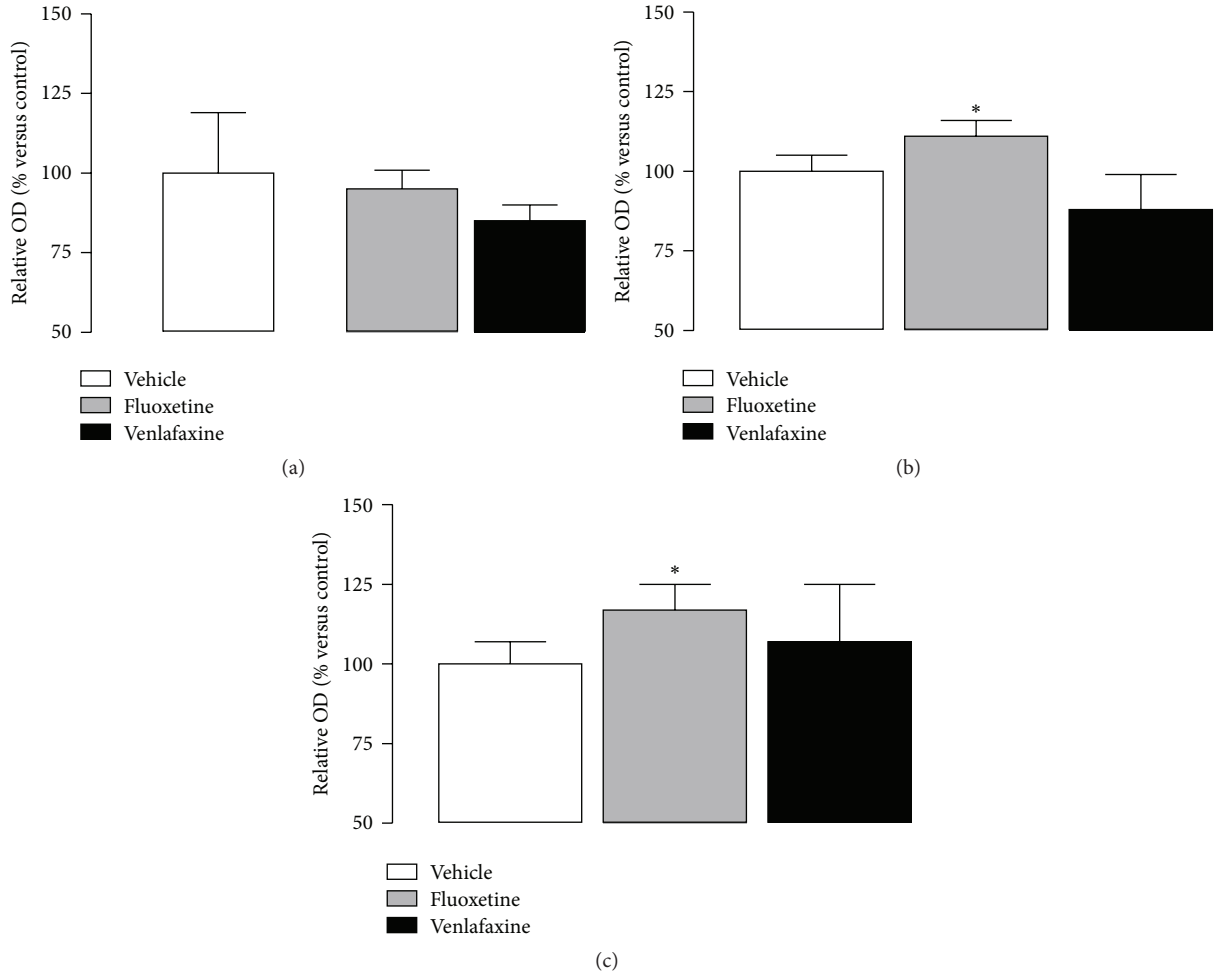


Figure 4: Western blot analyses of hippocampal CREB (a) and p-CREB (b) levels and ratio p-CREB/CREB (c) after chronic antidepressant treatment (14 days) with fluoxetine (10 mg/kg/day) and venlafaxine (40 mg/kg/day). Note that p-CREB levels and ratio p-CREB/CREB are increased after fluoxetine treatment, but not total CREB, or after venlafaxine treatment in total cell lysate from hippocampus of rats. Values are means ± S.E.M. Corresponding to densitometry levels of the proteins expressed as the percentage of the same proteins in vehicle-treated animals. $^{*}P < 0.05$ versus vehicle. Modified from Mostany et al., 2008 [10], and Mato et al., 2010 [100].

seems to be area and drug dependent [11, 103, 105–108]. Thus, increased phosphorylated CREB levels in hippocampus are linked to antidepressant-like behaviour [44], as observed after viral-mediated overexpression of CREB in hippocampus in behavioural models of depression [44]. Contrary to what could be expected, CREB overexpression in the nucleus accumbens produces prodepressive effects [111], and lowered CREB in the nucleus accumbens in mice exhibits an antidepressant-like response [196]. A different pattern appears also for amygdala, in which high CREB levels produce opposite effects depending on the timing. Thus, when CREB overexpression is induced before learned helplessness training, there is a prodepressant effect, while the increase of CREB after the training is antidepressant [109]. Studies in postmortem human brain indicate lower levels of CREB protein in depressed antidepressant-free subjects, in contrast to the increased CREB level in patients taking an antidepressant at the time of death [104]. These results are parallel to studies in human fibroblasts of patients with major depression [110], which is consistent with animal studies. Among the several target genes regulated by CREB, two of the more relevant, are the brain-derived neurotrophic factor (BDNF) and the vascular endothelium growth factor (VEGF) [19, 60, 197, 198].

A growing body of data shows that other signalling cascades can modulate CREB activity through phosphorylation, such as the calcium/calmodulin-dependent kinase (CaMKII) and the mitogen-activated protein (MAP) kinase cascades, and may also be implicated in the mechanism of action of antidepressants [11, 199].

Initially, all effects of cAMP increase were attributed to the activation of PKA/CREB, but two novel targets as the cAMP-regulated ion channels and Epac (exchange protein directly activated by cAMP) are now known to be involved in mediating cAMP responses. An increase in Epac-2 levels, but not Epac-1, has been found in postmortem samples of prefrontal cortex and hippocampus of depressed subjects [112].

3.3.2. Wnt/β-Catenin Pathway. The Wingless-type (Wnt) family of proteins has key roles in many fundamental processes during neurodevelopment [200]. The role of this pathway in neural development, through the modulation of neural stem cells' (NSC) proliferation and differentiation, has been clearly demonstrated [201]. Some of the processes regulated by Wnt/β-catenin pathway activity are neural differentiation [202], hippocampal formation [203, 204], dendritic morphogenesis [205, 206], axon guidance [207, 208], and synapse formation [209]. Moreover, it also plays an important role in spatial learning [210] and memory, including long-term potentiation (LTP) phenomena [159].

In the absence of Wnt signaling, β-catenin function is blocked by a destruction complex consisting of Axin, APC, and GSK-3β and CK1a kinases [211], which phosphorylates β-catenin for destruction in the proteasome [212, 213]. Wnts act through both canonical and noncanonical signal transduction pathways. Canonical Wnt signaling results in the inhibition of GSK-3β which is constitutively active, and the nonphosphorylated β-catenin is stabilized in the cytoplasm and translocated to the nucleus, which is essential for canonical Wnt signaling [214]. Once in the nucleus, β-catenin forms a complex with the T-cell factor/lymphoid enhancer factor (TCF/LEF) transcription factors, to activate the expression of Wnt target genes. TCF/LEF transcription factors are bound to Groucho, a protein producing repressive effects [214]. Nuclear β-catenin promotes the displacement of Groucho and the binding of the histone acetylase cyclic AMP response element-binding protein (CREB), activating the transcription machinery [214, 215]. The noncanonical pathway or β-catenin independent is mediated through Rac/Rho (Wnt/PCP) or through calcium (Wnt/Ca^{2+}) [201].

In the last years several evidences have implicated Wnt-signaling pathway in the pathophysiology and treatment of mood disorders and other cognitive pathologies. GSK-3β and β-catenin are regulated either directly or indirectly by lithium, valproate, antidepressants, and antipsychotics [8, 10, 113–117], while GSK-3β has also been identified as a target for the treatment of Alzheimer's disease [216] (Table 2).

Postmortem human brain samples from depressed subjects and teenage suicide victims present a dysregulation of Wnt/GSK-3β signaling with a decrease in β-catenin expression in prefrontal cortex [124]. β-catenin knock-out mice with 50–70% decrease of β-catenin expression in forebrain regions present an increased immobility time in the tail suspension test indicating a depression-like state, but not in other anxiety tests [125].

The inhibition of GSK-3β activity, either pharmacologically [117–119], or through deletion in mouse forebrain, results in an increase in brain β-catenin levels, as well as in antidepressant-like effects or decreased anxiety [120], as observed by the direct overexpression of β-catenin in mouse brain [126]. GSK-3β inhibition by lithium is an important regulator of cell survival related to mood stabilizers [215] and displays antidepressant efficacy [115, 118, 215, 217, 218]. In contrast, GSK-3β knockin mice displayed increased susceptibility to stress-induced depressive-like behaviour [121], presenting decreased cell proliferation in the subgranular zone of the dentate gyrus, accompanied by a reduction in VEGF, but not BDNF, and blunted neurogenesis in response to antidepressant treatments [219]. These data support the importance of the Wnt pathway activation and β-catenin levels associated to mood disorders and their treatment. In addition, SNP variation in the promoter region of GSK-3β plays a protective role in the onset of bipolar illness [122] and increased antidepressant response [123].

Recent studies have identified the Wnt/GSK-3β/β-catenin-signaling pathway as a key regulator of adult neurogenesis in hippocampus [220, 221] or subventricular zone [222], highlighting the role of GSK-3β on neural progenitor homeostasis [200]. Wnt proteins are signaling molecules that are released from hippocampal neural stem cells (NSC) and astrocytes, acting autocrinally to regulate proliferation via Wnt canonical pathway [220, 221].

Wnt/β-catenin pathway is activated by antidepressant treatments as electroconvulsive therapy [26], chronic treatments with classical antidepressants as the dual serotonin-noradrenaline reuptake inhibitor (SNRI) venlafaxine (Figure 5(a)) [10], and 5-HT_4 partial agonists [8]. The antidepressant-induced β-catenin increase is observed in the subgranular zone (SGZ) of the dentate gyrus (DG) of the hippocampus, in membrane and nuclear fractions [10, 26]. The increased proliferation observed in SGZ after chronic antidepressant treatments is localized in cell clusters that also show a positive β-catenin staining [8, 10].

Other treatments with antidepressant-like efficacy, such as the subchronic administration of SSRI fluoxetine together with the 5-HT_{2A} antagonist ketanserin, also produce a β-catenin increase in the membrane fraction but not in the nuclear one, which corresponds with a lack of changes in hippocampal proliferation (Figure 5(b)) [83]. The increase in membrane-associated β-catenin is parallel to an elevation of N-cadherin protein [83], both members of the β-catenin/N-cadherin complex present in pre- and postsynaptic terminals [223, 224], where β-catenin recruits scaffolding proteins [225], conforming cell-cell adhesion complexes [226], recruiting synaptic vesicles [209, 227], and acting on the development of new synapses [225]. This suggests a preference of modifications in synaptic plasticity instead of proliferation, as previously reported for other antidepressant treatments [60].

In addition, Frizzled receptors and GPCRs can interact through several pathways [228, 229]. Some GPCRs act through Gq and/or Gi proteins activating PKB (protein kinase B)/Akt which inhibits GSK-3β via phosphorylation. These receptors can also activate Gs proteins that activate prostaglandin E2 (PGE2), phosphoinositide 3-kinase (PI3K), and PKB/Akt, leading to the inhibition of GSK-3β. Other receptors act on Gq or G12/13 proteins, activating the phospholipase Cβ (PLCβ) and protein kinase C (PKCs) and inhibiting GSK-3β [228]. Taken together, these data support the possible existence of interactions between the GSK-3β/β-catenin pathway and other neurotransmitter systems involved in depression, including serotonin. The pharmacological modulation of the different elements of the Wnt/β-catenin pathway with antidepressant purposes has to be clarified in the near future, probably modulating at the

Table 2: Involvement of cAMP, Wnt/β-catenin, and mTOR-signaling pathways in major depression (MDD) and antidepressant treatment.

Signaling pathways		Changes related to disease or treatment						Direct effect	
		Nontreated MDD (versus control)			Treated MDD (versus untreated)				
		Change	Region	Reference	Change	Region	Reference	Change	Reference
cAMP/PKA	cAMP	↓ AC (induced by FK, β-AR, α2-AR)		[87–94]	↑ cAMP ↓ 5-HT_{1A}-induced AC inhibition/↓ 5-HT_4-induced AC stimulation	Human PFCx	[95, 96] [8, 97, 98]	cAMP (administration/degradation inhibition) → AD-like effect	[99]
	PKA		*No data*		↑ AC CB1-induced inhibition ↑ PKA activity		[100] [101]	*No data*	
	CREB	↓ CREB		[102–104]	↑ CREB (protein, expression, activity)	Hp, Cx	[11, 103–110]	CREB viral expression in Hp → AD-like effect CREB viral expression in Acb → depression-like effect	[44] [111]
	Epac	↑ Epac-2	Hp, PFCx	[112]	*No data*			*No data*	
Wnt/β-catenin	GSK-3β	↓ GSK-3β	Hp	[113]	↑ GSK-3β	Hp	[8, 10, 113–117]	GSK-3β inhibition → AD-like and anxiolytic effect GSK-3β knock in → stress-induced depression GSK-3β SNP (−50 T/C; rs334558) → later onset of bipolar depression GSK-3β SNP (−50 T/C; rs334558) → ↑ antidepressant response	[117–120] [121] [122] [123]
	β-catenin	↓ β-catenin	PFCx	[124]	↑ β-catenin	Hp	[8, 10, 26, 83, 113–117]	β-catenin ko mice → depression-like effect β-catenin OE mice → AD-like effect	[125] [126]
mTOR	mTOR	↓ mTOR, p70S6K, eIF4B, p-eIF4B		[127–129]	↑ p-mTOR/mTOR	PFCx	[27, 130]	Activation (NMDAr antagonists) → AD-like effect	[27, 131]

↑ : increase; ↓ : decrease; AC: adenylyl cyclase activity; FK: forskolin; Hp: hippocampus; PFCx: prefrontal cortex; Acb: accumbens nucleus; OE: overexpressing.

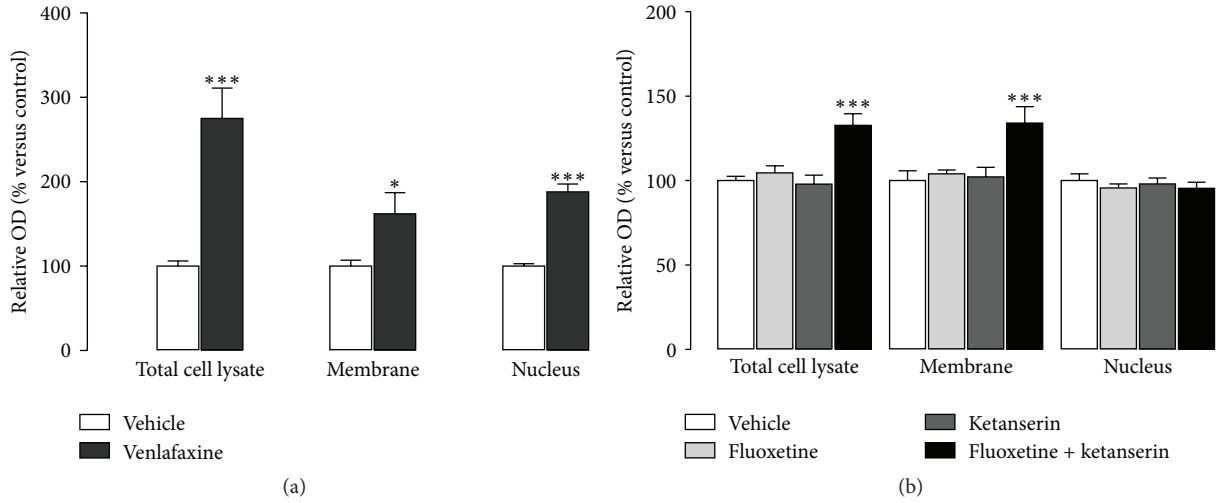


FIGURE 5: Implication on β-catenin subcellular distribution of the chronic (14 days) antidepressant treatment with the SNRI venlafaxine (40 mg/kg/day) (a), and 7-day treatment with the SSRI fluoxetine (5 mg/kg/day), the 5-HT_{2A} antagonist ketanserin (0.1 mg/kg/day), and the combination of both (b). Note that chronic treatment with venlafaxine produces an increase in both membrane-associated and nuclear β-catenin, while the subchronic treatment with fluoxetine + ketanserin, only increases β-catenin in the membrane but not in the nuclear fraction. $^{*}P < 0.05$ and $^{***}P < 0.001$ versus vehicle. (a) Modified from Mostany et al., 2008 [10], and (b) modified from Pilar-Cuellar et al., 2012 [83].

level of Wnts or β-catenin activity. Interestingly, a number of patents regarding GSK-3β inhibition as the therapeutic mechanism for treatment of neuropsychiatric disorders are being launched, including treatment of depression.

3.3.3. mTOR Pathway. Target of rapamycin (TOR) genes, members of the phosphoinositol kinase-related kinase (PIKK) family of kinases [230], was first described in yeast as the pharmacological targets of the microbicide rapamycin [231]. TORs were subsequently described in other invertebrate and vertebrate organisms. mTOR, the mammalian form of this protein, exists in two different functional multiprotein complexes within the cells, mTORC1 and mTORC2, which are evolutionarily conserved from yeast to mammals [232, 233]. mTORC2 is involved in cytoskeletal remodeling [234] and in the regulation of cell survival and cell cycle progression. mTORC1, the primary target of rapamycin, is involved in cell proliferation, cell growth and survival by protein translation, energy regulation, and autophagy in response to growth factors, mitogens, nutrients, and stress [235–237].

In neurons, mTORC1 activity is regulated by phosphorylation in response to growth factors, as BDNF, mitogens, hormones, and neurotransmitters through the activation of G protein-coupled receptors (GPCRS) or ionotropic receptors. The mTORC1 phosphorylation is mediated by ERK/MAPK, PI3K, PKA, and Epac. The activation of mTORC1 results in the phosphorylation and activation of several downstream targets as the eukaryotic initiation factor 4E-binding protein 1 (4E-BP1), p70 ribosomal S6 kinase (p70S6K), RNA helicase cofactor eIF4A, extracellular signal-regulated kinase (ERK, including both ERK1 and ERK2), or PKB/Akt; and the inhibition of the eukaryotic elongation factor 2 kinase (eEF2) [238, 239].

mTOR has been extensively studied related to cancer, development, metabolism, and more recently to the central nervous system (CNS) physiology and diseases [238, 240, 241]. mTOR-signaling pathway is involved in synaptic plasticity, memory retention, neuroendocrine regulation associated with food intake and puberty, and modulation of neuronal repair following injury. The target proteins of mTOR, 4E-BP1, and eukaryotic initiation factor-4E (eIF4E) have been detected in cell bodies and dendrites in cultured hippocampal neurons and their distribution completely overlaps with the postsynaptic density protein-95 (PSD-95) at synaptic sites, suggesting the postsynaptic localization of these proteins [242]. The activation of mTOR has been functionally linked with local protein synthesis localized presynaptically as synapsin I, or postsynaptically as PSD-95 and GluR1, and cytoskeletal proteins as the activity-regulated cytoskeletal-associated protein (Arc) [27, 241, 243].

mTOR-signaling pathway has been also related to a number of neurological diseases, such as Alzheimer's disease, Parkinson's disease, and Huntington's disease, tuberous sclerosis, neurofibromatosis, fragile X syndrome, epilepsy, brain injury, and ischemic stroke [244]. Dysfunction of mTORC1 is associated with the pathogenic mechanisms of Alzheimer's disease, and the activation of p70S6K, downstream of mTORC1, has been identified as a contributor to hyperphosphorylated tau accumulation in neurons with neurofibrillary tangles [245].

Recent studies have also associated mTOR signaling in affective disorders since the administration of ketamine produces a fast-acting antidepressant-like effect in animals [27] and human [131]. In stressed rats, a reduction in PI3K-Akt-mTOR-signaling pathway has been reported in PFCx [127, 128] or amygdala [129]. The inhibition in mPFCx of calcineurin, a serine/threonine protein phosphatase that

participates in the regulation of neurotransmission, neuronal structure and plasticity, and neuronal excitability, induces a depression-like behaviour [246], accompanied by a decrease in mTOR activity [130]. This effect can be reverted by the activation of mTOR by NMDA or the chronic administration of the antidepressant venlafaxine, promoting an antidepressant-like effect [130]. In human postmortem samples of prefrontal cortex of depressed subjects, there is a decrease in the expression of mTOR, as well as some of the downstream targets of this pathway, as p70S6 kinase (p70S6K), eIF4B, and its phosphorylated form, which suggests the impairment of the mTOR pathway in major depressive disorder (MDD) that would lead to a reduction in protein translation [247] (Table 2).

The subchronic, but not acute, administration of rapamycin in rodents has an antidepressant-like effect shown in two behavioural tests as forced swimming and tail suspension tests [248]. Acute administration of different NMDA receptor antagonists as the ketamine [27], Ro 25-6981 [27], and MK-801 [249] or antagonists of the group II of the metabotropic glutamate receptors (mGlu2/3), as MGS0039 and LY341495, produce a fast antidepressant effect [250, 251] mediated by mTOR-signaling pathway activation. Ketamine rapidly activates the mammalian target of rapamycin (mTOR) pathway, increases synaptogenesis, including increased density and function of spine synapses, in the prefrontal cortex of rats [27, 243], and increases hippocampal BDNF expression [252], that results in a rapid antidepressant-like effect in rats [27, 243] and humans [253]. Moreover, blockade of mTOR signalling by the specific antagonist rapamycin completely blocks the ketamine induction of synaptogenesis and behavioural responses in models of depression [27]. Other antidepressant strategies as the electroconvulsive treatment (ECT) also activate the mTOR pathway, leading to an increase in VEGF [254]. Therefore, modulation of mTOR could be a novel approach to develop strategies for the treatment of affective disorders [255].

4. A Further Step: Neuroplasticity versus Proliferation

The neurogenesis hypothesis of depression was based upon the demonstration that stress decreased adult neurogenesis in the hippocampus. This reduction in the production of newborn granule cells in the hippocampal dentate gyrus is related to the pathophysiology of depression. Since then, several studies have established that newborn neurons in the dentate gyrus are required for mediating some of the beneficial effects of antidepressant treatments since the increase in cell proliferation after antidepressant treatment is only observed in the SGZ and not in SVZ, suggesting a specificity of the antidepressants to regulate hippocampal neurogenesis. Moreover, psychotropic drugs without antidepressant activity do not increase neurogenesis [135, 256]. The disruption of hippocampal proliferation by irradiation is not sufficient to drive a depression-like phenotype. Both X-irradiation and genetic manipulation approaches demonstrated a requirement of hippocampal neurogenesis in mediating some of the antidepressant treatment effects [71, 76, 257], while mice exposed to X-irradiation of the SVZ or cerebellum responded normally to the antidepressants. However, some drugs with potential antidepressant action do not mediate their effect through the increase in hippocampal proliferation, as drugs acting on corticotrophin releasing factor receptor (CRF) or arginine vasopressin 1b (V1b) receptors [76], as indicated previously.

The appearance of the antidepressant-like effect in behavioural tests after 2-3 weeks parallels the time needed for the growth of newborn cells in hippocampus [258]. However, this time course does not always take so long. For classic antidepressants as the serotonin transporter inhibitors, a chronic regime is needed to observe that increased proliferation rate [10, 60], while, for others as ECS and 5-HT_4 agonists [8, 64], an acute or subacute treatment, respectively, is enough to increase proliferation.

The putative role of changes in synaptic plasticity and/or neural proliferation in the depressive pathology is proposed some time ago [19]. Synaptic plasticity, as indicated for proliferation, is also modulated by antidepressant treatments [43, 44]. The neural plasticity is not only functional but structural and is impaired in animal models [259]. For example, there is a decrease in spine number in hippocampal CA1 and CA3 areas in bulbectomized animals that are reverted with antidepressant treatment [259–261]. This structural plasticity is more striking when new neurons are born [53], or there is an increase in neuron survival as a consequence of antidepressant treatment or ECS [262]. The new dendritic spines formed are associated to smaller postsynaptic densities (PSDs) and a higher frequency of miniexcitatory postsynaptic currents (mEPSCs), suggesting an increased number of new and active glutamatergic synapses [263].

The rapid antidepressant response to drugs as ketamine acting through the blockade of NMDA receptors appears as a new target for having fast-acting effects on the treatment of mood disorders compared to the weeks or months required for standard medications. Ketamine and other glutamate antagonists through the increase of the number and function of new spine synapses in rat prefrontal cortex by the activation of mTOR [27] do not modify hippocampal cell proliferation [264].

It would also be critical for future work to validate the relative importance of antidepressant-induced neurogenesis and synaptic plasticity in the antidepressant effects. However, evidence is strong that neurogenesis is required for at least some of the beneficial effects of antidepressant treatment. The exact role of neuroplastic/neuroproliferative changes in other brain structures as mPFCx and amygdala should be elucidated.

5. Conclusion

As indicated in this review, the importance of either proliferation or plasticity, or both, is still a matter of debate. As the involvement of proliferation and plasticity has been mainly studied in hippocampus, we might be underestimating its role in the antidepressant effect. In this sense, as the hippocampus

is responsible for the learning and cognition part of the depressive disorder, the fact that the impairment of hippocampal proliferation would not block the antidepressant effect of some drugs does not necessarily conclude that the proliferation is only dependent on hippocampus. In the last years, prefrontal cortex, a structure with a great importance in mood control and working memory, is gaining increasing relevance in the plastic changes linked to antidepressant effects promoted by drugs as ketamine. In this sense, hippocampal proliferation would be only a small part of the plastic changes that are taking place within the hippocampus, and other brain areas. Thus, we must not underestimate the implication of synaptic plasticity in those antidepressant treatments that are not accompanied with increased proliferation.

Acknowledgments

The authors wish to thank Rebeca Madureira, Alicia Martín, Beatriz Romero, Isabel Ruiz, María Josefa Castillo, and Lourdes Lanza for their technical assistance. The scientific work of former coworkers Olga Guitérrez, Susana Mato, Elena del Olmo, Antonio Rodríguez-Gaztelumendi, and María Luisa Rojo is kindly acknowledged. This research was supported by Ministry of Science, SAF04-00941, SAF07-61862, Fundación Alicia Koplowitz, Fundación de Investigación Médica Mutua Madrileña, Instituto de Salud Carlos III, and University of Cantabria-FAES research contract. Rebecca Vidal has been the recipient of a fellowship from University of Cantabria-FAES, and a CIBERSAM contract.

References

[1] WHO, "The world health report," 2001.

[2] American Psychiatric Association, *Diagnostic and Statistical Manual of Mental Disorders (DSM-IV)*, APA Press, 1994.

[3] L. M. Bylsma, B. H. Morris, and J. Rottenberg, "A meta-analysis of emotional reactivity in major depressive disorder," *Clinical Psychology Review*, vol. 28, no. 4, pp. 676–691, 2008.

[4] M. T. Treadway and D. H. Zald, "Reconsidering anhedonia in depression: lessons from translational neuroscience," *Neuroscience and Biobehavioral Reviews*, vol. 35, no. 3, pp. 537–555, 2011.

[5] M. Koenigs and J. Grafman, "The functional neuroanatomy of depression: distinct roles for ventromedial and dorsolateral prefrontal cortex," *Behavioural Brain Research*, vol. 201, no. 2, pp. 239–243, 2009.

[6] J. L. Price and W. C. Drevets, "Neurocircuitry of mood disorders," *Neuropsychopharmacology*, vol. 35, no. 1, pp. 192–216, 2010.

[7] J. L. Price and W. C. Drevets, "Neural circuits underlying the pathophysiology of mood disorders," *Trends in Cognitive Sciences*, vol. 16, pp. 61–71, 2012.

[8] J. Pascual-Brazo, E. Castro, A. Diaz et al., "Modulation of neuroplasticity pathways and antidepressant-like behavioural responses following the short-term (3 and 7 days) administration of the 5-HT(4) receptor agonist RS67333," *International Journal of Neuropsychopharmacology*, vol. 15, no. 5, pp. 631–643, 2012.

[9] L. Xia, C. Delomenie, I. David et al., "Ventral hippocampal molecular pathways and impaired neurogenesis associated with 5-HT(1)A and 5-HT(1)B receptors disruption in mice," *Neuroscience Letters*, vol. 521, no. 1, pp. 20–25, 2012.

[10] R. Mostany, E. M. Valdizán, and A. Pazos, "A role for nuclear β-catenin in SNRI antidepressant-induced hippocampal cell proliferation," *Neuropharmacology*, vol. 55, no. 1, pp. 18–26, 2008.

[11] D. Tardito, J. Perez, E. Tiraboschi, L. Musazzi, G. Racagni, and M. Popoli, "Signaling pathways regulating gene expression, neuroplasticity, and neurotrophic mechanisms in the action of antidepressants: a critical overview," *Pharmacological Reviews*, vol. 58, no. 1, pp. 115–134, 2006.

[12] E. J. Nestler, M. Barrot, R. J. DiLeone, A. J. Eisch, S. J. Gold, and L. M. Monteggia, "Neurobiology of depression," *Neuron*, vol. 34, no. 1, pp. 13–25, 2002.

[13] O. Berton and E. J. Nestler, "New approaches to antidepressant drug discovery: beyond monoamines," *Nature Reviews Neuroscience*, vol. 7, no. 2, pp. 137–151, 2006.

[14] A. Caspi, K. Sugden, T. E. Moffitt et al., "Influence of life stress on depression: moderation by a polymorphism in the 5-HTT gene," *Science*, vol. 301, no. 5631, pp. 386–389, 2003.

[15] T. A. Ban, "Pharmacotherapy of depression: a historical analysis," *Journal of Neural Transmission*, vol. 108, no. 6, pp. 707–716, 2001.

[16] J. J. Schildkraut, "The catecholamine hypothesis of affective disorders: a review of supporting evidence," *The American Journal of Psychiatry*, vol. 122, no. 5, pp. 509–522, 1965.

[17] H. G. Ruhé, N. S. Mason, and A. H. Schene, "Mood is indirectly related to serotonin, norepinephrine and dopamine levels in humans: a meta-analysis of monoamine depletion studies," *Molecular Psychiatry*, vol. 12, no. 4, pp. 331–359, 2007.

[18] R. S. Duman, G. R. Heninger, and E. J. Nestler, "A molecular and cellular theory of depression," *Archives of General Psychiatry*, vol. 54, no. 7, pp. 597–606, 1997.

[19] R. S. Duman, J. Malberg, and J. Thome, "Neural plasticity to stress and antidepressant treatment," *Biological Psychiatry*, vol. 46, no. 9, pp. 1181–1191, 1999.

[20] B. Czéh, T. Michaelis, T. Watanabe et al., "Stress-induced changes in cerebral metabolites, hippocampal volume, and cell proliferation are prevented by antidepressant treatment with tianeptine," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 98, no. 22, pp. 12796–12801, 2001.

[21] J. E. Malberg and R. S. Duman, "Cell proliferation in adult hippocampus is decreased by inescapable stress: reversal by fluoxetine treatment," *Neuropsychopharmacology*, vol. 28, no. 9, pp. 1562–1571, 2003.

[22] J. L. Warner-Schmidt and R. S. Duman, "Hippocampal neurogenesis: opposing effects of stress and antidepressant treatment," *Hippocampus*, vol. 16, no. 3, pp. 239–249, 2006.

[23] A. Sahay and R. Hen, "Adult hippocampal neurogenesis in depression," *Nature Neuroscience*, vol. 10, no. 9, pp. 1110–1115, 2007.

[24] A. Sahay, M. R. Drew, and R. Hen, "Dentate gyrus neurogenesis and depression," *Progress in Brain Research*, vol. 163, pp. 697–722, 2007.

[25] C. H. Duman, L. Schlesinger, D. S. Russell, and R. S. Duman, "Voluntary exercise produces antidepressant and anxiolytic behavioral effects in mice," *Brain Research C*, vol. 1199, pp. 148–158, 2008.

[26] T. M. Madsen, S. S. Newton, M. E. Eaton, D. S. Russell, and R. S. Duman, "Chronic electroconvulsive seizure up-regulates

β-catenin expression in rat hippocampus: role in adult neurogenesis," *Biological Psychiatry*, vol. 54, no. 10, pp. 1006–1014, 2003.

[27] N. Li, B. Lee, R. J. Liu et al., "mTOR-dependent synapse formation underlies the rapid antidepressant effects of NMDA antagonists," *Science*, vol. 329, no. 5994, pp. 959–964, 2010.

[28] H. J. Lee, J. W. Kim, S. V. Yim et al., "Fluoxetine enhances cell proliferation and prevents apoptosis in dentate gyrus of maternally separated rats," *Molecular Psychiatry*, vol. 6, no. 6, pp. 725–728, 2001.

[29] V. M. Heine, S. Maslam, J. Zareno, M. Joëls, and P. J. Lucassen, "Suppressed proliferation and apoptotic changes in the rat dentate gyrus after acute and chronic stress are reversible," *European Journal of Neuroscience*, vol. 19, no. 1, pp. 131–144, 2004.

[30] K. Jaako-Movits, T. Zharkovsky, M. Pedersen, and A. Zharkovsky, "Decreased hippocampal neurogenesis following olfactory bulbectomy is reversed by repeated citalopram administration," *Cellular and Molecular Neurobiology*, vol. 26, no. 7-8, pp. 1559–1570, 2006.

[31] Y. Xu, P. A. Barish, J. Pan, W. O. Ogle, and J. M. O'Donnell, "Animal models of depression and neuroplasticity: assessing drug action in relation to behavior and neurogenesis," *Methods in Molecular Biology*, vol. 829, pp. 103–124, 2012.

[32] Y. I. Sheline, P. W. Wang, M. H. Gado, J. G. Csernansky, and M. W. Vannier, "Hippocampal atrophy in recurrent major depression," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 93, no. 9, pp. 3908–3913, 1996.

[33] J. D. Bremner, M. Narayan, E. R. Anderson, L. H. Staib, H. L. Miller, and D. S. Charney, "Hippocampal volume reduction in major depression," *American Journal of Psychiatry*, vol. 157, no. 1, pp. 115–117, 2000.

[34] C. A. Stockmeier, G. J. Mahajan, L. C. Konick et al., "Cellular changes in the postmortem hippocampus in major depression," *Biological Psychiatry*, vol. 56, no. 9, pp. 640–650, 2004.

[35] J. Beauquis, P. Roig, F. Homo-Delarche, A. De Nicola, and F. Saravia, "Reduced hippocampal neurogenesis and number of hilar neurones in streptozotocin-induced diabetic mice: reversion by antidepressant treatment," *European Journal of Neuroscience*, vol. 23, no. 6, pp. 1539–1546, 2006.

[36] B. Czéh, J. I. H. Müller-Keuker, R. Rygula et al., "Chronic social stress inhibits cell proliferation in the adult medial prefrontal cortex: hemispheric asymmetry and reversal by fluoxetine treatment," *Neuropsychopharmacology*, vol. 32, no. 7, pp. 1490–1503, 2007.

[37] M. Tochigi, K. Iwamoto, M. Bundo, T. Sasaki, N. Kato, and T. Kato, "Gene expression profiling of major depression and suicide in the prefrontal cortex of postmortem brains," *Neuroscience Research*, vol. 60, no. 2, pp. 184–191, 2008.

[38] R. M. Tordera, A. L. Garcia-García, N. Elizalde et al., "Chronic stress and impaired glutamate function elicit a depressive-like phenotype and common changes in gene expression in the mouse frontal cortex," *European Neuropsychopharmacology*, vol. 21, no. 1, pp. 23–32, 2011.

[39] A. Blugeot, C. Rivat, E. Bouvier et al., "Vulnerability to depression: from brain neuroplasticity to identification of biomarkers," *Journal of Neuroscience*, vol. 31, no. 36, pp. 12889–12899, 2011.

[40] M. Kodama, T. Fujioka, and R. S. Duman, "Chronic olanzapine or fluoxetine administration increases cell proliferation in hippocampus and prefrontal cortex of adult rat," *Biological Psychiatry*, vol. 56, no. 8, pp. 570–580, 2004.

[41] J. E. Castro, E. Varea, C. Márquez, M. I. Cordero, G. Poirier, and C. Sandi, "Role of the amygdala in antidepressant effects on hippocampal cell proliferation and survival and on depression-like behavior in the rat," *PLoS ONE*, vol. 5, no. 1, Article ID e8618, 2010.

[42] D. M. Diamond, A. Campbell, C. R. Park, and R. M. Vouimba, "Preclinical research on stress, memory, and the brain in the development of pharmacotherapy for depression," *European Neuropsychopharmacology*, vol. 14, supplement 5, pp. S491–S495, 2004.

[43] J. Thome, N. Sakai, K. H. Shin et al., "cAMP response element-mediated gene transcription is upregulated by chronic antidepressant treatment," *Journal of Neuroscience*, vol. 20, no. 11, pp. 4030–4036, 2000.

[44] A. C. H. Chen, Y. Shirayama, K. H. Shin, R. L. Neve, and R. S. Duman, "Expression of the cAMP response element binding protein (CREB) in hippocampus produces an antidepressant effect," *Biological Psychiatry*, vol. 49, no. 9, pp. 753–762, 2001.

[45] P. Penzes, M. E. Cahill, K. A. Jones, J. E. Vanleeuwen, and K. M. Woolfrey, "Dendritic spine pathology in neuropsychiatric disorders," *Nature Neuroscience*, vol. 14, no. 3, pp. 285–293, 2011.

[46] S. C. Cook and C. L. Wellman, "Chronic stress alters dendritic morphology in rat medial prefrontal cortex," *Journal of Neurobiology*, vol. 60, no. 2, pp. 236–248, 2004.

[47] J. J. Radley, H. M. Sisti, J. Hao et al., "Chronic behavioral stress induces apical dendritic reorganization in pyramidal neurons of the medial prefrontal cortex," *Neuroscience*, vol. 125, no. 1, pp. 1–6, 2004.

[48] D. Öngür, W. C. Drevets, and J. L. Price, "Glial reduction in the subgenual prefrontal cortex in mood disorders," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 95, no. 22, pp. 13290–13295, 1998.

[49] M. P. Bowley, W. C. Drevets, D. Öngür, and J. L. Price, "Low glial numbers in the amygdala in major depressive disorder," *Biological Psychiatry*, vol. 52, no. 5, pp. 404–412, 2002.

[50] G. Rajkowska, J. J. Miguel-Hidalgo, J. Wei et al., "Morphometric evidence for neuronal and glial prefrontal cell pathology in major depression," *Biological Psychiatry*, vol. 45, no. 9, pp. 1085–1098, 1999.

[51] M. Popoli, Z. Yan, B. S. McEwen, and G. Sanacora, "The stressed synapse: the impact of stress and glucocorticoids on glutamate transmission," *Nature Reviews Neuroscience*, vol. 13, pp. 22–37, 2012.

[52] J. J. Cerqueira, F. Mailliet, O. F. X. Almeida, T. M. Jay, and N. Sousa, "The prefrontal cortex as a key target of the maladaptive response to stress," *Journal of Neuroscience*, vol. 27, no. 11, pp. 2781–2787, 2007.

[53] P. J. Lucassen, M. B. Müller, F. Holsboer et al., "Hippocampal apoptosis in major depression is a minor event and absent from subareas at risk for glucocorticoid overexposure," *American Journal of Pathology*, vol. 158, no. 2, pp. 453–468, 2001.

[54] A. Bachis, M. I. Cruz, R. L. Nosheny, and I. Mocchetti, "Chronic unpredictable stress promotes neuronal apoptosis in the cerebral cortex," *Neuroscience Letters*, vol. 442, no. 2, pp. 104–108, 2008.

[55] T. A. Kosten, M. P. Galloway, R. S. Duman, D. S. Russell, and C. D'Sa, "Repeated unpredictable stress and antidepressants differentially regulate expression of the Bcl-2 family of apoptotic genes in rat cortical, hippocampal, and limbic brain structures," *Neuropsychopharmacology*, vol. 33, no. 7, pp. 1545–1558, 2008.

[56] M. Nibuya, S. Morinobu, and R. S. Duman, "Regulation of BDNF and trkB mRNA in rat brain by chronic electroconvulsive seizure and antidepressant drug treatments," *Journal of Neuroscience*, vol. 15, no. 11, pp. 7539–7547, 1995.

[57] T. Saarelainen, P. Hendolin, G. Lucas et al., "Activation of the TrkB neurotrophin receptor is induced by antidepressant drugs and is required for antidepressant-induced behavioral effects," *Journal of Neuroscience*, vol. 23, no. 1, pp. 349–357, 2003.

[58] P. J. Lucassen, E. Fuchs, and B. Czéh, "Antidepressant treatment with tianeptine reduces apoptosis in the hippocampal dentate gyrus and temporal cortex," *Biological Psychiatry*, vol. 55, no. 8, pp. 789–796, 2004.

[59] S. Ueda, S. Sakakibara, and K. Yoshimoto, "Effect of long-lasting serotonin depletion on environmental enrichment-induced neurogenesis in adult rat hippocampus and spatial learning," *Neuroscience*, vol. 135, no. 2, pp. 395–402, 2005.

[60] J. E. Malberg, A. J. Eisch, E. J. Nestler, and R. S. Duman, "Chronic antidepressant treatment increases neurogenesis in adult rat hippocampus," *Journal of Neuroscience*, vol. 20, no. 24, pp. 9104–9110, 2000.

[61] M. Sairanen, G. Lucas, P. Ernfors, M. Castrén, and E. Castrén, "Brain-derived neurotrophic factor and antidepressant drugs have different but coordinated effects on neuronal turnover, proliferation, and survival in the adult dentate gyrus," *Journal of Neuroscience*, vol. 25, no. 5, pp. 1089–1094, 2005.

[62] R. Alonso, G. Griebel, G. Pavone, J. Stemmelin, G. Le Fur, and P. Soubrié, "Blockade of CRF1 or V1b receptors reverses stress-induced suppression of neurogenesis in a mouse model of depression," *Molecular Psychiatry*, vol. 9, no. 3, pp. 278–286, 2004.

[63] G. Lucas, J. Du, T. Romeas et al., "Selective serotonin reuptake inhibitors potentiate the rapid antidepressant-like effects of serotonin4 receptor agonists in the rat," *PLoS ONE*, vol. 5, no. 2, Article ID e9253, 2010.

[64] G. Lucas, V. V. Rymar, J. Du et al., "Serotonin4 (5-HT4) receptor agonists are putative antidepressants with a rapid onset of action," *Neuron*, vol. 55, no. 5, pp. 712–725, 2007.

[65] A. Tamburella, V. Micale, A. Navarria, and F. Drago, "Antidepressant properties of the 5-HT4 receptor partial agonist, SL65.0155: behavioral and neurochemical studies in rats," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 33, no. 7, pp. 1205–1210, 2009.

[66] A. Ferres-Coy, F. Pilar-Cuellar, R. Vidal et al., "RNAi-mediated serotonin transporter suppression rapidly increases serotonergic neurotransmission and hippocampal neurogenesis," *Translational Psychiatry*, vol. 3, article e211, 2013.

[67] O. F. O'Leary, S. Zandy, T. G. Dinan, and J. F. Cryan, "Lithium augmentation of the effects of desipramine in a mouse model of treatment-resistant depression: a role for hippocampal cell proliferation," *Neuroscience*, vol. 228, pp. 36–46, 2013.

[68] K. Huo, Y. Sun, H. Li et al., "Lithium reduced neural progenitor apoptosis in the hippocampus and ameliorated functional deficits after irradiation to the immature mouse brain," *Molecular and Cellular Neuroscience*, vol. 51, pp. 32–42, 2012.

[69] J. M. Encinas, A. Vaahtokari, and G. Enikolopov, "Fluoxetine targets early progenitor cells in the adult brain," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 103, no. 21, pp. 8233–8238, 2006.

[70] E. Segi-Nishida, J. L. Warner-Schmidt, and R. S. Duman, "Electroconvulsive seizure and VEGF increase the proliferation of neural stem-like cells in rat hippocampus," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 32, pp. 11352–11357, 2008.

[71] L. Santarelli, M. Saxe, C. Gross et al., "Requirement of hippocampal neurogenesis for the behavioral effects of antidepressants," *Science*, vol. 301, no. 5634, pp. 805–809, 2003.

[72] J. E. Malberg and L. E. Schecter, "Increasing hippocampal neurogenesis: a novel mechanism for antidepressant drugs," *Current Pharmaceutical Design*, vol. 11, no. 2, pp. 145–155, 2005.

[73] A. Dranovsky and R. Hen, "Hippocampal eurogenesis: regulation by stress and antidepressants," *Biological Psychiatry*, vol. 59, no. 12, pp. 1136–1143, 2006.

[74] M. Banasr, M. Hery, R. Printemps, and A. Daszuta, "Serotonin-induced increases in adult cell proliferation and neurogenesis are mediated through different and common 5-HT receptor subtypes in the dentate gyrus and the subventricular zone," *Neuropsychopharmacology*, vol. 29, no. 3, pp. 450–460, 2004.

[75] J. W. Wang, D. J. David, J. E. Monckton, F. Battaglia, and R. Hen, "Chronic fluoxetine stimulates maturation and synaptic plasticity of adult-born hippocampal granule cells," *Journal of Neuroscience*, vol. 28, no. 6, pp. 1374–1384, 2008.

[76] A. Surget, M. Saxe, S. Leman et al., "Drug-dependent requirement of hippocampal neurogenesis in a model of depression and of antidepressant reversal," *Biological Psychiatry*, vol. 64, no. 4, pp. 293–301, 2008.

[77] S. A. Arnold and T. Hagg, "Serotonin 1A receptor agonist increases species- and region-selective adult CNS proliferation, but not through CNTF," *Neuropharmacology*, vol. 63, pp. 1238–1247, 2012.

[78] F. Pilar-Cuéllar, R. Vidal, A. Díaz et al., "Pathways involved in antidepressant-induced proliferationand synaptic plasticity," *Current Pharmaceutical Design*. In press.

[79] G. J. Huang and J. Herbert, "The role of 5-HT1A receptors in the proliferation and survival of progenitor cells in the dentate gyrus of the adult hippocampus and their regulation by corticoids," *Neuroscience*, vol. 135, no. 3, pp. 803–813, 2005.

[80] A. D. Fricker, C. Rios, L. A. Devi, and I. Gomes, "Serotonin receptor activation leads to neurite outgrowth and neuronal survival," *Molecular Brain Research*, vol. 138, no. 2, pp. 228–235, 2005.

[81] S. Jha, R. Rajendran, K. A. Fernandes, and V. A. Vaidya, "5-HT2A/2C receptor blockade regulates progenitor cell proliferation in the adult rat hippocampus," *Neuroscience Letters*, vol. 441, no. 2, pp. 210–214, 2008.

[82] A. Soumier, M. Banasr, S. Lortet et al., "Mechanisms contributing to the phase-dependent regulation of neurogenesis by the novel antidepressant, agomelatine, in the adult rat hippocampus," *Neuropsychopharmacology*, vol. 34, no. 11, pp. 2390–2403, 2009.

[83] F. Pilar-Cuellar, R. Vidal, and A. Pazos, "Subchronic treatment with fluoxetine and ketanserin increases hippocampal brain-derived neurotrophic factor, beta-catenin and antidepressant-like effects," *British Journal of Pharmacology*, vol. 165, pp. 1046–1057, 2012.

[84] A. G. Foley, W. D. Hirst, H. C. Gallagher et al., "The selective 5-HT6 receptor antagonists SB-271046 and SB-399885 potentiate NCAM PSA immunolabeling of dentate granule cells, but not neurogenesis, in the hippocampal formation of mature Wistar rats," *Neuropharmacology*, vol. 54, no. 8, pp. 1166–1174, 2008.

[85] O. Mnie-Filali, C. Faure, L. Lambas-Senas et al., "Pharmacological blockade of 5-HT7 receptors as a putative fast acting antidepressant strategy," *Neuropsychopharmacology*, vol. 36, no. 6, pp. 1275–1288, 2011.

[86] G. Sarkisyan and P. B. Hedlund, "The 5-HT7 receptor is involved in allocentric spatial memory information processing," *Behavioural Brain Research*, vol. 202, no. 1, pp. 26–31, 2009.

[87] J. A. Menninger and B. Tabakoff, "Forskolin-stimulated platelet adenylyl cyclase activity is lower in persons with major depression," *Biological Psychiatry*, vol. 42, no. 1, pp. 30–38, 1997.

[88] E. M. Valdizán, O. Gutierrez, and A. Pazos, "Adenylate cyclase activity in postmortem brain of suicide subjects: reduced response to β-adrenergic stimulation," *Biological Psychiatry*, vol. 54, no. 12, pp. 1457–1464, 2003.

[89] I. Extein, J. Tallman, C. C. Smith, and F. K. Goodwin, "Changes in lymphocyte beta-adrenergic receptors in depression and mania," *Psychiatry Research*, vol. 1, no. 2, pp. 191–197, 1979.

[90] J. P. Halper, R. P. Brown, J. A. Sweeney, J. H. Kocsis, A. Peters, and J. J. Mann, "Blunted β-adrenergic responsivity of peripheral blood mononuclear cells in endogenous depression. Isoproterenol dose-response studies," *Archives of General Psychiatry*, vol. 45, no. 3, pp. 241–244, 1988.

[91] J. J. Mann, J. P. Halper, P. J. Wilner et al., "Subsensitivity of adenylyl cyclase-coupled receptors on mononuclear leukocytes from drug-free inpatients with a major depressive episode," *Biological Psychiatry*, vol. 42, no. 10, pp. 859–870, 1997.

[92] P. Mazzola-Pomietto, J. M. Azorin, V. Tramoni, and R. Jeanningros, "Relation between lymphocyte beta-adrenergic responsivity and the severity of depressive disorders," *Biological Psychiatry*, vol. 35, pp. 920–925, 1994.

[93] G. N. Pandey, P. Sudershan, and J. M. Davis, "Beta adrenergic receptor function in depression and the effect of antidepressant drugs," *Acta Pharmacologica et Toxicologica Supplementum*, vol. 56, no. 1, pp. 66–79, 1985.

[94] E. M. Valdizn, R. Dez-Alarcia, J. Gonzlez-Maeso et al., "α2-adrenoceptor functionality in postmortem frontal cortex of depressed suicide victims," *Biological Psychiatry*, vol. 68, no. 9, pp. 869–872, 2010.

[95] H. Ozawa and M. M. Rasenick, "Chronic electroconvulsive treatment augments coupling of the GTP-binding protein G(s) to the catalytic moiety of adenylyl cyclase in a manner similar to that seen with chronic antidepressant drugs," *Journal of Neurochemistry*, vol. 56, no. 1, pp. 330–338, 1991.

[96] V. J. Watts and K. A. Neve, "Sensitization of adenylate cyclase by Galpha i/o-coupled receptors," *Pharmacology & Therapeutics*, vol. 106, pp. 405–421, 2005.

[97] R. Vidal, E. M. Valdizan, R. Mostany, A. Pazos, and E. Castro, "Long-term treatment with fluoxetine induces desensitization of 5-HT4 receptor-dependent signalling and functionality in rat brain," *Journal of Neurochemistry*, vol. 110, no. 3, pp. 1120–1127, 2009.

[98] R. Vidal, E. M. Valdizan, M. T. Vilaro, A. Pazos, and E. Castro, "Reduced signal transduction by 5-HT4 receptors after long-term venlafaxine treatment in rats," *British Journal of Pharmacology*, vol. 161, no. 3, pp. 695–706, 2010.

[99] H. T. Zhang, "Cyclic AMP-specific phosphodiesterase-4 as a target for the development of antidepressant drugs," *Current Pharmaceutical Design*, vol. 15, no. 14, pp. 1688–1698, 2009.

[100] S. Mato, R. Vidal, E. Castro, A. Díaz, A. Pazos, and E. M. Valdizán, "Long-term fluoxetine treatment modulates cannabinoid type 1 receptor-mediated inhibition of adenylyl cyclase in the rat prefrontal cortex through 5-hydroxytryptamine 1A receptor-dependent mechanisms," *Molecular Pharmacology*, vol. 77, no. 3, pp. 424–434, 2010.

[101] E. J. Nestler, R. Z. Terwilliger, and R. S. Duman, "Chronic antidepressant administration alters the subcellular distribution of cyclic AMP-dependent protein kinase in rat frontal cortex," *Journal of Neurochemistry*, vol. 53, no. 5, pp. 1644–1647, 1989.

[102] J. Alfonso, L. R. Frick, D. M. Silberman, M. L. Palumbo, A. M. Genaro, and A. C. Frasch, "Regulation of hippocampal gene expression is conserved in two species subjected to different stressors and antidepressant treatments," *Biological Psychiatry*, vol. 59, no. 3, pp. 244–251, 2006.

[103] S. Morley-Fletcher, J. Mairesse, A. Soumier et al., "Chronic agomelatine treatment corrects behavioral, cellular, and biochemical abnormalities induced by prenatal stress in rats," *Psychopharmacology*, vol. 217, no. 3, pp. 301–313, 2011.

[104] D. Dowlatshahi, G. M. MacQueen, J. F. Wang, and L. T. Young, "Increased temporal cortex CREB concentrations and antidepressant treatment in major depression," *Lancet*, vol. 352, no. 9142, pp. 1754–1755, 1998.

[105] D. Dowlatshahi, G. M. MacQueen, J. F. Wang, J. S. Reiach, and L. T. Young, "G protein-coupled cyclic AMP signaling in postmortem brain of subjects with mood disorders: effects of diagnosis, suicide, and treatment at the time of death," *Journal of Neurochemistry*, vol. 73, no. 3, pp. 1121–1126, 1999.

[106] Y. Odagaki, J. A. García-Sevilla, P. Huguelet, R. La Harpe, T. Koyama, and J. Guimón, "Cyclic AMP-mediated signaling components are upregulated in the prefrontal cortex of depressed suicide victims," *Brain Research*, vol. 898, no. 2, pp. 224–231, 2001.

[107] S. Yamada, M. Yamamoto, H. Ozawa, P. Riederer, and T. Saito, "Reduced phosphorylation of cyclic AMP-responsive element binding protein in the postmortem orbitofrontal cortex of patients with major depressive disorder," *Journal of Neural Transmission*, vol. 110, no. 6, pp. 671–680, 2003.

[108] D. Laifenfeld, R. Karry, E. Grauer, E. Klein, and D. Ben-Shachar, "Antidepressants and prolonged stress in rats modulate CAM-L1, laminin, and pCREB, implicated in neuronal plasticity," *Neurobiology of Disease*, vol. 20, no. 2, pp. 432–441, 2005.

[109] T. L. Wallace, K. E. Stellitano, R. L. Neve, and R. S. Duman, "Effects of cyclic adenosine monophosphate response element binding protein overexpression in the basolateral amygdala on behavioral models of depression and anxiety," *Biological Psychiatry*, vol. 56, no. 3, pp. 151–160, 2004.

[110] D. H. Manier, R. C. Shelton, and F. Sulser, "Noradrenergic antidepressants: does chronic treatment increase or decrease nuclear CREB-P?" *Journal of Neural Transmission*, vol. 109, no. 1, pp. 91–99, 2002.

[111] A. M. Pliakas, R. R. Carlson, R. L. Neve, C. Konradi, E. J. Nestler, and W. A. Carlezon, "Altered responsiveness to cocaine and increased immobility in the forced swim test associated with elevated cAMP response element-binding protein expression in nucleus accumbens," *Journal of Neuroscience*, vol. 21, no. 18, pp. 7397–7403, 2001.

[112] Y. Dwivedi, A. C. Mondal, H. S. Rizavi et al., "Differential and brain region-specific regulation of Rap-1 and Epac in depressed suicide victims," *Archives of General Psychiatry*, vol. 63, no. 6, pp. 639–648, 2006.

[113] R. Liu, W. Dang, M. Jianting et al., "Citalopram alleviates chronic stress induced depression-like behaviors in rats by activating GSK3 beta signaling in dorsal hippocampus," *Brain Research*, vol. 1467, pp. 10–17, 2012.

[114] G. Chen, L. D. Huang, Y. M. Jiang, and H. K. Manji, "The mood-stabilizing agent valproate inhibits the activity of glycogen synthase kinase-3," *Journal of Neurochemistry*, vol. 72, no. 3, pp. 1327–1330, 1999.

[115] T. D. Gould, G. Chen, and H. K. Manji, "In vivo evidence in the brain for lithium inhibition of glycogen synthase kinase-3," *Neuropsychopharmacology*, vol. 29, no. 1, pp. 32–38, 2004.

[116] X. Li, W. Zhu, M. S. Roh, A. B. Friedman, K. Rosborough, and R. S. Jope, "In vivo regulation of glycogen synthase kinase-3β (GSK3β) by serotonergic activity in mouse brain," *Neuropsychopharmacology*, vol. 29, no. 8, pp. 1426–1431, 2004.

[117] J. M. Beaulieu, X. Zhang, R. M. Rodriguiz et al., "Role of GSK3β in behavioral abnormalities induced by serotonin deficiency," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 4, pp. 1333–1338, 2008.

[118] O. Kaidanovich-Beilin, A. Milman, A. Weizman, C. G. Pick, and H. Eldar-Finkelman, "Rapid antidepressive-like activity of specific glycogen synthase kinase-3 inhibitor and its effect on β-catenin in mouse hippocampus," *Biological Psychiatry*, vol. 55, no. 8, pp. 781–784, 2004.

[119] W. T. O'Brien, A. D. Harper, F. Jové et al., "Glycogen synthase kinase-3β haploinsufficiency mimics the behavioral and molecular effects of lithium," *Journal of Neuroscience*, vol. 24, no. 30, pp. 6791–6798, 2004.

[120] C. Latapy, V. Rioux, M. J. Guitton, and J. M. Beaulieu, "Selective deletion of forebrain glycogen synthase kinase 3beta reveals a central role in serotonin-sensitive anxiety and social behaviour," *Philosophical Transactions of the Royal Society B*, vol. 367, pp. 2460–2474, 2012.

[121] A. Polter, E. Beurel, S. Yang et al., "Deficiency in the inhibitory serine-phosphorylation of glycogen synthase kinase-3 increases sensitivity to mood disturbances," *Neuropsychopharmacology*, vol. 35, no. 8, pp. 1761–1774, 2010.

[122] F. Benedetti, A. Serretti, C. Colombo, C. Lorenzi, V. Tubazio, and E. Smeraldi, "A glycogen synthase kinase 3-β promoter gene single nucleotide polymorphism is associated with age at onset and response to total sleep deprivation in bipolar depression," *Neuroscience Letters*, vol. 368, no. 2, pp. 123–126, 2004.

[123] F. Benedetti, S. Dallaspezia, C. Lorenzi et al., "Gene-gene interaction of glycogen synthase kinase 3-beta and serotonin transporter on human antidepressant response to sleep deprivation," *Journal of Affective Disorders*, vol. 136, pp. 514–519, 2012.

[124] F. Karege, N. Perroud, S. Burkhardt et al., "Protein levels of beta-catenin and activation state of glycogen synthase kinase-3beta in major depression. A study with postmortem prefrontal cortex," *Journal of Affective Disorders*, vol. 136, pp. 185–188, 2012.

[125] T. D. Gould, K. C. O'Donnell, A. M. Picchini, E. R. Dow, G. Chen, and H. K. Manji, "Generation and behavioral characterization of β-catenin forebrain-specific conditional knock-out mice," *Behavioural Brain Research*, vol. 189, no. 1, pp. 117–125, 2008.

[126] T. D. Gould, H. Einat, K. C. O'Donnell, A. M. Picchini, R. J. Schloesser, and H. K. Manji, "B-catenin overexpression in the mouse brain phenocopies lithium-sensitive behaviors," *Neuropsychopharmacology*, vol. 32, no. 10, pp. 2173–2183, 2007.

[127] K. R. Howell, A. Kutiyanawalla, and A. Pillai, "Long-term continuous corticosterone treatment decreases VEGF receptor-2 expression in frontal cortex," *PLoS ONE*, vol. 6, no. 5, Article ID e20198, 2011.

[128] L. Sui, J. Wang, and B. M. Li, "Role of the phosphoinositide 3-kinase-Akt-mammalian target of the rapamycin signaling pathway in long-term potentiation and trace fear conditioning memory in rat medial prefrontal cortex," *Learning and Memory*, vol. 15, no. 10, pp. 762–776, 2008.

[129] A. Chandran, A. H. Iyo, C. S. Jernigan, B. Legutko, M. C. Austin, and B. Karolewicz, "Reduced phosphorylation of the mTOR signaling pathway components in the amygdala of rats exposed to chronic stress," *Progress in Neuro-Psychopharmacology & Biological Psychiatry*, vol. 40, pp. 240–245, 2013.

[130] J. J. Yu, Y. Zhang, Y. Wang et al., "Inhibition of calcineurin in the prefrontal cortex induced depressive-like behavior through mTOR signaling pathway," *Psychopharmacology*, vol. 225, pp. 361–372, 2013.

[131] S. J. Mathew, K. Keegan, and L. Smith, "Glutamate modulators as novel interventions for mood disorders," *Revista Brasileira de Psiquiatria*, vol. 27, no. 3, pp. 243–248, 2005.

[132] J. M. Brezun and A. Daszuta, "Serotonergic reinnervation reverses lesion-induced decreases in PSA-NCAM labeling and proliferation of hippocampal cells in adult rats," *Hippocampus*, vol. 10, pp. 37–46, 2000.

[133] D. S. Cowen, L. F. Takase, C. A. Fornal, and B. L. Jacobs, "Age-dependent decline in hippocampal neurogenesis is not altered by chronic treatment with fluoxetine," *Brain Research C*, vol. 1228, pp. 14–19, 2008.

[134] M. W. Marlatt, P. J. Lucassen, and H. van Praag, "Comparison of neurogenic effects of fluoxetine, duloxetine and running in mice," *Brain Research C*, vol. 1341, pp. 93–99, 2010.

[135] N. D. Hanson, C. B. Nemeroff, and M. J. Owens, "Lithium, but not fluoxetine or the corticotropin-releasing factor receptor 1 receptor antagonist R121919, increases cell proliferation in the adult dentate gyrus," *Journal of Pharmacology and Experimental Therapeutics*, vol. 337, no. 1, pp. 180–186, 2011.

[136] J. J. Radley and B. L. Jacobs, "5-HT1A receptor antagonist administration decreases cell proliferation in the dentate gyrus," *Brain Research*, vol. 955, no. 1-2, pp. 264–267, 2002.

[137] G. J. Marek, L. L. Carpenter, C. J. McDougle, and L. H. Price, "Synergistic action of 5-HT2A antagonists and selective serotonin reuptake inhibitors in neuropsychiatric disorders," *Neuropsychopharmacology*, vol. 28, no. 2, pp. 402–412, 2003.

[138] D. K. Pandey, R. Mahesh, A. A. Kumar, V. S. Rao, M. Arjun, and R. Rajkumar, "A novel 5-HT(2A) receptor antagonist exhibits antidepressant-like effects in a battery of rodent behavioural assays: approaching early-onset antidepressants," *Pharmacology Biochemistry and Behavior*, vol. 94, no. 3, pp. 363–373, 2010.

[139] V. A. Vaidya, R. M. Z. Terwilliger, and R. S. Duman, "Role of 5-HT(2a) receptors in the stress-induced down-regulation of brain-derived neurotrophic factor expression in rat hippocampus," *Neuroscience Letters*, vol. 262, no. 1, pp. 1–4, 1999.

[140] A. I. Gulyas, L. Acsady, and T. F. Freund, "Structural basis of the cholinergic and serotonergic modulation of GABAergic neurons in the hippocampus," *Neurochemistry International*, vol. 34, pp. 359–372, 1999.

[141] P. Rosel, B. Arranz, M. Urretavizcaya, M. Oros, L. San, and M. A. Navarro, "Altered 5-HT2A and 5-HT4 postsynaptic receptors and their intracellular signalling systems IP3 and cAmP in brains from depressed violent suicide victims," *Neuropsychobiology*, vol. 49, no. 4, pp. 189–195, 2004.

[142] A. Wesołowska, "Potential role of the 5-HT6 receptor in depression and anxiety: an overview of preclinical data," *Pharmacological Reports*, vol. 62, no. 4, pp. 564–577, 2010.

[143] A. Wesołowska and A. Nikiforuk, "Effects of the brain-penetrant and selective 5-HT6 receptor antagonist SB-399885 in animal models of anxiety and depression," *Neuropharmacology*, vol. 52, no. 5, pp. 1274–1283, 2007.

[144] A. Wesołowska and A. Nikiforuk, "The selective 5-HT6 receptor antagonist SB-399885 enhances anti-immobility action of antidepressants in rats," *European Journal of Pharmacology*, vol. 582, no. 1–3, pp. 88–93, 2008.

[145] A. Wesołowska, A. Nikiforuk, and K. Stachowicz, "Anxiolytic-like and antidepressant-like effects produced by the selective

5-HT6 receptor antagonist SB-258585 after intrahippocampal administration to rats," *Behavioural Pharmacology*, vol. 18, no. 5-6, pp. 439–446, 2007.

[146] C. K. Callaghan, V. Hok, A. Della-Chiesa, D. J. Virley, N. Upton, and S. M. O'Mara, "Age-related declines in delayed non-match-to-sample performance (DNMS) are reversed by the novel 5HT6 receptor antagonist SB742457," *Neuropharmacology*, vol. 63, pp. 890–897, 2012.

[147] O. Mnie-Filali, L. Lambás-Señas, L. Zimmer, and N. Haddjeri, "5-HT7 receptor antagonists as a new class of antidepressants," *Drug News and Perspectives*, vol. 20, no. 10, pp. 613–618, 2007.

[148] S. M. Stahl, "The serotonin-7 receptor as a novel therapeutic target," *Journal of Clinical Psychiatry*, vol. 71, no. 11, pp. 1414–1415, 2010.

[149] P. B. Hedlund, S. Huitron-Resendiz, S. J. Henriksen, and J. G. Sutcliffe, "5-HT7 receptor inhibition and inactivation induce antidepressantlike behavior and sleep pattern," *Biological Psychiatry*, vol. 58, no. 10, pp. 831–837, 2005.

[150] E. Alvarez, V. Perez, M. Dragheim, H. Loft, and F. Artigas, "A double-blind, randomized, placebo-controlled, active reference study of Lu AA21004 in patients with major depressive disorder," *International Journal of Neuropsychopharmacology*, vol. 15, pp. 589–600, 2012.

[151] F. Calabrese, R. Molteni, G. Racagni, and M. A. Riva, "Neuronal plasticity: a link between stress and mood disorders," *Psychoneuroendocrinology*, vol. 34, supplement 1, pp. S208–S216, 2009.

[152] D. Taliaz, A. Loya, R. Gersner, S. Haramati, A. Chen, and A. Zangen, "Resilience to chronic stress is mediated by hippocampal brain-derived neurotrophic factor," *Journal of Neuroscience*, vol. 31, no. 12, pp. 4475–4483, 2011.

[153] K. Hashimoto, "Brain-derived neurotrophic factor as a biomarker for mood disorders: an historical overview and future directions," *Psychiatry and Clinical Neurosciences*, vol. 64, no. 4, pp. 341–357, 2010.

[154] S. Sen, R. Duman, and G. Sanacora, "Serum brain-derived neurotrophic factor, depression, and antidepressant medications: meta-analyses and implications," *Biological Psychiatry*, vol. 64, no. 6, pp. 527–532, 2008.

[155] W. Umene-Nakano, R. Yoshimura, A. Ikenouchi-Sugita et al., "Serum levels of brain-derived neurotrophic factor in comorbidity of depression and alcohol dependence," *Human Psychopharmacology*, vol. 24, no. 5, pp. 409–413, 2009.

[156] A. S. Gonul, F. Akdeniz, F. Taneli, O. Donat, C. Eker, and S. Vahip, "Effect of treatment on serum brain-derived neurotrophic factor levels in depressed patients," *European Archives of Psychiatry and Clinical Neuroscience*, vol. 255, no. 6, pp. 381–386, 2005.

[157] Y. K. Kim, H. P. Lee, S. D. Won et al., "Low plasma BDNF is associated with suicidal behavior in major depression," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 31, no. 1, pp. 78–85, 2007.

[158] L. M. Monteggia, M. Barrot, C. M. Powell et al., "Essential role of brain-derived neurotrophic factor in adult hippocampal function," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 101, no. 29, pp. 10827–10832, 2004.

[159] Z. Y. Chen, D. Jing, K. G. Bath et al., "Genetic variant BDNF (Val66Met) polymorphism alters anxiety-related behavior," *Science*, vol. 314, no. 5796, pp. 140–143, 2006.

[160] M. Adachi, M. Barrot, A. E. Autry, D. Theobald, and L. M. Monteggia, "Selective loss of brain-derived neurotrophic factor in the dentate gyrus attenuates antidepressant efficacy," *Biological Psychiatry*, vol. 63, no. 7, pp. 642–649, 2008.

[161] J. A. Siuciak, D. R. Lewis, S. J. Wiegand, and R. M. Lindsay, "Antidepressant-like effect of brain-derived neurotrophic factor (BDNF)," *Pharmacology Biochemistry and Behavior*, vol. 56, no. 1, pp. 131–137, 1997.

[162] B. A. Hoshaw, J. E. Malberg, and I. Lucki, "Central administration of IGF-I and BDNF leads to long-lasting antidepressant-like effects," *Brain Research*, vol. 1037, no. 1-2, pp. 204–208, 2005.

[163] Y. Shirayama, A. C. H. Chen, S. Nakagawa, D. S. Russell, and R. S. Duman, "Brain-derived neurotrophic factor produces antidepressant effects in behavioral models of depression," *Journal of Neuroscience*, vol. 22, no. 8, pp. 3251–3261, 2002.

[164] Y. Ye, G. Wang, H. Wang, and X. Wang, "Brain-derived neurotrophic factor (BDNF) infusion restored astrocytic plasticity in the hippocampus of a rat model of depression," *Neuroscience Letters*, vol. 503, pp. 15–19, 2011.

[165] H. D. Schmidt and R. S. Duman, "Peripheral BDNF produces antidepressant-like effects in cellular and behavioral models," *Neuropsychopharmacology*, vol. 35, no. 12, pp. 2378–2391, 2010.

[166] A. Tadić, S. Wagner, K. F. Schlicht et al., "The early non-increase of serum BDNF predicts failure of antidepressant treatment in patients with major depression: a pilot study," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 35, no. 2, pp. 415–420, 2011.

[167] R. S. Duman and L. M. Monteggia, "A neurotrophic model for stress-related mood disorders," *Biological Psychiatry*, vol. 59, no. 12, pp. 1116–1127, 2006.

[168] V. A. Vaidya, G. J. Marek, G. K. Aghajanian, and R. S. Duman, "5-HT(2A) receptor-mediated regulation of brain-derived neurotrophic factor mRNA in the hippocampus and the neocortex," *Journal of Neuroscience*, vol. 17, no. 8, pp. 2785–2795, 1997.

[169] S. H. Choi, Y. Li, L. F. Parada, and S. S. Sisodia, "Regulation of hippocampal progenitor cell survival, proliferation and dendritic development by BDNF," *Molecular Neurodegeneration*, vol. 4, no. 1, article 52, 2009.

[170] H. Son, M. Banasr, M. Choi et al., "Neuritin produces antidepressant actions and blocks the neuronal and behavioral deficits caused by chronic stress," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 109, no. 28, pp. 11378–11383, 2012.

[171] M. N. Alme, K. Wibrand, G. Dagestad, and C. R. Bramham, "Chronic fluoxetine treatment induces brain region-specific upregulation of genes associated with BDNF-induced long-term potentiation," *Neural Plasticity*, vol. 2007, Article ID 26496, 9 pages, 2007.

[172] M. F. Egan, M. Kojima, J. H. Callicott et al., "The BDNF val66met polymorphism affects activity-dependent secretion of BDNF and human memory and hippocampal function," *Cell*, vol. 112, no. 2, pp. 257–269, 2003.

[173] S. Sen, R. M. Nesse, S. F. Stoltenberg et al., "A BDNF coding variant is associated with the NEO personality inventory domain neuroticism, a risk factor for depression," *Neuropsychopharmacology*, vol. 28, no. 2, pp. 397–401, 2003.

[174] M. Verhagen, A. Van Der Meij, P. M. Van Deurzen et al., "Meta-analysis of the BDNF Val66Met polymorphism in major depressive disorder: effects of gender and ethnicity," *Molecular Psychiatry*, vol. 15, no. 3, pp. 260–271, 2010.

[175] H. Yu, D. D. Wang, Y. Wang, T. Liu, F. S. Lee, and Z. Y. Chen, "Variant brain-derived neurotrophic factor Val66Met polymorphism alters vulnerability to stress and response to

antidepressants," *Journal of Neuroscience*, vol. 32, no. 12, pp. 4092–4101, 2012.

[176] N. A. Kocabas, I. Antonijevic, C. Faghel et al., "Brain-derived neurotrophic factor gene polymorphisms: influence on treatment response phenotypes of major depressive disorder," *International Clinical Psychopharmacology*, vol. 26, pp. 1–10, 2011.

[177] T. D. Palmer, A. R. Willhoite, and F. H. Gage, "Vascular niche for adult hippocampal neurogenesis," *Journal of Comparative Neurology*, vol. 425, pp. 479–494, 2000.

[178] J. L. Warner-Schmidt, T. M. Madsen, and R. S. Duman, "Electroconvulsive seizure restores neurogenesis and hippocampus-dependent fear memory after disruption by irradiation," *European Journal of Neuroscience*, vol. 27, no. 6, pp. 1485–1493, 2008.

[179] V. M. Heine, J. Zareno, S. Maslam, M. Joëls, and P. J. Lucassen, "Chronic stress in the adult dentate gyrus reduces cell proliferation near the vasculature and VEGF and Flk-1 protein expression," *European Journal of Neuroscience*, vol. 21, no. 5, pp. 1304–1314, 2005.

[180] T. Kiuchi, H. Lee, and T. Mikami, "Regular exercise cures depression-like behavior via VEGF-Flk-1 signaling in chronically stressed mice," *Neuroscience*, vol. 207, pp. 208–217, 2012.

[181] S. S. Newton, E. F. Collier, J. Hunsberger et al., "Gene profile of electroconvulsive seizures: induction of neurotrophic and angiogenic factors," *Journal of Neuroscience*, vol. 23, no. 34, pp. 10841–10851, 2003.

[182] C. A. Altar, P. Laeng, L. W. Jurata et al., "Electroconvulsive seizures regulate gene expression of distinct neurotrophic signaling pathways," *Journal of Neuroscience*, vol. 24, no. 11, pp. 2667–2677, 2004.

[183] R. Sun, N. Li, and T. Li, "VEGF regulates antidepressant effects of lamotrigine," *European Neuropsychopharmacology*, vol. 22, pp. 424–430, 2012.

[184] J. S. Lee, D. J. Jang, N. Lee et al., "Induction of neuronal vascular endothelial growth factor expression by cAMP in the dentate gyrus of the hippocampus is required for antidepressant-like behaviors," *Journal of Neuroscience*, vol. 29, no. 26, pp. 8493–8505, 2009.

[185] M. Ventriglia, R. Zanardini, L. Pedrini et al., "VEGF serum levels in depressed patients during SSRI antidepressant treatment," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 33, no. 1, pp. 146–149, 2009.

[186] Z. Halmai, P. Dome, J. Dobos et al., "Peripheral vascular endothelial growth factor level is associated with antidepressant treatment response: results of a preliminary study," *Journal of Affective Disorders*, vol. 144, pp. 269–273, 2013.

[187] D. S. Cowen, "Serotonin and neuronal growth factors—a convergence of signaling pathways," *Journal of Neurochemistry*, vol. 101, no. 5, pp. 1161–1171, 2007.

[188] J. Peltier, A. O'Neill, and D. V. Schaffer, "PI3K/Akt and CREB regulate adult neural hippocampal progenitor proliferation and differentiation," *Developmental Neurobiology*, vol. 67, no. 10, pp. 1348–1361, 2007.

[189] T. D. Gould and H. K. Manji, "Signaling networks in the pathophysiology and treatment of mood disorders," *Journal of Psychosomatic Research*, vol. 53, pp. 687–697, 2002.

[190] J. A. Blendy, "The role of CREB in depression and antidepressant treatment," *Biological Psychiatry*, vol. 59, no. 12, pp. 1144–1150, 2006.

[191] A. J. Shaywitz and M. E. Greenberg, "CREB: a stimulus-induced transcription factor activated by a diverse array of extracellular signals," *Annual Review of Biochemistry*, vol. 68, pp. 821–861, 1999.

[192] H. Viola, M. Furman, L. A. Izquierdo et al., "Phosphorylated cAMP response element-binding protein as a molecular marker of memory processing in rat hippocampus: effect of novelty," *The Journal of Neuroscience*, vol. 20, no. 23, article RC112, 2000.

[193] G. E. Hardingham, F. J. L. Arnold, and H. Bading, "Nuclear calcium signaling controls CREB-mediated gene expression triggered by synaptic activity," *Nature Neuroscience*, vol. 4, no. 3, pp. 261–267, 2001.

[194] B. E. Lonze and D. D. Ginty, "Function and regulation of CREB family transcription factors in the nervous system," *Neuron*, vol. 35, no. 4, pp. 605–623, 2002.

[195] W. A. Carlezon Jr., R. S. Duman, and E. J. Nestler, "The many faces of CREB," *Trends in Neurosciences*, vol. 28, no. 8, pp. 436–445, 2005.

[196] O. Valverde, T. Mantamadiotis, M. Torrecilla et al., "Modulation of anxiety-like behavior and morphine dependence in CREB-deficient mice," *Neuropsychopharmacology*, vol. 29, no. 6, pp. 1122–1133, 2004.

[197] M. Nibuya, E. J. Nestler, and R. S. Duman, "Chronic antidepressant administration increases the expression of cAMP response element binding protein (CREB) in rat hippocampus," *Journal of Neuroscience*, vol. 16, no. 7, pp. 2365–2372, 1996.

[198] T. D. Perera, S. Park, and Y. Nemirovskaya, "Cognitive role of neurogenesis in depression and antidepressant treatment," *Neuroscientist*, vol. 14, no. 4, pp. 326–338, 2008.

[199] M. Johannessen, M. P. Delghandi, and U. Moens, "What turns CREB on?" *Cellular Signalling*, vol. 16, no. 11, pp. 1211–1227, 2004.

[200] W. Y. Kim, X. Wang, Y. Wu et al., "GSK-3 is a master regulator of neural progenitor homeostasis," *Nature Neuroscience*, vol. 12, pp. 1390–1397, 2009.

[201] F. Li, Z. Z. Chong, and K. Maiese, "Vital elements of the Wnt-frizzled signaling pathway in the nervous system," *Current Neurovascular Research*, vol. 2, no. 4, pp. 331–340, 2005.

[202] L. Ciani and P. C. Salinas, "WNTs in the vertebrate nervous system: from patterning to neuronal connectivity," *Nature Reviews Neuroscience*, vol. 6, no. 5, pp. 351–362, 2005.

[203] J. Galceran, E. M. Miyashita-Lin, E. Devaney, J. L. R. Rubenstein, and R. Grosschedl, "Hippocampus development and generation of dentate gyrus granule cells is regulated by LEF1," *Development*, vol. 127, no. 3, pp. 469–482, 2000.

[204] C. J. Zhou, C. Zhao, and S. J. Pleasure, "Wnt signaling mutants have decreased dentate granule cell production and radial glial scaffolding abnormalities," *Journal of Neuroscience*, vol. 24, no. 1, pp. 121–126, 2004.

[205] X. Yu and R. C. Malenka, "Beta-catenin is critical for dendritic morphogenesis," *Nature Neuroscience*, vol. 6, pp. 1169–1177, 2003.

[206] X. Gao, P. Arlotta, J. D. Macklis, and J. Chen, "Conditional knock-out of β-catenin in postnatal-born dentate gyrus granule neurons results in dendritic malformation," *Journal of Neuroscience*, vol. 27, no. 52, pp. 14317–14325, 2007.

[207] A. A. Zaghetto, S. Paina, S. Mantero et al., "Activation of the Wnt-βcatenin pathway in a cell population on the surface of the forebrain is essential for the establishment of olfactory axon connections," *Journal of Neuroscience*, vol. 27, no. 36, pp. 9757–9768, 2007.

[208] S. A. Purro, L. Ciani, M. Hoyos-Flight, E. Stamatakou, E. Siomou, and P. C. Salinas, "Wnt regulates axon behavior through changes in microtubule growth directionality: a new role for adenomatous polyposis coli," *Journal of Neuroscience*, vol. 28, no. 34, pp. 8644–8654, 2008.

[209] S. X. Bamji, K. Shimazu, N. Kimes et al., "Role of β-catenin in synaptic vesicle localization and presynaptic assembly," *Neuron*, vol. 40, no. 4, pp. 719–731, 2003.

[210] F. Hernández, J. Borrell, C. Guaza, J. Avila, and J. J. Lucas, "Spatial learning deficit in transgenic mice that conditionally overexpress GSK-3β in the brain but do not form tau filaments," *Journal of Neurochemistry*, vol. 83, no. 6, pp. 1529–1533, 2002.

[211] C. Yost, M. Torres, J. R. Miller, E. Huang, D. Kimelman, and R. T. Moon, "The axis-inducing activity, stability, and subcellular distribution of β-catenin is regulated in Xenopus embryos by glycogen synthase kinase 3," *Genes and Development*, vol. 10, no. 12, pp. 1443–1454, 1996.

[212] H. Aberle, A. Bauer, J. Stappert, A. Kispert, and R. Kemler, "β-catenin is a target for the ubiquitin-proteasome pathway," *EMBO Journal*, vol. 16, no. 13, pp. 3797–3804, 1997.

[213] L. Li, H. Yuan, C. D. Weaver et al., "Axin and Frat1 interact with Dvl and GSK, bridging Dvl to GSK in Wnt-mediated regulation of LEF-1," *EMBO Journal*, vol. 18, no. 15, pp. 4233–4240, 1999.

[214] C. Y. Logan and R. Nusse, "The Wnt signaling pathway in development and disease," *Annual Review of Cell and Developmental Biology*, vol. 20, pp. 781–810, 2004.

[215] R. S. Jope and G. N. Bijur, "Mood stabilizers, glycogen synthase kinase-3β and cell survival," *Molecular Psychiatry*, vol. 7, supplement 1, pp. S35–S45, 2002.

[216] F. Hernández, J. D. Nido, J. Avila, and N. Villanueva, "GSK3 inhibitors and disease," *Mini-Reviews in Medicinal Chemistry*, vol. 9, no. 9, pp. 1024–1029, 2009.

[217] P. S. Klein and D. A. Melton, "A molecular mechanism for the effect of lithium on development," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 93, pp. 8455–8459, 1996.

[218] A. Wada, H. Yokoo, T. Yanagita, and H. Kobayashi, "Lithium: potential therapeutics against acute brain injuries and chronic neurodegenerative diseases," *Journal of Pharmacological Sciences*, vol. 99, no. 4, pp. 307–321, 2005.

[219] T. Y. Eom and R. S. Jope, "Blocked inhibitory serine-phosphorylation of glycogen synthase kinase-3alpha/beta impairs in vivo neural precursor cell proliferation," *Biological Psychiatry*, vol. 66, no. 5, pp. 494–502, 2009.

[220] D. C. Lie, S. A. Colamarino, H. J. Song et al., "Wnt signalling regulates adult hippocampal neurogenesis," *Nature*, vol. 437, no. 7063, pp. 1370–1375, 2005.

[221] E. M. Wexler, P. Andres, H. I. Kornblum, T. D. Palmer, and D. H. Geschwind, "Endogenous Wnt signaling maintains neural progenitor cell potency," *Stem Cells*, vol. 27, no. 5, pp. 1130–1141, 2009.

[222] K. Adachi, Z. Mirzadeh, M. Sakaguchi et al., "β-catenin signaling promotes proliferation of progenitor cells in the adult mouse subventricular zone," *Stem Cells*, vol. 25, no. 11, pp. 2827–2836, 2007.

[223] N. Uchida, Y. Honjo, K. R. Johnson, M. J. Wheelock, and M. Takeichi, "The catenin/cadherin adhesion system is localized in synaptic junctions bordering transmitter release zones," *Journal of Cell Biology*, vol. 135, no. 3, pp. 767–779, 1996.

[224] F. Miskevich, Y. Zhu, B. Ranscht, and J. R. Sanes, "Expression of multiple cadherins and catenins in the chick optic tectum," *Molecular and Cellular Neurosciences*, vol. 12, no. 4-5, pp. 240–255, 1998.

[225] I. Yao, J. Iida, N. Tanaka, Y. Hata, T. Medical, and D. University, "Interaction of synaptic scaffolding molecule and β-catenin," *Journal of Neuroscience*, vol. 22, no. 3, pp. 757–765, 2002.

[226] W. J. Nelson and R. Nusse, "Convergence of Wnt, beta-catenin, and cadherin pathways," *Science*, vol. 303, no. 5663, pp. 1483–1487, 2004.

[227] H. Togashi, K. Abe, A. Mizoguchi, K. Takaoka, O. Chisaka, and M. Takeichi, "Cadherin regulates dendritic spine morphogenesis," *Neuron*, vol. 35, no. 1, pp. 77–89, 2002.

[228] S. P. Shevtsov, S. Haq, and T. Force, "Activation of β-catenin signaling pathways by classical G-protein-coupled receptors: mechanisms and consequences in cycling and non-cycling cells," *Cell Cycle*, vol. 5, no. 20, pp. 2295–2300, 2006.

[229] T. Jin, I. George Fantus, and J. Sun, "Wnt and beyond Wnt: multiple mechanisms control the transcriptional property of β-catenin," *Cellular Signalling*, vol. 20, no. 10, pp. 1697–1704, 2008.

[230] J. Kunz, R. Henriquez, U. Schneider, M. Deuter-Reinhard, N. R. Movva, and M. N. Hall, "Target of rapamycin in yeast, TOR2, is an essential phosphatidylinositol kinase homolog required for G1 progression," *Cell*, vol. 73, no. 3, pp. 585–596, 1993.

[231] J. Heitman, N. R. Movva, and M. N. Hall, "Targets for cell cycle arrest by the immunosuppressant rapamycin in yeast," *Science*, vol. 253, no. 5022, pp. 905–909, 1991.

[232] S. B. Helliwell, P. Wagner, J. Kunz, M. Deuter-Reinhard, R. Henriquez, and M. N. Hall, "TOR1 and TOR2 are structurally and functionally similar but not identical phosphatidylinositol kinase homologues in yeast," *Molecular Biology of the Cell*, vol. 5, no. 1, pp. 105–118, 1994.

[233] R. Loewith, E. Jacinto, S. Wullschleger et al., "Two TOR complexes, only one of which is rapamycin sensitive, have distinct roles in cell growth control," *Molecular Cell*, vol. 10, no. 3, pp. 457–468, 2002.

[234] E. Jacinto, R. Loewith, A. Schmidt et al., "Mammalian TOR complex 2 controls the actin cytoskeleton and is rapamycin insensitive," *Nature Cell Biology*, vol. 6, no. 11, pp. 1122–1128, 2004.

[235] T. Takahara, K. Hara, K. Yonezawa, H. Sorimachi, and T. Maeda, "Nutrient-dependent multimerization of the mammalian target of rapamycin through the N-terminal HEAT repeat region," *Journal of Biological Chemistry*, vol. 281, no. 39, pp. 28605–28614, 2006.

[236] F. C. Harwood, L. Shu, and P. J. Houghton, "mTORC1 signaling can regulate growth factor activation of p44/42 mitogen-activated protein kinases through protein phosphatase 2A," *Journal of Biological Chemistry*, vol. 283, no. 5, pp. 2575–2585, 2008.

[237] N. Hosokawa, T. Hara, T. Kaizuka et al., "Nutrient-dependent mTORCl association with the ULK1-Atg13-FIP200 complex required for autophagy," *Molecular Biology of the Cell*, vol. 20, no. 7, pp. 1981–1991, 2009.

[238] C. A. Hoeffer and E. Klann, "mTOR signaling: at the crossroads of plasticity, memory and disease," *Trends in Neurosciences*, vol. 33, no. 2, pp. 67–75, 2010.

[239] M. Livingstone, E. Atas, A. Meller, and N. Sonenberg, "Mechanisms governing the control of mRNA translation," *Physical Biology*, vol. 7, no. 2, Article ID 021001, 2010.

[240] N. Hay and N. Sonenberg, "Upstream and downstream of mTOR," *Genes and Development*, vol. 18, no. 16, pp. 1926–1945, 2004.

[241] R. Gong, S. P. Chang, N. R. Abbassi, and S. J. Tang, "Roles of glutamate receptors and the mammalian target of rapamycin (mTOR) signaling pathway in activity-dependent dendritic protein synthesis in hippocampal neurons," *Journal of Biological Chemistry*, vol. 281, no. 27, pp. 18802–18815, 2006.

[242] S. J. Tang, G. Reis, H. Kang, A. C. Gingras, N. Sonenberg, and E. M. Schuman, "A rapamycin-sensitive signaling pathway contributes to long-term synaptic plasticity in the hippocampus," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 99, no. 1, pp. 467–472, 2002.

[243] R. S. Duman, N. Li, R. J. Liu, V. Duric, and G. Aghajanian, "Signaling pathways underlying the rapid antidepressant actions of ketamine," *Neuropharmacology*, vol. 62, pp. 35–41, 2012.

[244] Z. Z. Chong, Y. C. Shang, L. Zhang, S. Wang, and K. Maiese, "Mammalian target of rapamycin: hitting the bull's-eye for neurological disorders," *Oxidative Medicine and Cellular Longevity*, vol. 3, no. 6, pp. 374–391, 2010.

[245] W. L. An, R. F. Cowburn, L. Li et al., "Up-regulation of phosphorylated/activated p70 S6 kinase and its relationship to neurofibrillary pathology in Alzheimer's disease," *American Journal of Pathology*, vol. 163, pp. 591–607, 2003.

[246] W. L. Zhu, H. S. Shi, S. J. Wang, P. Wu, Z. B. Ding, and L. Lu, "Hippocampal CA3 calcineurin activity participates in depressive-like behavior in rats," *Journal of Neurochemistry*, vol. 117, no. 6, pp. 1075–1086, 2011.

[247] C. S. Jernigan, D. B. Goswami, M. C. Austin et al., "The mTOR signaling pathway in the prefrontal cortex is compromised in major depressive disorder," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 35, no. 7, pp. 1774–1779, 2011.

[248] C. Cleary, J. A. S. Linde, K. M. Hiscock et al., "Antidepressive-like effects of rapamycin in animal models: implications for mTOR inhibition as a new target for treatment of affective disorders," *Brain Research Bulletin*, vol. 76, no. 5, pp. 469–473, 2008.

[249] S. C. Yoon, M. S. Seo, S. H. Kim et al., "The effect of MK-801 on mTOR/p70S6K and translation-related proteins in rat frontal cortex," *Neuroscience Letters*, vol. 434, no. 1, pp. 23–28, 2008.

[250] S. Maeng, C. A. Zarate Jr., J. Du et al., "Cellular mechanisms underlying the antidepressant effects of ketamine: role of α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid receptors," *Biological Psychiatry*, vol. 63, no. 4, pp. 349–352, 2008.

[251] H. Koike, M. Iijima, and S. Chaki, "Involvement of the mammalian target of rapamycin signaling in the antidepressant-like effect of group II metabotropic glutamate receptor antagonists," *Neuropharmacology*, vol. 61, pp. 1419–1423, 2011.

[252] C. Yang, Y. M. Hu, Z. Q. Zhou, G. F. Zhang, and J. J. Yang, "Acute administration of ketamine in rats increases hippocampal BDNF and mTOR levels during forced swimming test," *Upsala Journal of Medical Sciences*, vol. 118, no. 1, pp. 3–8, 2013.

[253] C. A. Zarate Jr., J. B. Singh, P. J. Carlson et al., "A randomized trial of an N-methyl-D-aspartate antagonist in treatment-resistant major depression," *Archives of General Psychiatry*, vol. 63, no. 8, pp. 856–864, 2006.

[254] B. Elfving and G. Wegener, "Electroconvulsive seizures stimulate the vegf pathway via mTORC1," *Synapse*, vol. 66, pp. 340–345, 2012.

[255] J. M. Dwyer, A. E. Lepack, and R. S. Duman, "mTOR activation is required for the antidepressant effects of mGluR(2)/(3) blockade," *International Journal of Neuropsychopharmacology*, vol. 15, pp. 429–434, 2012.

[256] A. J. Eisch, "Adult neurogenesis: implications for psychiatry," *Progress in Brain Research*, vol. 138, pp. 315–342, 2002.

[257] Y. Li, B. W. Luikart, S. Birnbaum et al., "TrkB regulates hippocampal neurogenesis and governs sensitivity to antidepressive treatment," *Neuron*, vol. 59, no. 3, pp. 399–412, 2008.

[258] C. Pittenger and R. S. Duman, "Stress, depression, and neuroplasticity: a convergence of mechanisms," *Neuropsychopharmacology*, vol. 33, no. 1, pp. 88–109, 2008.

[259] E. Fuchs, B. Czéh, M. H. P. Kole, T. Michaelis, and P. J. Lucassen, "Alterations of neuroplasticity in depression: the hippocampus and beyond," *European Neuropsychopharmacology*, vol. 14, supplement 5, pp. S481–S490, 2004.

[260] Y. Watanabe, E. Gould, and B. S. McEwen, "Stress induces atrophy of apical dendrites of hippocampal CA3 pyramidal neurons," *Brain Research*, vol. 588, no. 2, pp. 341–345, 1992.

[261] S. D. Norrholm and C. C. Ouimet, "Altered dendritic spine density in animal models of depression and in response to antidepressant treatment," *Synapse*, vol. 42, no. 3, pp. 151–163, 2001.

[262] S. Nakagawa, J. E. Kim, R. Lee et al., "Regulation of neurogenesis in adult mouse hippocampus by cAMP and the cAMP response element-binding protein," *Journal of Neuroscience*, vol. 22, no. 9, pp. 3673–3682, 2002.

[263] D. J. Christoffel, S. A. Golden, and S. J. Russo, "Structural and synaptic plasticity in stress-related disorders," *Annual Review of Neuroscience*, vol. 22, pp. 535–549, 2011.

[264] S. R. Lamont, B. J. Stanwell, R. Hill, I. C. Reid, and C. A. Stewart, "Ketamine pre-treatment dissociates the effects of electroconvulsive stimulation on mossy fibre sprouting and cellular proliferation in the dentate gyrus," *Brain Research*, vol. 1053, no. 1-2, pp. 27–32, 2005.

Hindawi Publishing Corporation
Neural Plasticity
Volume 2013, Article ID 318596, 24 pages
http://dx.doi.org/10.1155/2013/318596

Review Article

Immature Dentate Gyrus: An Endophenotype of Neuropsychiatric Disorders

Hideo Hagihara,[1,2] Keizo Takao,[1,2,3] Noah M. Walton,[4] Mitsuyuki Matsumoto,[4] and Tsuyoshi Miyakawa[1,2,3]

[1] *Division of Systems Medical Science, Institute for Comprehensive Medical Science, Fujita Health University, 1-98 Dengakugakubo, Kutsukake-cho, Toyoake, Aichi 470-1192, Japan*
[2] *CREST, Japan Science and Technology Agency, 4-1-8 Honcho, Kawaguchi, Saitama 332-0012, Japan*
[3] *Section of Behavior Patterns, Center for Genetic Analysis of Behavior, National Institute for Physiological Sciences, 5-1 Aza-Higashiyama, Myodaiji-cho, Okazaki, Aichi 444-8787, Japan*
[4] *CNS, Astellas Research Institute of America LLC, 8045 Lamon Avenue, Skokie, IL 60077, USA*

Correspondence should be addressed to Tsuyoshi Miyakawa; miyakawa@fujita-hu.ac.jp

Received 5 March 2013; Revised 17 April 2013; Accepted 19 April 2013

Academic Editor: Chitra D. Mandyam

Adequate maturation of neurons and their integration into the hippocampal circuit is crucial for normal cognitive function and emotional behavior, and disruption of this process could cause disturbances in mental health. Previous reports have shown that mice heterozygous for a null mutation in α-CaMKII, which encodes a key synaptic plasticity molecule, display abnormal behaviors related to schizophrenia and other psychiatric disorders. In these mutants, almost all neurons in the dentate gyrus are arrested at a pseudoimmature state at the molecular and electrophysiological levels, a phenomenon defined as "immature dentate gyrus (iDG)." To date, the iDG phenotype and shared behavioral abnormalities (including working memory deficit and hyperlocomotor activity) have been discovered in Schnurri-2 knockout, mutant SNAP-25 knock-in, and forebrain-specific calcineurin knockout mice. In addition, both chronic fluoxetine treatment and pilocarpine-induced seizures reverse the neuronal maturation, resulting in the iDG phenotype in wild-type mice. Importantly, an iDG-like phenomenon was observed in post-mortem analysis of brains from patients with schizophrenia/bipolar disorder. Based on these observations, we proposed that the iDG is a potential endophenotype shared by certain types of neuropsychiatric disorders. This review summarizes recent data describing this phenotype and discusses the data's potential implication in elucidating the pathophysiology of neuropsychiatric disorders.

1. Introduction

The exact mechanisms within the brain that underlie most psychiatric disorders remain largely unknown, and one of the major challenges in psychiatric research is to identify the pathophysiology in the brains of patients with these disorders. This is challenging because psychiatric disorders are diagnosed on the basis of behavioral characteristics and not biological criteria. Therefore, each psychiatric disorder likely consists of multiple biologically heterogeneous populations, which further complicates the search for underlying pathophysiologies.

Previously, studies have identified the "immature dentate gyrus (iDG)," a potential brain endophenotype shared by several psychiatric disorders, including schizophrenia and bipolar disorder. The iDG was identified in animal models of psychiatric disorders, which were selected using large-scale behavioral screening of genetically engineered mice [1]. In the iDG phenotype, most of the granule cells or principal neurons in the dentate gyrus (DG) within the hippocampus are arrested at a pseudoimmature status, in which the molecular and physiological properties are similar to those of normal immature neurons from the DG. To date, the iDG phenotype has been identified in several strains of mutant mice, including α-CaMKII heterozygous knockout (HKO) mice [1], Schnurri-2 (Shn-2) knockout (KO) mice [2], mutant SNAP-25 knock-in (KI) mice [3], and forebrain-specific calcineurin (CN) KO mice [4]. A signature pattern,

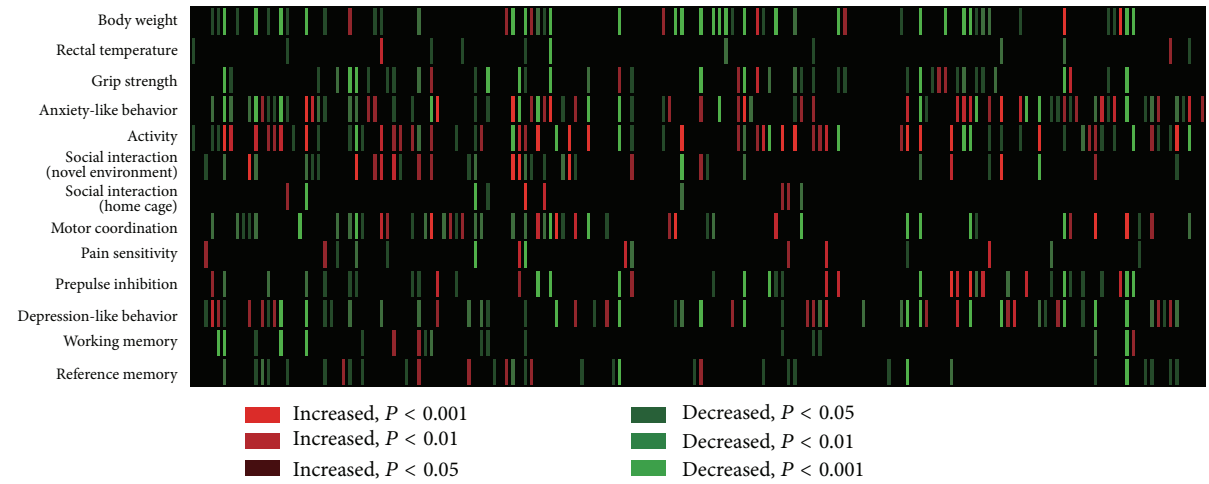


Figure 1: Heat map showing behavioral phenotypes of more than 160 strains of genetically engineered mice. Each column represents the mouse strain analyzed in the laboratory of the author's group (unpublished data). Each row represents a behavior category assessed by the comprehensive battery of behavioral tests. Colors represent an increase (red) or decrease (green), compared between wild-type and mutant mice. Adapted from Takao et al. [8].

quite similar to the iDG phenotype identified in these mutant mice, has also been found in mice treated with chronic fluoxetine [5] and in a pilocarpine-induced mouse model of epilepsy [6]. Moreover, postmortem analysis for molecular markers of neuronal maturity in the DG revealed an iDG-like signature for brains of patients with schizophrenia and bipolar disorder [7].

The purpose of this review is to introduce how the iDG phenotype was identified and its molecular and cellular signature. An additional goal is to summarize potential mechanisms underlying the iDG phenotype and its impact on brain functions.

2. Discovery of iDG in α-CaMKII HKO Mice

A high-throughput comprehensive behavioral testing battery was developed for many different strains of genetically engineered mice. The battery of tests covers many behavioral domains ranging from sensorimotor functions to highly cognitive functions, like learning and memory. In the course of a large-scale effort to phenotype the genetically engineered mice (mutant mice), this battery of tests was administered to more than 160 strains of knockout and transgenic mice (Figure 1) [8]. Surprisingly, when tested most of the strains of mutant mice displayed at least some aberrant behavioral phenotypes, which may suggest that many of the genes expressed in the brain may have some functional significance at the behavior level. In the course of screening the mutant mice populations, a number of mouse strains were identified that showed abnormal behaviors common to neuropsychiatric disorders (e.g., schizophrenia, bipolar disorder, and autism). Among these mutant mouse models, the α-CaMKII HKO mice showed a particularly dramatic series of abnormal behaviors. The first discovery of the iDG phenotype occurred during investigations into the neuropathophysiology underlying these behaviors.

Calmodulin-dependent protein kinase II (CaMKII) is a major downstream molecule of the *N*-methyl-d-aspartic acid (NMDA) receptor, which has presumed involvement in the pathophysiology of schizophrenia. The function of the NMDA receptor can be modulated by calcineurin, a potential susceptibility gene we previously identified [9, 10]. CaMKII is thought to play an essential role in neural plasticity, such as long-term potentiation and long-term depression [11].

Perhaps the most striking behavioral phenotype of α-CaMKII HKO mice is that they occasionally kill their cage mates (often littermates). Prior to 6 months of age, α-CaMKII HKO mice kill more than half of their group-housed cage mates. In addition, these mice show greatly increased locomotor activity in the open field test. While α-CaMKII HKO mice demonstrate normal reference memory, as assessed by the reference memory version of the eight-arm radial maze test, their working memory is severely impaired. Likewise, in the T-maze forced-alternation test, the working memory of the α-CaMKII HKO mice was found to be severely impaired, while they showed normal performance in a simple left/right discrimination task using the same apparatuses. Previous studies have presented representative videos of the performance of α-CaMKII HKO mice in these tasks [12]. Using the home cage locomotor activity monitoring system, which measures distance travelled in the home cage, it was shown that the locomotor activity patterns of the α-CaMKII HKO mice slowly and dramatically change over time. These mice show a periodic mood-change-like behavior in their home cage (Figure 2(a)), and 1 cycle lasts approximately 10–20 d. The manifestations of this behavioral phenotype are so clear that these mutant mice can be easily identified using these activity patterns.

The next task was to understand the underlying pathophysiology in the brains of the α-CaMKII mutants that show this dramatic array of behavioral abnormalities. First, a transcriptome analysis of the hippocampus of mutant mice

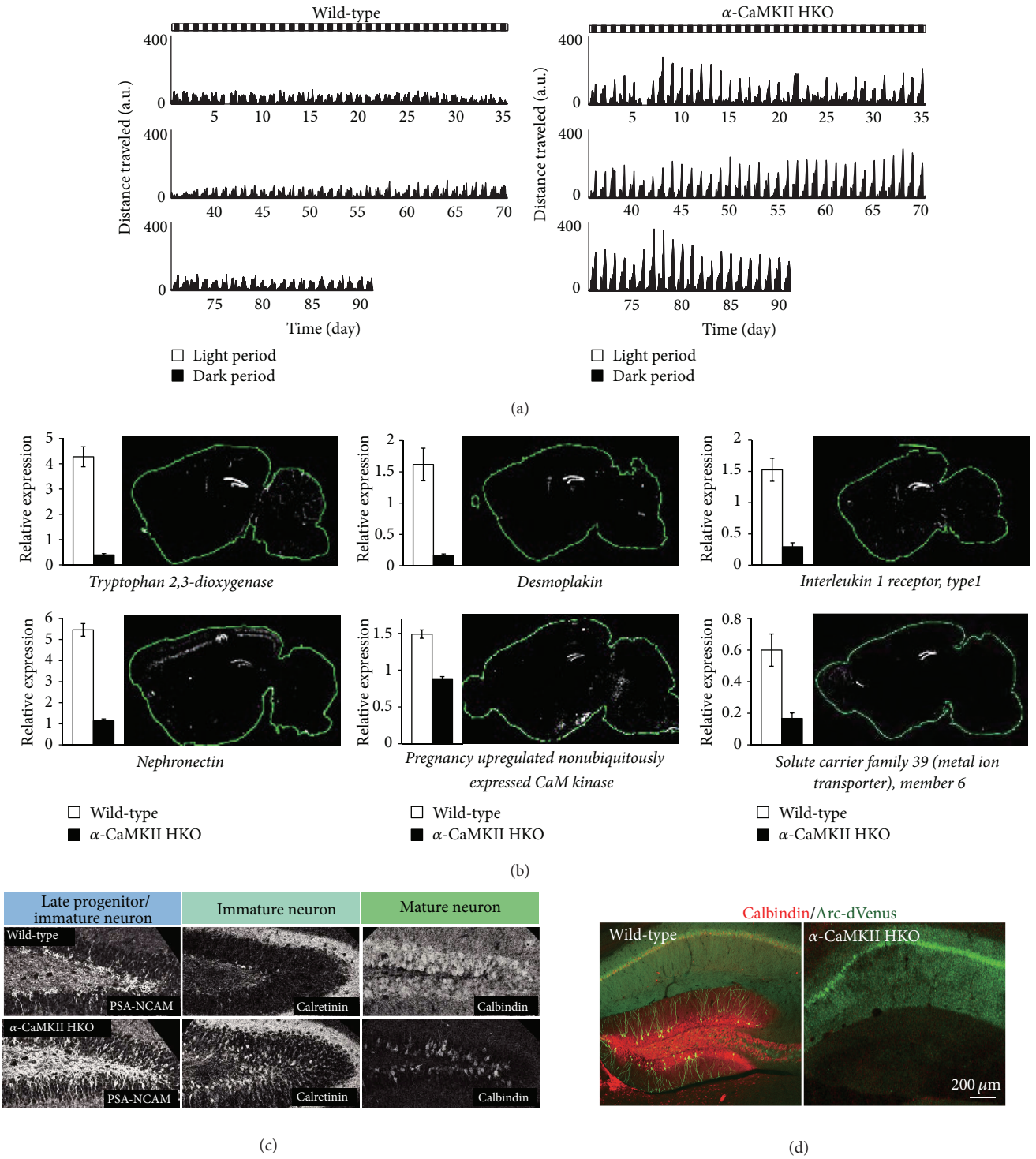


Figure 2: Mood-change-like behavior and the iDG phenotype in α-CaMKII HKO mice. (a) Pattern of locomotor activity of wild-type and α-CaMKII HKO mice in their home cage. Mutant mice were hyperactive and showed a periodic mood-change-like activity pattern. A.U: arbitrary unit. (b) Among the downregulated genes in the hippocampus of α-CaMKII HKO mice, several genes were expressed selectively in the DG (Allen Brain Atlas [42]. Seattle, WA: Allen Institute for Brain Science. © 2004–2008. Available from: http://www.brain-map.org). Graphs indicate the relative expression levels of each gene in the microarray experiment. (c) In α-CaMKII HKO mice, expression of PSA-NCAM (a late-progenitor and immature-neuron marker) and calretinin (an immature neuron marker) was markedly increased, and expression of calbindin (a mature neuron marker) was decreased. (d) Expression of Arc-dVenus in the DG of α-CaMKII HKO mice after the working memory task (eight-arm radial maze test) was completely abolished. *Red:* calbindin; *green:* Arc-dVenus. Adapted from Yamasaki et al. [1] and Matsuo et al. [13].

was conducted. Gene expression in α-CaMKII HKO mice was highly dysregulated, with altered expressions of more than 2,000 genes in the hippocampus of the mutants. Surprisingly, of the top 30 hippocampal genes with downregulated expression in α-CaMKII mutants, 6 genes showed highly selective expression in the DG (Figure 2(b)). These data implied the existence of some unknown abnormalities in the DG of these mutants.

Adult neurogenesis was then examined and a large upregulation (>50%) of neurogenesis was demonstrated in α-CaMKII mutant mice. Traditionally, the process of adult neurogenesis involves a recapitulation during which stem/progenitor cells give way to immature postmitotic neurons, which then mature to adopt distinct morphological and physiological properties in the hippocampus. Based on the observed upregulation of adult neurogenesis in the α-CaMKII mutant mice, it was interesting to determine whether the α-CaMKII mutant mice followed the traditional path of neurogenesis. Gene chip analysis of hippocampus demonstrated a reduced expression of the gene for calbindin, a marker for mature neurons in the DG, in the mutants. Furthermore, calbindin, as assessed by immunohistochemistry, was dramatically reduced in the DG of the α-CaMKII mutants (Figure 2(c)). In addition, expression of calretinin, a marker for immature neurons in the DG, was increased, as was expression of polysialylated neuronal cell adhesion molecule (PSA-NCAM), a marker for late progenitor cells and immature neurons. Taken together, these findings suggest that in α-CaMKII mutant mice the number of immature neurons are upregulated and the number of mature neurons are downregulated.

Golgi staining was also performed to examine the morphology of the DG neurons. Interestingly, staining of the DG was difficult to achieve in the α-CaMKII mutant mice, presenting a challenge in the quantification of morphological alterations. Fewer dendritic intersections and shorter overall dendritic length were demonstrated in the α-CaMKII mutant mice, consistent with the idea that DG neurons in these mice are immature.

Moreover, DG neurons in the α-CaMKII mutant mice were found to have many electrophysiological features that are known characteristics of immature dentate neurons, including high input resistance and a decreased number of spikes during sustained depolarization. This finding indicates that, in the α-CaMKII mutant mice, most of the DG neurons have electrophysiological properties consistent with those of immature neurons. Interestingly, α-CaMKII HKO mice also exhibit abnormal synaptic transmission/plasticity at mossy fiber-CA3 synapses. Specifically, α-CaMKII mutant mice have increased basal transmission and dramatically decreased facilitation at these synapses.

The α-CaMKII HKO mice were mated with transgenic mice expressing destabilized Venus (dVenus) under the Arc promoter. Arc is known to be induced by neural activity. In normal mice, a number of hippocampal neurons (including DG neurons) become positive for Arc-dVenus after performing working memory tasks (Figure 2(d)). In α-CaMKII HKO mice, Arc-dVenus expression is almost completely abolished in the DG after performing a working memory task, suggesting that the DG in these mutants is not fully functional [13].

These experiments were the first to demonstrate that the most of the DG neurons in α-CaMKII HKO mice are arrested at a pseudoimmature status. This arrest of maturity was termed the "immature dentate gyrus (iDG)" phenotype.

3. iDG Is an Endophenotype Shared by Genetically Engineered Mice with Abnormal Behaviors Characteristic for Neuropsychiatric Disorders

After identification of the iDG phenotype, it was important to investigate whether other strains of mutant mice share this phenotype. Over the past 10 years, the behaviors of mutant mice have been assessed continuously using the comprehensive behavioral test battery previously described [8]. The majority of the behavioral phenotypes reported are registered in the mouse phonotype database (http://www.mousephenotype.org). This database was used to identify strains of mice that exhibited behavioral phenotype(s) similar to that shown by the α-CaMKII mutants. More than 160 different strains of mutant mice were investigated, and the best behavioral match was identified in a strain of the mice lacking a transcription factor called Schnurri-2 (Shn-2 KO mice) [2].

Shn-2 KO mice show an array of behavioral abnormalities that is quite similar to those of α-CaMKII HKO mice, including hyperlocomotor activity and severe deficits in working memory. In Shn-2 KO mice, locomotor activity is dramatically increased and prepulse inhibition (PPI) is impaired [2]. However, Shn-2 KO mice have normal reference memory, as assessed by the left-right discrimination task performed in a T-maze. In addition, the Shn-2 KO mice have severely impaired working memory, as suggested by their performance in the T-maze forced-alternation task and the eight-arm radial maze (Figures 3(a)–3(d)) [2].

To identify the pathophysiologic abnormalities responsible for the behavioral phenotypes in Shn-2 KO mice, an additional gene chip analysis was conducted and the hippocampal transcriptome patterns of Shn-2 KO mice were compared to those of α-CaMKII HKO mice. Remarkably, the 2 mutant strains showed strikingly similar gene expression patterns in the hippocampus (Figures 4(a) and 4(b)), with expression of >100 genes altered in a similar manner with regard to both the direction and the magnitude of the alterations. For example, as observed in α-CaMKII HKO mice, calbindin expression was dramatically decreased in the DG of Shn-2 KO mice (Figure 4(c)) [2]. Whole-cell patch recordings made from granule cells in the DG revealed that DG neurons in Shn-2 KO mice have electrophysiological features shown by immature neurons, such as a lower current threshold for firing, a short latency to the first spike, and a decreased number of spikes during sustained depolarization [2]. This evidence supports the conclusion that Shn-2 KO mice also express iDG phenotype. Additional strains of mutant mice with the iDG phenotype have also been identified.

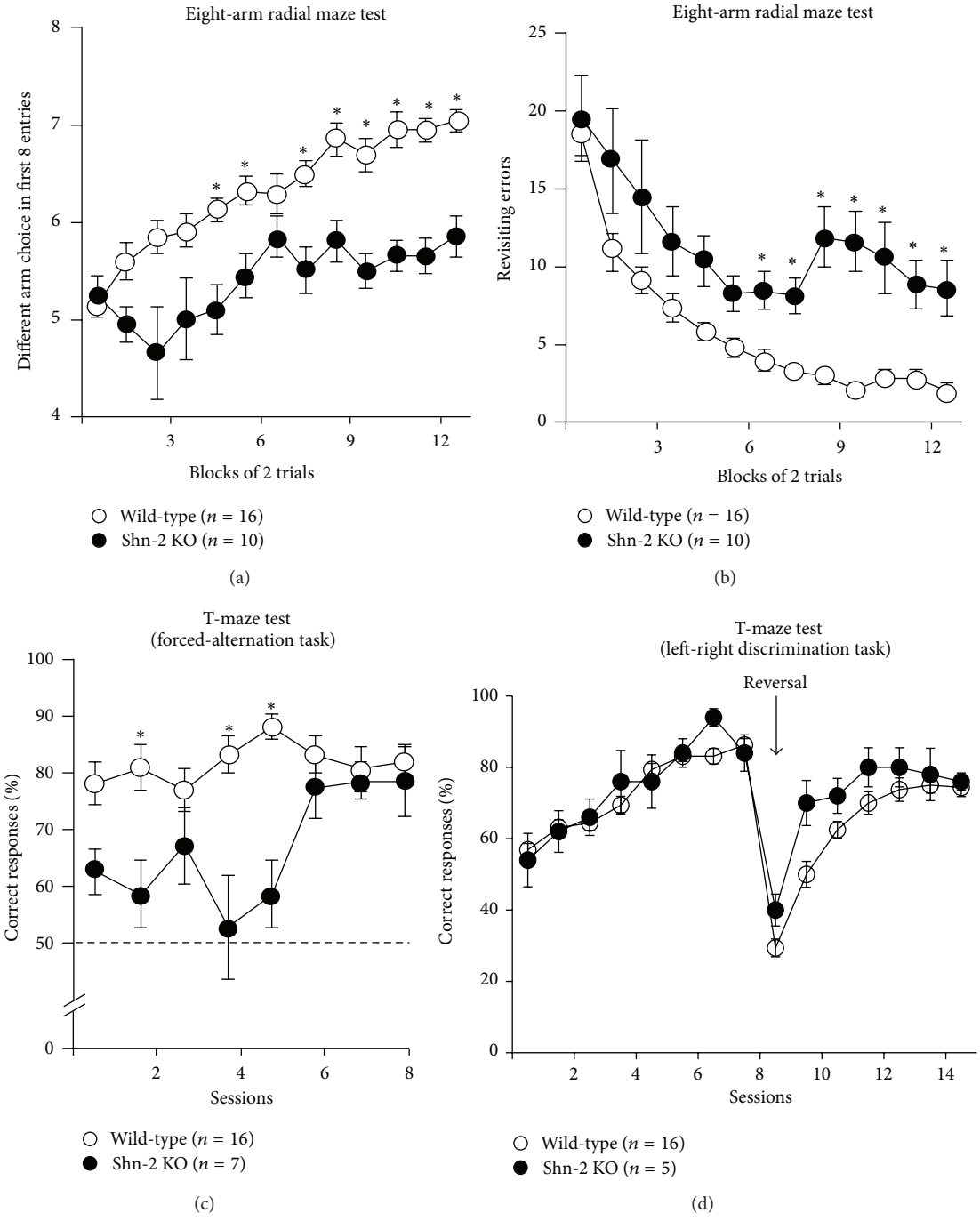


Figure 3: Impaired working memory performance in Shn-2 KO mice. (a) In the spatial working memory version of the eight-arm radial maze, compared to controls, Shn-2 KO mice performed significantly worse with respect to the number of different arm choices in the first 8 entries (genotype effect: $F_{1,24} = 62.104$, $P < 0.0001$). (b) Mutants made significantly more revisiting errors than controls (genotype effect: $F_{1,24} = 45.597$, $P < 0.0001$; genotype × trial block interaction: $F_{12,228} = 1.470$, $P = 0.1345$). (c) Shn-2 KO mice also showed poor working memory performance in the T-maze forced-alternation task (genotype effect: $F_{1,21} = 20.497$: $P = 0.0002$; genotype × session interaction: $F_{7,147} = 3.273$: $P = 0.0029$). (d) Shn-2 KO and wild-type mice were comparable in the left-right discrimination task (genotype effect: $F_{1,19} = 0.209$, $P = 0.6529$) and reversal learning (genotype effect: $F_{1,19} = 5.917$, $P = 0.0251$). Asterisks indicate statistical significance determined using the Student's t-test with a correction for multiple comparisons in each block (a, b) and each session (c) ($^*P < 0.05$). Adapted from Takao et al. [2].

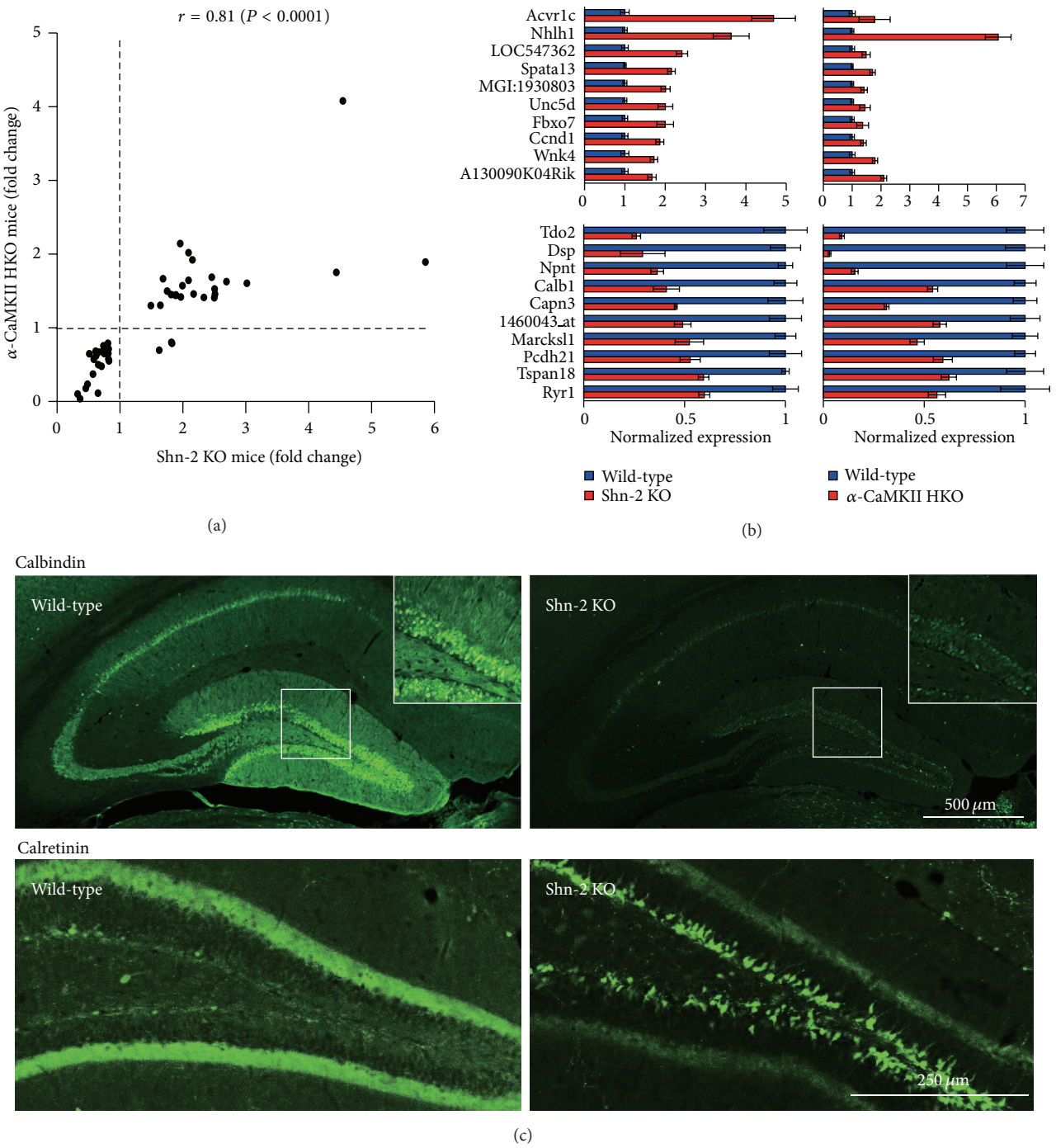


Figure 4: Maturation abnormalities in DG neurons of Shn-2 KO mice. (a) The hippocampal transcriptome pattern of Shn-2 KO mice is similar to that of α-CaMKII HKO mice, which also demonstrated maturation abnormalities in the DG. Genes showing differential expression between genotypes at $P < 0.005$ in both experiments were plotted. (b) Normalized gene expression of differentially expressed genes in Shn-2 KO and α-CaMKII HKO mice. The top 10 genes are indicated in the graphs. (c) Expression of the mature neuronal marker calbindin was decreased, and the expression of the immature neuronal marker calretinin was markedly increased in the DG of Shn-2 KO mice. Adapted from Takao et al. [2].

Synaptosomal-associated protein of 25 kDa (SNAP-25) regulates exocytosis of neurotransmitters and is thought to be involved in the neuropsychiatric disorders such as schizophrenia [14, 15], attention-deficit/hyperactivity disorder (AD/HD) [16, 17], and epilepsy [18, 19]. The iDG phenotype was also identified in SNAP-25 knock-in (KI) mice with a single amino acid substitution [3]. Behaviorally, these mice also show severe deficits in working memory. Of note, chronic administration of valproic acid, an antiepileptic drug, rescued both the iDG phenotype and working memory

deficit in SNAP-25 KI mice. CN is a key signal transduction molecule in the brain, and several studies have suggested a link between dysfunction of CN signaling and schizophrenia [9, 20]. Conditional forebrain-specific CN KO mice also exhibit abnormal behaviors related to schizophrenia [10, 21], including a severe working memory deficit, and an iDG phenotype was also observed in CN KO mice [4].

Further studies have established a method for assessing the existence of the iDG phenotype using real-time polymerase chain reaction (PCR) with 3 probes (Figure 5) [22] selected using gene chip data from α-CaMKII HKO mice. The molecular markers for iDG include increased expression of dopamine receptor D1A (*Drd1a*) and decreased expression of tryptophan 2,3-dioxygenase (*Tdo2*) and desmoplakin (*Dsp*) in the DG of mutant mice [1]. To date, based on these molecular markers for iDG, 9 of the 16 strains of mice that exhibited abnormal behaviors (e.g., hyperlocomotor activity, reduced anxiety-like behavior, and impaired working memory) have been shown to express the iDG phenotype. These results suggest that iDG is a common endophenotype shared by mice with behavioral abnormalities characteristic for neuropsychiatric disorders.

Furthermore, the current results demonstrated that hippocampal gene expression patterns of mice expressing a constitutively active cAMP-response element-binding protein (CREB) variant (VP16-CREB) were similar to those of α-CaMKII HKO, Shn-2 KO, or SNAP-25 KI mice. Notably, VP16-CREB mice exhibited dramatic alteration in expression of *Calb1*, *Dsp*, glial fibrillary acidic protein (*GFAP*), and complement genes in the hippocampus [23]. It has been shown previously that CREB activation is observed in immature granule cells and plays important roles in differentiation, survival, and maturation of neurons [24]. Thus, it is possible that chronic activation of CREB induced a maturation abnormality of the granule cells, resulting in an iDG-like phenotype.

4. Induction of iDG in Wild-Type Animals

The iDG phenotype can be seen not only in genetically engineered mice, but also in properly manipulated wild-type normal mice. For example, Kobayashi et al. found that chronic treatment with the antidepressant fluoxetine (FLX), one of the most widely used selective serotonin reuptake inhibitors (SSRI), can induce iDG phenotypes in adult mice [5]. In FLX-treated mice, expression of *Calb1*, *Tdo2*, and *Dsp*, markers for mature granule cells, was greatly reduced in the DG. In contrast, expression of calretinin, a marker for immature granule cells, was increased in the DG [5]. These findings suggest that the number of immature granule cells is upregulated and the number of mature granule cells is downregulated in FLX-treated mice. Electrophysiologically, FLX-treated granule cells showed higher excitability, which is a functional characteristic observed in immature granule cells. FLX treatment was also found to reduce mossy fiber synaptic facilitation to juvenile levels [5]. These molecular and electrophysiological phenotypes found in FLX-treated mice are strikingly similar to those found in α-CaMKII HKO, Shn-2 KO, and SNAP-25 KI mice (see Table 1). This likely represents an example of "dematuration," in which mature neurons are reversed to a pseudoimmature status. In the study by Kobayashi et al., the dose of FLX used to treat wild-type mice was higher than the doses used in other studies using both wild-type mice [25, 26] and mouse model of depression [27, 28]. This FLX-induced iDG caused by "dematuration" may be related to the antidepressant-induced mania/psychosis observed in clinical settings.

Recently, a second group reported that FLX treatment induced an immature state in basolateral amygdala neurons in adult brains [36]. In their study, chronic FLX treatment reduced the percentage of neurons containing perineuronal nets (PNNs) and expressing parvalbumin (PV) in the region. PNNs predominantly surround mature PV-positive fast-spiking (FS) neurons. PNNs develop late in postnatal development and their formation coincides with the closure of critical developmental periods and the maturation of PV-positive neurons [37, 38]. During the critical period, absence of PNNs is thought to allow the induction of synaptic plasticity. Degradation of PNNs could reactivate synaptic plasticity in adult brain [39, 40]. Thus, FLX treatment shifted the PV- and PNN-containing neurons toward an immature state in the adult basolateral amygdala [36]. The amygdala plays a crucial role in fear-related behaviors. Chronic FLX treatment has been thought to increase synaptic plasticity, converting fear memory circuitry to a more immature state, subsequently allowing erasure of that fear memory. A bidirectional regulation of neuronal maturation in both the DG and amygdala could be an important mechanism of FLX action. In terms of regulation of neuronal maturation, chronic FLX treatment has also been shown to reactivate ocular dominance plasticity in the adult visual cortex [41]. Considering that ocular dominance plasticity is restricted to a critical period during postnatal development [39], chronic FLX treatment could reinstate a juvenile-like form of that plasticity in adulthood. These findings suggest that "dematuration" of neurons caused by FLX is not specific to the DG granule cells.

In addition to FLX, another SSRI, paroxetine, is most likely to induce "dematuration" of DG granule cells in adult mice. Chronic paroxetine treatment has been found to suppress calbindin and *Dsp* expression in the DG [5, 31] and reduce frequency facilitation at mossy fiber-CA3 synapses (mossy fiber synapses formed by immature granule cells exhibit smaller frequency facilitation) [5]. These results suggest that serotonergic antidepressants can reverse the state of neuronal maturation in the adult DG.

Individuals with epilepsy are at significant risk of psychosis with emerging behavioral features that include hallucinations, mood instability, mixed irritability, and mania. Recent genetic studies have shown that chromosome copy number variations, such as the 15q13.3 microdeletion, as conferring an increased risk for developing psychosis and epilepsy and suggest that common phenotypes in epilepsy and psychosis may share a genetic basis.

Pilocarpine-induced seizure is a well-established rodent model of temporal lobe epilepsy. Based on previous evidence demonstrating a permanent reduction in calbindin mRNA and protein expression from the hippocampus of rats following pilocarpine treatment (and a similar reduction in

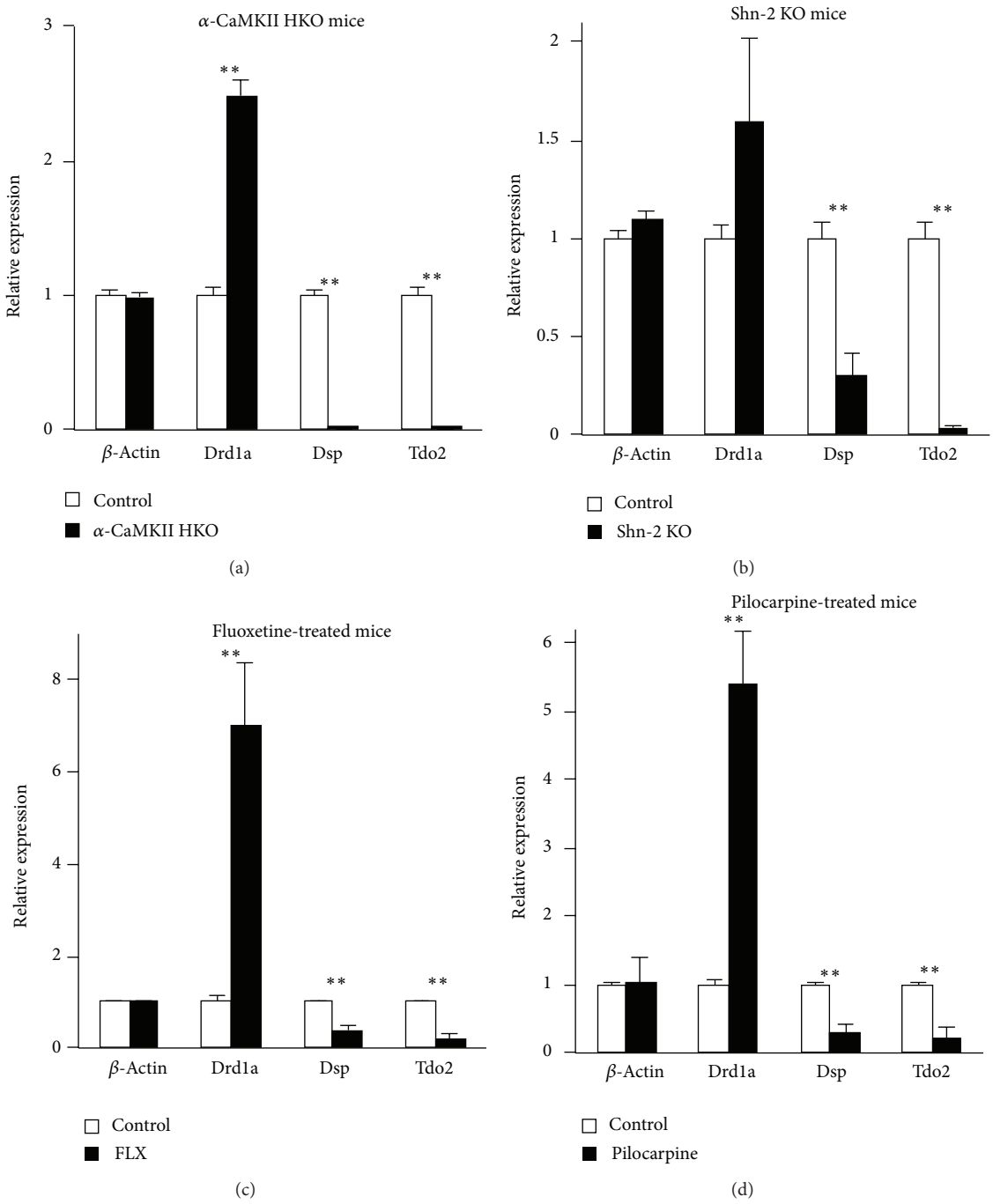


FIGURE 5: Identification of the iDG phenotype using real-time polymerase chain reaction. iDG is characterized by upregulation of *dopamine receptor D1a* (*Drd1a*) and downregulation of both *desmoplakin* (*Dsp*) and *tryptophan 2,3-dioxygenase* (*Tdo2*) in the hippocampus. Asterisks indicate statistical significance determined using the Student's *t*-test ($^{**}P < 0.01$, $n = 4$–7 per group). Adapted from Yamasaki et al. [1], Takao et al. [2], Kobayashi et al. [5], and Shin et al. [6].

human epileptic brains post mortem), it was hypothesized that mice with pilocarpine-induced seizures also exhibit an iDG phenotype. Establishment of the iDG phenotype in this mouse model of epilepsy was expected to further support the link between the pathophysiologies of epilepsy and psychosis. Molecular markers for the iDG phenotype were demonstrated in mice with pilocarpine-induced seizures, including dysregulated gene/protein expression of calretinin/calbindin, as well as hallmark alterations in other iDG markers such as *Drd1a*, *Tdo2*, and *Dsp*. Mice with pilocarpine-induced seizures also displayed characteristic iDG phenotypes at the electrophysiological levels, including decreased polarization of resting membrane potentials, lower spike threshold currents in their DG granule cells [6]. Although these

Table 1: Behavioral, electrophysiological, and molecular phenotypes in mice with iDG.

		α-CaMKII HKO [1]	Shn-2 KO [2]	SNAP-25 KI [3]	CN KO [4, 10, 21]	FLX treatment [5]	Pilocarpine treatment [6]
Behavioral phenotypes	Locomotor activity	↑	↑	↑	↑	↑↓ [29]	↑
	Working memory	↓ [1–12]	↓	↓	↓		↓ [6, 30]
	Social interaction	Mutants kill cagemates	↓	↓	↓		↓
	PPI	—	↓	↓	↓	—	
Electrophysiological phenotypes	*Mossy fibre-CA3 synapse*						
	Basal transmission	↑	↑	—		—	
	Frequency facilitation	↓	↓	↓		↓	—
	Granule cell soma						
	Passive						
	Input resistance	↑	↑	—		—	—
	Active						
	Excitability	↑	↑	↑		↑	↑
	Spike number	↓	↓	↑		↑	↑
Molecular phenotypes	*Markers of immature granule cells*						
	Dopamine D1 receptor (Drd1a)	↑	—	↑	↑	↑ [31]	↑
	Calretinin	↑	↑	↑	↑	↑	↑
	Markers of mature granule cells						
	Tryptophan 2,3-dioxygenase (Tdo2)	↓	↓	↓	↓	↓	↓
	Desmoplakin (Dsp)	↓	↓	↓	↓	↓	↓
	Calbindin	↓	↓	↓	↓	↓	↓
	GluR1	↓ [32]	↓		↓	—	
	Immediate early genes induction						
	c-Fos or Arc-dVenus	↓ [1, 13]	↓	↓	↓	↓	
	Inflammation-related molecules						
	Glial fibrillary acidic protein (GFAP)	—	↑	↑	↑	↑ [31]	↑ [33]
	Complement components	—	↑	↑		↑ [31]	↑ [34]
	MHC genes	—	↑	↑		↑ [31]	↑ [34]
	Adult neurogenesis						
	BrdU incorporation	↑		↓	↑	↑ [25]	↑ [33, 35]

↑: Increased or upregulated.
↓: Decreased or downregulated.
—: Nonsignificant.

characteristics resemble those found in α-CaMKII HKO, DG granule cells in pilocarpine-treated mice exhibited a significant increase in repetitive spiking by sustained depolarizing currents. This was in sharp contrast to the α-CaMKII HKO, which showed relatively few spikes following the same stimulation protocol. Granule cells from pilocarpine-treated mice resemble neurons observed in chronic FLX-treated mice. Therefore, similar to what was observed as a result of pilocarpine treatment in mice, seizure-induced changes in the epileptic brain may lead to a "dematuration" of DG granule cells resulting in an iDG [6]. Remarkably, the behavioral phenotypes of mice with pilocarpine-induced seizures are similar to those found in earlier iDG mouse models, specifically hyperlocomotor activity, working memory deficits, and social withdrawal.

5. iDG Phenotype and Human Psychiatric Disorders

The discovery of an iDG phenotype in multiple strains of mice with behavioral abnormalities suggests a link between iDG and the distinct behavioral traits characteristic of schizophrenia and other psychiatric disorders. To assess whether iDG is related to human psychiatric disorders, comprehensive gene expression data from 166 hippocampi were analyzed from human brains postmortem, including brains

of 21 patients with psychiatric disorders. Comprehensive gene expression analysis was performed using a specific biomarker set derived from α-CaMKII HKO mice, including 10 genes selected (*ADCY8*, *CCND1*, *LOC151835*, *LOC284018*, *NTNG1*, *PDYN*, *PIP3-E*, *PNCK*, *SPATA13,* and *TDO2*) due to differential expression in the iDG mutant hippocampus. Statistical clustering of the gene expression data was performed using all 166 hippocampi from human brains collected postmortem. The analysis roughly classified subjects into 2 clusters, one of which (schizophrenia-enriched cluster) contained 19 of the 21 patients with psychiatric disorders (including schizophrenia, schizoaffective, and bipolar patients) [1]. Furthermore, the gene expression profile was compared between the patients with schizophrenia in the schizophrenia-enriched cluster and the individuals with no major psychiatric diagnosis, and 26 differentially expressed probes were identified in the patients with schizophrenia. Interestingly, 12 of the 26 identified genes were related to neurogenesis and cell-maturation/migration; these 12 included calbindin whose expression was reduced by >60% in the schizophrenia-enriched cluster. In a similar, independent study, Altar et al. conducted DG transcriptome analysis in human patients with schizophrenia [43] and indicated that calbindin was significantly downregulated in the DG of these patients.

Patients with schizophrenia and those with bipolar disorder have also been examined for the molecular characteristics of the iDG phenotype. Postmortem immunohistochemical analysis of brain tissue revealed that patients with schizophrenia and those with bipolar disorder display significantly elevated calretinin expression in the DG, compared to both control subjects and patients having major depression. The increase of calretinin is strongly correlated with a history of psychosis. Moreover, compared to both control subjects and patients having major depression, patients with schizophrenia and those with bipolar disorder have significantly elevated ratio of calretinin to calbindin [7]. These facts demonstrate that an iDG-like condition may be also found in human patients with schizophrenia and/or bipolar disorder.

A mouse model of schizophrenia that expresses the iDG phenotype (Shn-2 KO mice) has been shown to display a series of brain phenotypes found in schizophrenia. These phenotypes include decreased gamma-aminobutyric acidergic (GABAergic) neuronal molecules (i.e., PV, glutamic acid decarboxylase isoform 67 (GAD67), *Gabra1*, etc.), reduction of oligodendrocyte markers (myelin basic protein and 2′,3′-cyclic-nucleotide 3′-phosphodiesterase (CNPase)), thinner cortex, and abnormal electroencephalographic findings (increased theta waves and decreased gamma waves). The gene expression pattern in Shn-2 KO mice was compared with that in patients with mental disorders obtained from publicly available array data using the bioinformatics tool, NextBio (Cupertino, CA, USA). NextBio is a repository of analyzed microarray datasets that allows an investigator to search results and the expression profiles of publicly available microarray datasets. This analysis determined that the highest degree of gene expression overlap was detected from Broadmann area 10 (BA10) in postmortem tissue from patients with schizophrenia and from control subjects, with 100 genes commonly altered in both Shn-2 KO mice and patients with schizophrenia [2]. The similarity was extraordinal ($P = 9.5 \times 10^{-14}$), which is unlikely to be obtained by chance. Furthermore, the expression levels of 76 of these 100 genes were altered in the same directions (Figures 6(a) and 6(b)). These findings support the idea that iDG or an equivalent phenomenon could also be present in human brain.

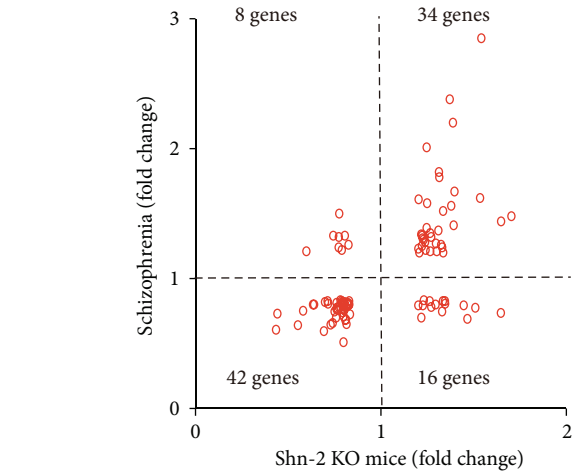
6. Maturation Abnormalities of Neurons other than DG Granule Cells in Humans with Psychiatric Disorders

Growing evidence from postmortem research implicates abnormal neurodevelopment in the pathogenesis of schizophrenia and other psychiatric disorders. In addition to detection of the iDG-like condition mentioned earlier, immaturity of neurons other than DG granule cells has also been shown in the brains of patients with psychiatric disorders.

The maturation of GABA signaling is characterized by progressive switches in expression from GAD25 to GAD67 and from NKCC1 to KCC2 in the prefrontal cortex (PFC) and hippocampus of human brain. GAD25 is predominantly expressed in the fetus, whereas GAD67 is stably expressed across development [44]. GABA synthesis is increased concurrently with the switch from GAD25 to GAD67. The cation-chloride cotransporters, NKCC1 and KCC2, contribute to change of GABA from an excitatory to an inhibitory neurotransmitter in developing brain. NKCC1 expression predominates early in the developmental period and KCC2 expression rises as brain development progresses [44, 45]. In the hippocampus of patients with schizophrenia, GAD25/GAD67 and NKCC1/KCC2 ratios are increased and KCC2 expression is significantly decreased, indicating a potentially immature state in a certain type of GABAergic neurons [44].

Decreased levels of PV have been shown in brains of individuals with schizophrenia [46–49] and bipolar disorder [50]. Because PV is known to be a marker of mature FS inhibitory neurons, FS cells were hypothesized to be immature in these psychiatric disorders [51, 52]. In this context, Gandal et al. developed an FS cell maturation index. Using time-course gene expression data from developing FS cells that were positively correlated with PV expression levels, they showed a reduction of the index in cortices of patients with schizophrenia, bipolar disorder, and autism [52]. These results suggest that FS neurons are arrested in an immature-like state in the cortices of patients with these psychiatric disorders.

Furthermore, reduction of PNNs has been reported in the lateral nucleus of the amygdala and in layer II of the lateral entorhinal cortex of subjects with schizophrenia [38, 53]. PNNs are chondroitin sulfate proteoglycan-(CSPG-) containing neural extracellular matrices and are particularly enriched around PV-positive inhibitory neurons. Because numbers of PV-positive cells are normal within these regions in schizophrenia, the reduction of PNNs indicate downregulated CSPG levels, rather than loss of PNN-associated neurons [53, 54]. Disruption of PNN formation is thought to lead maturation abnormalities in selective neuronal populations, including those expressing PV [39, 40]. Since PNNs develop



	Gene name	Fold change	
		Shn-2 KO	Schizophrenia
*	Serpina3n	1.54	2.85
*	C1qb	1.37	2.38
*	C1qc	1.39	2.20
	C10orf10	1.25	2.01
*	Rgs4	−1.25	−1.96
*	Ifitm2	1.31	1.82
*	C1qa	1.31	1.78
	Myt1l	1.22	−1.70
*	Tac1	−1.44	−1.68
*	Cebpd	1.40	1.67
	Frzb	−2.30	−1.65
*	Ifitm1	1.53	1.62
*	Tgfbr1	1.20	1.61
	Frzb	−2.30	−1.59
*	Ddit4	1.25	1.58
*	Ifitm1	1.53	1.58
	CbIn4	−1.81	−1.56
*	Tgm2	1.38	1.56
	Ma12	−1.35	−1.52
	Sic14a1	1.34	1.52
	Mthfd2	−1.29	1.50
*	Gfap	1.70	1.48
	Rgs4	−1.25	−1.48
*	D0H4S114	−1.24	−1.47
	Fbxo9	−1.23	−1.46
	Syn2	1.47	−1.45
	Gadd45b	1.65	1.44
	Gabral	−1.32	−1.43
	ChmI	1.22	−1.43
*	Cebpb	1.39	1.41
	Car10	1.27	−1.41
	Rab3b	−1.25	−1.41
	Tasp1	−1.24	−1.40
	Hk2	1.25	1.39
	RP11–35N6.1	−1.20	−1.38
	Gng4	−2.26	−1.37
	Ms4a6b	1.31	1.37
*	Klf10	1.65	−1.36
	Fcgr3	1.27	1.35
	Nrxn3	−1.22	−1.35

(b)

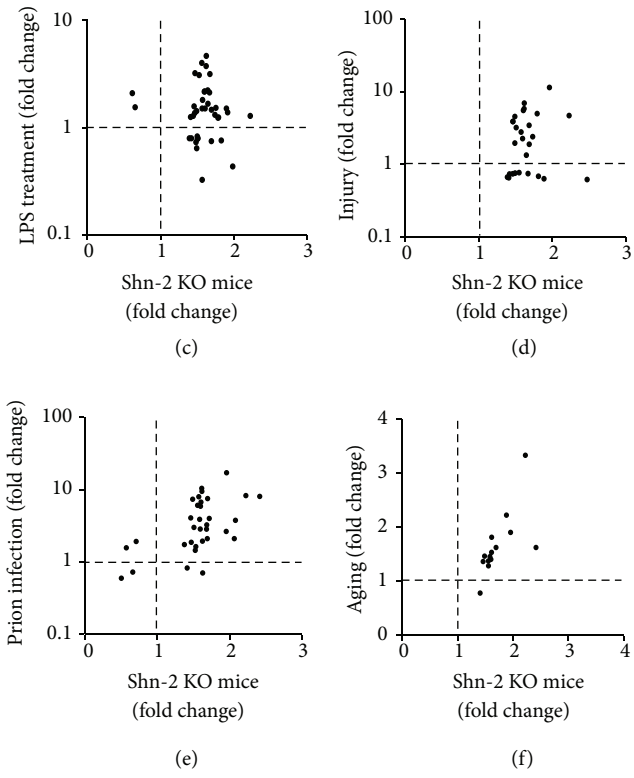


Figure 6: Comparison of gene expression profiles between Shn-2 KO mice and individuals with schizophrenia. (a) Scatter plot of gene expression fold change values in the medial prefrontal cortex (mPFC) of Shn-2 KO mice and Brodmann area (BA) 10 of postmortem schizophrenia brain. (b) Genes differentially expressed in both Shn-2 KO mice and in schizophrenia. Red indicates gene upregulation and blue indicates downregulation in both Shn-2 KO mice and in schizophrenia. The top 40 genes with respect to the fold change values are included. Asterisks indicate inflammation-related genes. (c)–(f) The hippocampal transcriptome pattern of Shn-2 KO mice was similar to the transcriptome data from lipopolysaccharide (LPS) treatment ((c), $P = 5.6 \times 10^{-9}$), injury ((d), $P = 5.7 \times 10^{-24}$), prion infection ((e), $P = 1.0 \times 10^{-18}$), and aging ((f), $P = 1.4 \times 10^{-26}$). Adapted from Takao et al. [2].

late in postnatal development and coincide with the maturation of PV-positive neurons [38, 40], the reduction of PNNs may imply that the PV-positive neurons remain at an immature-like state in the amygdala and entorhinal cortex of subjects with schizophrenia.

Taken together, these findings suggest that immaturity of neurons in adult brain can be seen, not only in the DG, but also in other brain regions in humans with psychiatric disorders. Further studies that focus on defining the maturation state of cells could identify maturation abnormalities of cells in additional brain regions.

7. Potential Mechanisms Underlying iDG

When attempting to fully understand the iDG phenotype, a logical next question is: what are the underlying mechanisms causing iDG? Both genetic and pharmacological manipulations have been shown to induce the iDG phenotype, and these factors could provide clues towards identifying the mechanisms behind this phenotype.

7.1. Inflammation. One clue into these mechanisms may lie in the normal functions of genes that, when altered, produce mouse models that express the iDG phenotype. For example, Shn-2 can be linked to the inflammatory cascade in that Shn-2 was originally identified as a nuclear factor-kappa B (NF-κB) site-binding protein that tightly binds to the enhancers of major histocompatibility complex (MHC) genes in the MHC regions of chromosome 6 [55]. Recent genome-wide association studies have identified a number of single nucleotide polymorphisms (SNPs) in the MHC region associated with schizophrenia [56–60]. There are several MHC class I genes (*HLA-A*, *HLA-B*, etc.), each with similar promoter structures that have 2 NF-κB binding motifs [61]. Upon inflammation, NF-κB is known to bind to these enhancers and to initiate transcription of genes involved in immune functions. Shn-2 binds these motifs but does not substitute for the transcriptional function of NF-κB [62]. Instead, Shn-2 competes with NF-κB for binding to the motifs and inhibits NF-κB activity. It has been reported that Shn-2 KO mice have constitutive NF-κB activation and that T-cells of these mice show dramatic enhancement in differentiation towards T-helper Type 2 cells (Th2), a phenomenon called "Th2 slant," [63] which is observed in patients with schizophrenia [64]. Thus, Shn-2 is an inhibitor of NF-κB, a major mediator of inflammation.

Due to the link between Shn-2 and inflammation, altered gene expression patterns were expected in Shn-2 KO mice. Using the bioinformatics tool, NextBio, publicly available datasets were analyzed to find the conditions where transcriptome patterns are similar to those found in the PFC of Shn-2 KO mice. Gene expression patterns in the PFC of Shn-2 KO mice were found to be similar to those seen in inherently inflammatory conditions (e.g., prion infection, malaria encephalopathy, spinal cord injury, brain injury, and kidney tissue treated with lipopolysaccharide) (Figures 6(c)–6(f)). However, this analysis highlighted that the scale of inflammation for Shn-2 KO mice is quite different from other inflammatory conditions (i.e., the inflammation in Shn-2 KO mice is much weaker in terms of the magnitude of the changes). Additionally, it was found that many of the genes with altered expression in both Shn-2 KO mice and patients with schizophrenia were inflammation-related genes (Figure 6(b)). Astrocytic activation is considered an indication of inflammation and GFAP, a reactive astrocyte marker, is increased not only in Shn-2 KO mice but also in SNAP-25 KI [3], FLX-treated [65], and pilocarpine-treated mice [33]. Therefore, the iDG phenotype can be associated with inflammation-like phenomena.

7.2. Neuronal Hyperexcitation. In pilocarpine-induced epilepsy mouse models, a single episode of status epilepticus (SE) was not enough to induce the iDG phenotype. Chronic models of animal epilepsy develop spontaneous recurrent seizures (SRS) following SE over a period of months or years. In mice with pilocarpine-induced epilepsy, the markers of the iDG phenotype were identified in mice that developed SRS following SE, but not in mice that, after pilocarpine-induced SE, did not develop SRS [6]. These findings have 2 potential interpretations: (1) tonic/chronic hyperexcitation of hippocampal neurocircuits leading to SRS is needed to induce the iDG phenotype, or (2) the iDG phenotype itself is necessary for the development of SRS. However, a representative model of iDG (α-CaMKII HKO mice) has a more than 4-fold increase in sensitivity to pilocarpine for triggering seizures, but not developing SRS.

Recently, it has been reported that SNAP-25 KI mice sometimes exhibit spontaneously occurring convulsive seizures after postnatal days 21–24 [66]. These seizures can be suppressed with chronic administration of valproic acid, an anticonvulsant, starting at postnatal day 16 [67]. These findings led to an investigation of whether iDG phenotypes and behavioral abnormalities could be rescued in mutant mice by suppression of recurrent epileptic seizures with valproic acid. Results showed that in the DG of SNAP-25 KI mice decreased expression of calbindin and increased expression of calretinin could be rescued with chronic administration of valproic acid. Valproic acid treatment also reduced the previously observed increase in the size of the DG and significantly improved working memory in these mice [3]. These results suggest that recurrent epileptic seizures underlie "dematuration" of DG granule cells, which induce the iDG phenotype.

Hyperexcitability of the DG granule cells is a feature shared among mice that express the iDG phenotype. The threshold current required to induce an action potential is reduced in α-CaMKII HKO, Shn-2 KO, SNAP-25 KI, and FLX-treated mice (Table 1). Furthermore, the increased sensitivity to pilocarpine (with respect to the seizure threshold) in α-CaMKII HKO mice supports the idea that hyperexcitation induces the iDG phenotype [6]. Previous studies have reported region-specific loss of GABAergic inhibition following SE/SRS, especially from the CA1, CA3, and hilar regions in the hippocampus. Notable molecular/morphological deficits observed in iDG mouse models include a reduction of the GABAergic system in the hippocampus (e.g., a reduction of PV-positive interneurons and decrease of GAD67 expression, Takao et al. [2]). Although it is probably too simplistic

to conclude (or even to speculate) that a simple loss of GABAergic cells and/or function from the hippocampus may contribute to induction of the iDG phenotype, these data suggest that sustained hippocampal neuronal hyperexcitation, caused by loss of GABAergic inhibition, may be a common driving force for initiating a cascade of molecular events that lead to development of the iDG phenotype. Collectively, these findings support the idea that neuronal hyperexcitation leads to "dematuration" of mature granule cells in the DG, thereby contributing to iDG phenotypes.

It should also be noted that iDG phenotypes are usually associated with mild chronic inflammation. Shn2-KO, SNAP-25 KI, pilocarpine-treated, FLX-treated, and paroxetine-treated mice commonly demonstrate upregulated expression of complement and MHC genes and GFAP, suggesting that neuronal hyperexcitation is a source of inflammatory-like processes in the brains of these mice. In contrast, recent research has highlighted the involvement of the immune system in the development of epilepsy. For instance, inflammatory cytokines are increased in the central nervous system (CNS) and plasma of epilepsy model animals and of patients with epilepsy [68, 69]. Moreover, inflammation in the CNS is associated with damage to the blood-brain barrier (potentially leading to leakage) that is known to enhance neuronal excitability and has been implicated in epileptogenesis [70–72]. In this context, Fabene et al. demonstrated that pilocarpine-induced SE enhances leukocytic inflammatory changes in the CNS vasculature, consequently generating epileptic activity [73]. Based on these data, a possible positive feedback mechanism between neuronal hyperexcitation and inflammatory conditions could subsequently induce iDG phenotypes.

More recently, it has been reported that knockdown of *Disc1* (disrupted in schizophrenia 1), specifically in adult-born DG neurons, results in enhanced excitability [74]. The *Disc1* gene was originally discovered in a Scottish family with a high incidence of psychiatric disorders, including schizophrenia and bipolar disorder [75]. *Disc1* knockdown also caused pronounced cognitive and affective deficits, which could be reversed when affected DG neurons were inactivated [74]. These findings are consistent with the hypothesis that hyperexcitation of DG neurons may underlie behavioral abnormalities related to schizophrenia and other neuropsychiatric disorders.

7.3. Other Clues. The iDG phenotype is associated with several other cellular/molecular alterations, and these changes may serve as clues to the mechanisms underlying the phenotype. The GluR1 subunit of α-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid-type glutamate receptor is dramatically reduced in most, but not all, mice that express the iDG phenotype. For example, it has been reported that GluR1 expression is reduced in the DG of α-CaMKII HKO [32] and Shn-2 KO [2]. Interestingly, GluR1 is expressed primarily in mature granule cells and can be used as a marker for maturation of granule cells [32]. Physiologically, Ca^{2+}-permeability mediated by GluR1 and/or GluR3 subunits is strongly associated with granule cell maturation [76–78]. The GluR1 reduction demonstrated in these mutants may be consistent with the idea that DG granule cells are immature in these mice.

Arc and c-Fos are immediate-early genes (IEGs) that are commonly used as markers for the maturity of in vivo activity-dependent responsiveness of granule cells, whose expression can be stimulus-induced in 3- to 5-week-old cells [79, 80]. As mentioned previously, Arc promoter activity was dramatically reduced in the DG of α-CaMKII mice after performing working memory tasks [13]. Likewise, Arc induction after exposure to a novel environment or receiving electrical foot shocks was reduced in the DG of Shn-2 KO [2] and SNAP-25 KI mice [3]. In FLX-treated mice, expression of c-Fos was reduced after receiving foot shocks [5]. Since activity-dependent processes have been implicated in the maturation of adult neural progenitors, if reduction of IEG expression (or, conversely, promotion of expression) represents a state of low-responsiveness to behavioral stimulations in the granule cells of mice expressing the iDG phenotype, activity-dependent maturational mechanisms are also likely impaired in these cells. However, granule cells are more excitable in α-CaMKII HKO [1], Shn-2 KO [2], SNAP-25 KI [3], and FLX-treated mice [5]. Therefore, suppressed IEG induction in mice expressing the iDG phenotype might be the result of decreased activity and/or impairments in activity-dependent gene regulation in the DG.

Adult neurogenesis in the DG was assessed using incorporation of 5-bromo-2′-deoxyuridine (BrdU) and was elevated in α-CaMKII HKO [1], FLX-treated [25], and pilocarpine-treated mice. In the pilocarpine model of epilepsy, BrdU incorporation was increased in the acute phase after SE [33], as well as in the period of SRS [35]. However, despite the occurrence of epileptic seizures in SNAP-25 KI mice, reduced adult neurogenesis was observed in these mutants [3]. Further studies are needed to fully assess whether altered adult neurogenesis is involved in formation of the iDG phenotype. Electrophysiologically, an increase in granule cell excitability and a reduction of frequency facilitation at mossy fibre-CA3 synapses were found in α-CaMKII HKO, Shn-2 KO, SNAP-25 KI, and FLX-treated mice. These shared molecular and physiological alternations in mice with the iDG phenotype may provide important clues to investigate the mechanisms underlying iDG.

Table 1 summarizes the behavioral, electrophysiological, and molecular patterns in different mice with the iDG phenotype. Some endophenotypes are perfectly shared, but others are not. There appear to be subgroups of the iDG phenotype, which may potentially correspond to different types of psychiatric disorders. Furthermore, there are likely some shared and some unshared mechanisms between the different groups of mice with the iDG phenotype.

8. Face and Construct Validity of iDG Mouse Models as Models of Psychiatric Disorders

Mice with the iDG phenotype possess face validity as animal models of schizophrenia at both the behavioral and molecular levels. In addition, various phenomena accompanied by the iDG phenotype are in good agreement with most of the major

hypotheses of the disorder, supporting the construct validity of these models.

8.1. Face Validity. As mentioned earlier, mutant mice with the iDG phenotype (CN KO, α-CaMKII HKO, Shn-2 KO, and SNAP-25 KI mice) have been identified as potential models of schizophrenia and bipolar disorder, originally by behavioral screening designed to assess the face validity of this model. These mutant mice strains commonly exhibited severe working memory deficits, decreased PPI, impaired social behaviors, and hyperactivity. Impairments in working memory [81, 82], PPI [83], and social withdrawal [84] are prominent features of schizophrenia symptomatology. Hyperactivity is characteristic of rodent models of schizophrenia [85] and could correspond to the psychomotor agitation often present in patients with schizophrenia. Furthermore, FLX-treated and pilocarpine-treated mice express iDG and behavioral phenotypes shown by mutant iDG mice (Table 1). Therefore, when evaluated at a behavioral level, mice expressing the iDG phenotype display a strikingly conserved behavioral phenotype.

Among these models, Shn-2 KO mice showed outstanding face validity as an animal model of schizophrenia. In addition, with respect to molecular profiles, Shn-2 KO mice shared altered molecular expression patterns with postmortem brain tissue from patients with schizophrenia. Significant similarities in expression direction and magnitude were detected when the gene expression pattern in Shn-2 KO medial PFC (mPFC) was compared with the pattern in schizophrenic frontal cortex (Brodmann area 10, BA10) examined postmortem [86]; 100 altered genes were detected in both Shn-2 KO mice and patients with schizophrenia. Of the 100 altered genes, 76 showed the same directional change in expression and, of the 76 genes, 42 were downregulated and 34 were upregulated [2]. A large number of genes previously implicated in schizophrenia or bipolar disorder were included in these groups. For example, 89 of the 100 similarly expressed genes have been identified as related to these disorders. Shn-2 KO mice also displayed decreased levels of PV in the frontal cortex, which has been widely observed in schizophrenia [46–49]. Fast-spiking PV-positive interneurons have been shown to be immature in the cortices of patients with schizophrenia, bipolar disorder, and autism [52]. This suggests that immaturity of this type of neuron in the cortex may underlie some of the cognitive deficits seen in these disorders. Shn-2 KO mice also exhibit decreased expression of PV and GAD67 in the hippocampus, which has been observed in the brains of patients with schizophrenia examined postmortem [87–89]. Furthermore, Shn-2 KO mice had significantly thinner cortex and reduced cell density in the prelimbic and primary visual cortices, consistent with observations in human patients with schizophrenia [90].

In addition to anatomical abnormalities, the cortex of Shn-2 KO mice exhibits physiological alternations. Cortical EEG analysis in mutant mice showed an increase in slow waves and a decrease in fast waves, both of which are observed in patients with schizophrenia [91–93]. Although present studies have evaluated the molecular and physiological validity using Shn-2 KO mice in detail, future studies are needed to address whether other mouse strains that express the iDG phenotype similarly fulfill the criteria for face validity.

8.2. Hyperdopaminergic/Hypoglutamatergic Hypotheses. The longstanding "dopamine hypothesis of schizophrenia" is based primarily on 2 facts: (1) all antipsychotics currently available to treat the positive symptoms of schizophrenia possess dopamine D2 receptor blocking activity and (2) hyperactive dopamine release in subcortical limbic brain regions, especially in the nucleus accumbens/striatum, has been consistently observed in patients with schizophrenia [94, 95]. Dopamine D2 blockers are widely used for treating bipolar mania and hyperactivity of the dopaminergic system during manic phases has also been proposed [96]. A hyperdopaminergic state in subcortical regions is believed to be associated with psychosis, a core constituent of positive symptoms of schizophrenia and a phenotype often observed in manic phase of the bipolar disorder, and a symptom that can be ameliorated using treatment with D2 blockers.

Intriguingly, α-CaMKII HKO mice have been proposed as a model of schizophrenia by Novak and Seeman [97], based on their finding that D2High receptors were elevated in the striatum (representing hyperdopaminergic state analogous to schizophrenia) of this mutant strain. Increasing evidence also supports that mice with iDG phenotypes are in a hyperdopaminergic state. For example, administration of haloperidol, a typical antipsychotic, significantly improved PPI impairment in Shn-2 KO mice [2]. Currently available antipsychotics (including haloperidol) work primarily by antagonizing dopamine D2 receptors and raising intracellular cAMP levels. Therefore, intracellular stimulation of cAMP levels is thought to have effects similar to treatment with antipsychotic medication [98, 99]. Rolipram is a cAMP-specific phosphodiesterase inhibitor that elevates intracellular cAMP levels. Chronic treatment with rolipram (in combination with ibuprofen) rescued a subset of mice expressing the iDG phenotype and improved working memory and nest building in Shn-2 KO mice. These findings suggest that lower intracellular cAMP levels are involved in a subset of behavioral abnormalities and iDG phenotypes.

Interestingly, dopamine D1 signaling is also upregulated in the DG of α-CaMKII HKO and Shn-2 KO mice, when assessed using receptor binding assay and receptor agonist sensitivity, respectively. Increased expression of *Drd1a* mRNA also suggests elevated D1 signaling in α-CaMKII [1], SNAP-25 KI [3], FLX-treated [65], and pilocarpine-treated mice [6]. Upregulated D1 signaling might serve as a compensatory mechanism to normalize intracellular cAMP levels in these mice. These results suggest the presence of a hyperdopaminergic state in the brains of mice with the iDG phenotype.

This hyperdopaminergic state is thought to be a consequence of NMDA receptor hypofunction. This assertion is supported by findings showing that acute application of an NMDA receptor antagonist stimulates dopamine release in both animal models and humans. CaMKII is a major downstream molecule of the NMDA receptor and plays an important role in long-term potentiation and long-term depression [100]. Since deficiency of α-CaMKII causes schizophrenia-related behavioral and

iDG phenotypes, NMDA receptor hypofunction might be involved in the formation of the iDG phenotype. In addition, in α-CaMKII HKO mice, NMDA receptor- binding was also downregulated in the hippocampus, especially in the DG [1], supporting the idea of NMDA receptor hypofunction in these mutant mice. Additional evidence for this hypothesis includes the fact that MK-801, an NMDA receptor antagonist that causes schizophrenia-like psychosis in humans, produced significantly higher levels of drug-stimulated motor activation in CN KO [10] and Shn-2 KO mice [2]. Taken together, these findings suggest that hypoglutamatergic/hyperdopaminergic function is involved in the iDG phenotype.

8.3. Neurodevelopmental/Neurogenesis Hypothesis. The neurodevelopmental hypothesis is one of the major theories regarding the pathogenesis of schizophrenia [101–103]. This hypothesis is supported by several lines of evidence, including increased frequency of obstetric complications in patients with schizophrenia, the presence of neurological, cognitive, and behavioral dysfunction long before illness onset, the absence of postmortem evidence of neurodegeneration, and the induction of schizophrenia-like phenomena by neonatal hippocampal lesions in model animals [101, 103–105]. These findings are consistent with a neurodevelopmental model in which the etiology of schizophrenia may involve pathologic processes caused by both genetic and environmental factors that begin before adolescence when the brain approaches its adult anatomical state.

The iDG phenotype literally reflects a neurodevelopmental problem in which DG granule cells arrest in a pseudoimmature state. Takao et al. revealed that iDG phenotypes emerge at 2–4 weeks of age in Shn-2 KO mice [2]. In 2-week-old mice, there were no significant differences in the expression of calbindin and calretinin in the DG of Shn-2 KO and wild-type mice. However, when mice were evaluated at 1 month of age, calbindin expression was decreased and calretinin was increased in the DG of Shn-2 KO mice, indicating the postnatal (possibly adolescent) emergence of an iDG phenotype.

Specifically, the number of calbindin-positive cells was decreased at age of 4 weeks, compared to age of 2 weeks in Shn-2 KO mice, whereas, in wild-type mice, these cells increased throughout development. Even in adult wild-type mice, "dematuration" of mature granule cells and schizophrenia-related behavioral phenotypes, could be induced by chronic FLX treatment [5] or with pilocarpine-induced SRS [6]. These findings suggest that genetic and/or environmental factors could induce the maturational abnormalities of DG granule cells during the postnatal developmental period (and during adulthood).

Elevated neurogenesis has been defined as a core feature of the pathology associated with the iDG phenotype, with few exceptions (see [3]). One study supports a linkage between schizophrenia and decreased hippocampal neurogenesis, as tissues from human patients with schizophrenia had fewer proliferating Ki-67 cells than tissues from control subjects [112]. However, that study excluded several patients with neurogenesis levels greater than 5-fold baseline levels, which may represent a subset of patients with schizophrenia/bipolar disorder that display hyperactive neurogenesis.

8.4. Inflammation Hypothesis. The well-established role of inflammation in the etiology of schizophrenia is often referred to as the inflammation hypothesis [64, 113–115]. The link between prenatal infection and schizophrenia was first identified in an epidemiological study demonstrating increased schizophrenia risk in the offspring of women exposed to influenza during pregnancy [64]. Several other infectious factors have also been implicated in the pathogenesis of schizophrenia [64], suggesting that the disease may result from maternal immune response to infection. To this end, prenatal treatment with polyinosinic:polycytidylic acid (poly I:C), viral infection, and LPS treatment are used as rodent models of schizophrenia [114, 116–119]. As mentioned previously, Shn-2 KO mice were shown to exhibit mild chronic inflammation of the brain, as evidenced by increased inflammation markers (including complement and MHC class I genes, GFAP, and NADH/NADPH oxidase p22-phox) and genome-wide gene expression patterns similar to various inflammatory conditions [2]. Similarly, SNAP-25 KI mice, which develop epileptic seizures, showed increased expression of complement and MHC class I genes, and *GFAP* in the hippocampus [3]. In FLX-treated [65], paroxetine-treated [31], and pilocarpine-treated animals [34], gene chip data reveals hippocampal upregulation of complement and MHC genes, and *GFAP*. Therefore, the changes in inflammation-related molecules in these mice with iDG are consistent with inflammatory features found in the brains of patients with schizophrenia and epilepsy.

8.5. Collection of Genetic Association Study Results. Schizophrenia is highly heritable and multiple genetic and environmental factors are probably be involved [120]. Mice with iDG phenotype exhibit significantly altered expression of genes that have been reported to associate with schizophrenia.

For example, mutated genes that cause iDG phenotype have been implicated in schizophrenia by association studies. Based on the observation that CN KO mice display a spectrum of behavioral abnormalities strikingly similar to those observed in patients with schizophrenia [21], Gerber et al. examined whether CN dysfunction is involved in the etiology of schizophrenia [9]. By conducting transmission disequilibrium studies in a large sample of affected families, they reported evidence supporting an association between the *PPP3CC* gene encoding the CNA-gamma catalytic subunit and schizophrenia in populations from the United States and South Africa. The allelic and haplotypic associations of *PPP3CC* with schizophrenia were later confirmed in populations from Japan [20]. Thus, CN KO mice have good construct and face validity as a model for schizophrenia.

Moreover, recent genome-wide association studies have identified a number of SNPs in the MHC region associated with schizophrenia [56–59]. Many of these SNPs are located in or near NF-κB binding sites that may be associated with susceptibility to schizophrenia [2]. Some of the genes surrounding these SNPs are dysregulated in the brains of

patients with schizophrenia examined postmortem and/or the PFC of Shn-2 KO mice. As Shn-2 is a MHC enhancer-binding protein that binds to the NF-κB binding motif, abnormal transcription of these genes may be induced by dysregulated NF-κB signaling pathways, independent of any deficiency or mutation in Shn-2 itself.

In addition, recent human genetic studies have discovered associations between SNAP-25 and various psychiatric and neurological disorders, including schizophrenia [14, 15], ADHD [16, 17], and epilepsy [18, 19]. Translational convergent functional genomic study demonstrates that SNAP-25 is one of the top 42 candidate genes for schizophrenia [121]. Therefore, SNAP-25 KI mice also have strong construct validity for schizophrenia.

Many of the genes with decreased expression in both schizophrenic BA10 and Shn-2 KO mPFC have been reported to be associated with schizophrenia. Chowdari et al. reported that several SNPs in *RGS4*, regulator of G-protein signaling-4, are associated with schizophrenia [122]. It has been previously shown that *RGS4* expression is decreased across 3 cortical areas (including the PFC) of subjects with schizophrenia [123]. Consistent with these observations, *RGS4* mRNA expression was decreased in the mPFC of Shn-2 KO mice [2]. Additionally, significant associations were detected between samples from patients with schizophrenia and SNPs in *Cbln4* [57], *Gabra1* [124], and *Nrxn3* [57]. These genes are involved in synaptic transmission or synaptic plasticity, and expression of these genes is commonly decreased in both schizophrenic BA10 and Shn-2 KO mPFC. Thus, decreased expression of genes associated with schizophrenia could play a significant role in developing schizophrenic conditions in the PFC of iDG mice, especially of Shn-2 KO mice.

Genes related to neuronal excitation and inflammatory responses have been reported to be associated with schizophrenia [125]. As previously mentioned, these conditions are closely related to induction of the iDG phenotype. A meta-analysis of clinical data showed an association between SNPs in *KCNH2* (a human Ether-à-go-go-family potassium channel) and schizophrenia [126]. The *KCNH2*-3.1 isoform modulates neuronal firing, and its expression is increased in the hippocampi from individuals with schizophrenia, suggesting increased neuronal excitability in the schizophrenic brain [126].

Regarding inflammatory or immune response-related genes, as previously mentioned, a number of SNPs in the MHC region are associated with schizophrenia. In addition, many inflammatory response-related genes such as *C1QA*, *C1QB* [127], *TAC1* [128, 129], *TGFBR1* [129], and *TGM2* [130] have been associated with schizophrenia. *GFAP* is also known to have a significant association with schizophrenia [15]. These mRNA expression changes were in the same direction as changes seen in schizophrenic BA10 and Shn-2 KO mPFC [2]. Thus, it has been established that altered expression of genes related to neuronal excitation and inflammatory conditions could be involved in the brains of patients with schizophrenia as well as in mice with iDG phenotype.

Taken together, these findings indicate that mice with the iDG phenotype have high construct validity as animal models of schizophrenia, with many of the genes showing a significant association with schizophrenia.

8.6. Other Factors. Some other factors, such as dysfunction of oligodendrocytes or mitochondria, have been implicated in psychiatric disorders. Several potential deficits were observed in key oligodendrocyte markers, including decreases in CNPase and myelin basic protein (MBP), which is a major constituent of the myelin sheath of oligodendrocytes and Schwann cells. Decreased CNPase protein levels have been reported in schizophrenia [131]. Similarly, *MBP* mRNA levels were decreased in the DG as well as the mPFC of Shn-2 KO mice, and proteome analysis reveled that many mitochondria-related proteins (e.g., dihydrolipoamide *S*-acetyltransferase (DLAT), ubiquinol-cytochrome *c* reductase core protein (UQCRC), and NADH dehydrogenase [ubiquinone] Fe-S protein (NDUFS)) in the DG are affected in Shn-2 KO mice [2, 132]. These findings suggest that changes in the expression of molecules related to oligodendrocytes or mitochondria in the DG of Shn-2 KO mice would be consistent with the oligodendrocyte and mitochondria hypotheses of schizophrenia, respectively.

Figure 7 shows a schematic representation of the model detailing how the iDG phenotype may be involved in schizophrenia, bipolar disorder, and other psychiatric disorders. This proposed model is based on the findings that mice with the iDG phenotype possess face and construct validity as animal models of these disorders. In these disorders, multiple genetic and environmental factors could induce mild chronic inflammation, and this could subsequently result in multiple endophenotypes (including iDG) in the brain, that may, as a whole, cause behavioral abnormalities in psychiatric patients.

9. A Possible Role of iDG in Behavioral Abnormalities Relevant to Neuropsychiatric Disorders

9.1. iDG and Working Memory. Impairment in working memory is one of the core behavioral abnormalities in mice with the iDG phenotype. CN KO [21], α-CaMKII HKO [1, 12], Shn-2KO [2], SNAP-25 KI mice [3], and pilocarpine-treated animals [30] have been reported to display severe working memory deficits in the spatial working memory version of the eight-arm radial maze and/or in the T-maze forced-alteration task.

The hippocampal structure has been shown to be critically involved in working memory function [133, 134]. The DG is a key input node for the hippocampal structure. Perforant path fibers originating in the entorhinal cortex provide a major source of highly processed sensory information to DG granule cells. When rats with lesions in the DG were tested on working memory version of the eight-arm radial maze, they showed impairment in their ability to perform this version of the task [135–137]. Considering the evidence supporting involvement of the DG in working memory function, it is likely that the iDG phenotype has a negative impact on working memory function. However, the possibility that deficits in regions other than the DG cause

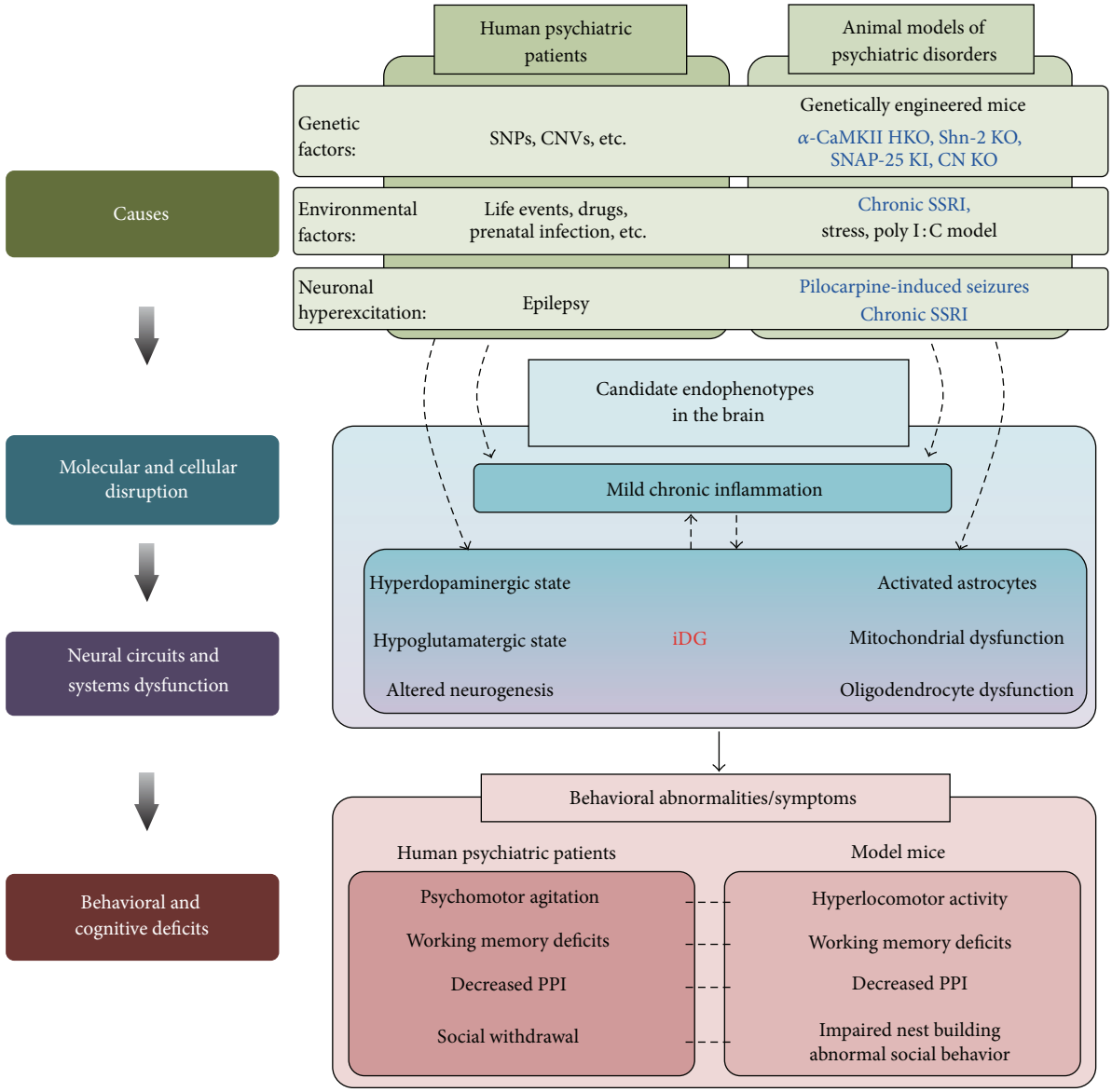


Figure 7: A schematic representation of the model of involvement of the iDG phenotype in psychiatric disorders. In psychiatric disorders, such as schizophrenia, bipolar disorder, and depression, multiple genetic and environmental factors could induce mild chronic inflammation. This could subsequently result in multiple endophenotypes in the brain, including iDG. Inflammation could induce alterations in neurogenesis [106, 107], mitochondrial dysfunction [108, 109], and oligodendrocyte dysfunction [110, 111]. Involvement of inflammation in induction of a hypoglutamatergic state or hyperdopaminergic state remains unclear. These possible endophenotypes may affect each other, which may, in turn, cause behavioral abnormalities in psychiatric patients. There may not be a single "principal" or "core" mechanism that underlies the behavioral symptom of psychiatric disorders. Indeed, a shared or preserved set of multiple endophenotypes, as a whole, may be the principal mechanisms of these disorders.

impairments in working memory in the mice with the iDG phenotype cannot be excluded.

In working memory tasks, animals must rapidly establish and maintain a memory of visited areas based on single within-trial exposures and must suppress interference by memories obtained during previous trials. Deficits in either function could cause impairments in working memory tasks. Mice with the iDG phenotype commonly display a reduction of frequency facilitation at mossy fibre-CA3 synapses [1–3, 5]. Frequency facilitation is a potential mechanism for the involvement of the CA3 region in the processing of working memory, especially in quickly encoding novel information into the hippocampal memory system [138]. Therefore, the deficit might contribute to impairments in establishment and maintenance of memory in mice with the iDG phenotype. In addition, mice with the iDG phenotype might have deficits in processes that reduce memory interference. Interference between already remembered information and to-be-remembered information can have a profound effect on mnemonic accuracy [139]. DG granule cells have been

proposed to play important roles in temporal pattern separation, in which an initial event should be separated from a later event [140, 141]. If this hypothesis proves true, mice with the iDG phenotype might have deficits in suppressing memory interference, leading to impaired working memory.

9.2. iDG, Pattern Separation, and Psychosis. Among hippocampal subfields, computational models have highlighted the essential role of the DG for disambiguating similar events. This so-called pattern separation refers to the computational process for making representations for similar input patterns more orthogonal for better discrimination [142–145]. Experimental efforts have tested these computational hypotheses using hippocampal region-selective lesions [146–149], *in vivo* electrophysiological recording [150], and genetic manipulation [151]. More recently, investigation of adult-born DG neurons in memory formation has focused on specific function-pattern integration and pattern separation [152, 153]. Marín-Burgin et al. showed enhanced excitation/inhibition balance and low input specificity in immature granule cells [153]. These electrophysiological features of immature granule cells have been proposed to be crucial for pattern integration [153]. For example, mature granule cells are presumably very selective in their responses, which may contribute to pattern separation by making the stimulus inputs independent of one another. In contrast, the physiological properties of immature granule cells probably make them less selective, which may result in firing in response to multiple events [152, 153].

As mentioned earlier, granule cells in the pseudoimmature state are hyperexcitable in α-CaMKII HKO [1], Shn-2 KO [2], SNAP-25 KI [3], and FLX-treated mice [5]. Given that the physiological properties of pseudoimmature granule cells are comparable to those of immature granule cells in normal development, it is tempting to hypothesize that, due to a lack of appropriate memory separation, the iDG phenotype causes excessive integration and association of memories to actual and imaginary events.

However, Nakashiba et al. have reported that pattern separation requires adult-born immature granule cells, not mature granule cells [154]. The role of immature and pseudoimmature granule cells on pattern separation/integration remains controversial. Therefore, future studies are needed to examine input selectivity of pseudoimmature granule cells in mice with the iDG phenotype and to address whether they exhibit deficits in pattern separation.

An iDG signature, increased calretinin immunoreactivity, has been identified from patients with schizophrenia and bipolar disorder. The increase in the calretinin signal was positively correlated with a history of psychosis in these patients [7]. Similarly, in iDG mouse models, hyperlocomotor activity, generally thought to be linked to a hyperdopaminergic/psychosis state, has been consistently observed [1–3, 10]. Indeed, α-CaMKII HKO mice show hyperlocomotion and simultaneously display hypersensitivity to amphetamine-induced hyperlocomotion (Kogan & Matsumoto, unpublished observation), a well-established behavioral trait that is believed to reflect a hyperdopaminergic/psychosis state in patients with schizophrenia/bipolar disorder. Based on this evidence, iDG may represent a pathophysiological deficit shared among patients with psychosis, a deficit presumably linked to a hyperdopaminergic state. Interestingly, iDG signatures have also been identified in pilocarpine-induced epilepsy mouse models [6]. Epidemiological evidence indicates that psychosis is often comorbid with epilepsy and vice-versa [155]. Thus, these recent findings suggest that a common hyperdopaminergic state may further strengthen the link between epilepsy and psychosis [156].

How iDG contributes to psychosis and a hyperdopaminergic state remains an open question. While not fully answered, iDG may fit as a potential player in the disturbed neurocircuits, based on recent studies that have proposed that hippocampal neurocircuits influence dopaminergic neuronal tones that underlie psychotic and schizophrenic conditions [157, 158].

The consequences of iDG (specifically, excess immature/less-mature granule cells) on the function of the hippocampus also remain open questions. It has recently been proposed that immature granule cells in the DG may inhibit or destabilize activity of the entire DG via activation of inhibitory GABAergic input from hilar interneurons [159]. Because immature granule cells preferentially innervate hilar interneurons and these interneurons innervate all DG granule cells, excess immature neurons may consequently lead to gross inhibition of the DG. Although immature granule cells in iDG models are hyperexcitable compared to mature granule cells [1–3, 5], their hyperexcitability may cause further gross inhibition of DG by the proposed feedback mechanism from hilar interneurons. A synaptic competition for perforant path connections between older, more-mature granule cells and newly generated immature cells has also been proposed as a potential cause of the gross inhibition of the DG by immature granule cells [159]. In iDG mouse models, mature granule cells are dramatically reduced. This reduction of mature neurons could be easily connected to dysfunction of the DG, especially after considering the reduced arborization of DG granule cells in iDG models [1].

In iDG models, the DG also does not respond to stimulus (e.g., lack of c-Fos expression in DG following electric foot shock in α-CaMKII HKO mice [1] and dramatically reduced Arc induction in a novel environment in Shn-2 KO mice [2]). It currently remains unclear whether DG activity is actually decreased. Furthermore, it remains unclear if decreased DG activity leads to less glutamatergic input to CA3 from the DG through mossy fibers. Although mossy fibers from the DG to CA3 are glutamatergic and come into direct contact with CA3 pyramidal neurons, many mossy fiber synapses are connected to interneurons in CA3 that provide GABAergic inhibitory input to CA3 pyramidal cells. As a consequence, DG and mossy fiber activation causes a net inhibition within the CA3 network and presumably activates only a specific subset of CA3 pyramidal neurons [160, 161]. From this vantage point, net inhibition of the DG by immature granule cells may cause a tonic net activation of CA3. Induction of Arc transcription after foot shock was greatly suppressed in almost every region of the Shn-2 KO brain. Despite this near-ubiquitous reduction of Arc, comparable Arc induction was observed in the CA1 and CA3 regions of the hippocampus in Shn-2 KO and control mice. The relatively higher Arc induction in these

regions suggests that CA areas in these mutant mice may be more active than other brain regions.

This functional failure in the hippocampal formation *per se* is proposed by Tamminga et al. [158] as an underlying mechanism of psychosis, in which hypoactive DG (weakened pattern separation) and hyperactive CA3 (strengthened pattern completion) confer psychosis (cognitive "mistakes") in patients with schizophrenia and other psychiatric conditions. Since an iDG signature was identified in patients with schizophrenia and bipolar disorder and due to its strong association with psychosis, it is conceivable that iDG is one of the potential mechanisms causing dysfunction of the DG and subsequent tonic activation of CA3-CA1 in patients with psychiatric conditions.

As discussed earlier, a subcortical hyperdopaminergic state in patients is also believed to play a pivotal role in the development of psychosis and is the only biochemical finding supported by the mechanism-of-action of currently available drugs. A hypothesis has been proposed by Grace et al. [157, 162] suggesting that overdrive of the subcortical dopamine system is caused by hyperactivation of the ventral hippocampus. Tonic activation of ventral CA3-CA1 induced by the iDG phenotype theoretically leads to activation of the ventral subiculum (vSub) through direct glutamatergic transmission. Glutamatergic afferents to the nucleus accumbens (NAc) from the vSub then activate GABAergic projections from the NAc to the ventral pallidum, suppressing pallidal GABAergic efferent neurons. Because dopaminergic neurons in the ventral tegmental area (VTA) that project to the subcortical limbic region (including the NAc) are governed by GABAergic inhibitory projections from the ventral pallidum, activation of the NAc by the vSub suppresses tonic inhibitory GABAergic inputs from the ventral pallidum and consequently leads to tonic activation of VTA dopamine neurons.

10. Conclusion

In the course of large-scale behavioral screening of mouse models of neuropsychiatric disorders, a number of strains of mice displayed a series of schizophrenia-related behavioral abnormalities, including increased locomotor activity, severe working memory deficits, and decreased social interaction. Among these models, α-CaMKII HKO, Shn-2 KO, SNAP-25 KI, and CN KO mice all possessed an iDG phenotype. Subsequently, it was shown that chronic FLX treatment or pilocarpine-induced SRS reverses neuronal maturation, resulting in the iDG phenotype in wild-type mice. Importantly, iDG-like phenomena have been observed in the brains of patients with schizophrenia and bipolar disorder when examined postmortem. Based on these observations, iDG is proposed as a potential endophenotype shared by certain types of neuropsychiatric disorders.

Due to limitations of the current psychiatric diagnostic methods, schizophrenia, bipolar disorder, and other related psychiatric disorders are considered biologically heterogeneous populations. Therefore, an endophenotype-based analysis would be preferable for establishing biological characteristics for the classification of psychiatric disorders, rather than an analysis based on current diagnostic methods [163]. In this context, the current findings suggest that the iDG phenotype could be used as a potential brain endophenotype shared by neuropsychiatric disorders (including schizophrenia, bipolar disorders, and epilepsy) and could be useful for phenotype-based classification of these disorders. Furthermore, at least partial rescue of iDG phenotypes and some behavioral abnormalities was possible in Shn-2 KO and SNAP-25 KI mice with treatment based on possible mechanisms underlying the iDG phenotype. Further investigation of iDG as an important factor in the precipitation of, as well as recovery from, episodes of schizophrenia and other psychiatric disorders would facilitate the study of these disorders.

Disclosure

Noah M. Walton and Mitsuyuki Matsumoto are employees of the Astellas Research Institute of America LLC, a subsidiary of Astellas Pharma Inc. The other authors declare no conflict of interests.

Acknowledgment

This work was supported by KAKENHI (24111546, 20023017, 18023022, and 17025023) from the Ministry of Education, Culture, Sports, Science and Technology (MEXT) of Japan, Promotion of Fundamental Studies in Health Sciences of the National Institute of Biomedical Innovation (NIBIO), Neuroinformatics Japan Center (NIJC), and by grants from CREST & BIRD of the Japan Science and Technology Agency (JST). The authors gratefully acknowledge Hisatsugu Koshimizu in Fujita Health University for his helpful comments on the paper.

References

[1] N. Yamasaki, M. Maekawa, K. Kobayashi et al., "Alpha-CaMKII deficiency causes immature dentate gyrus, a novel candidate endophenotype of psychiatric disorders," *Molecular Brain*, vol. 1, article 6, 2008.

[2] K. Takao, K. Kobayashi, H. Hagihara et al., "Deficiency of Schnurri-2, an MHC enhancer binding protein, induces mild chronic inflammation in the brain and confers molecular, neuronal, and behavioral phenotypes related to schizophrenia," *Neuropsychopharmacology*, 2013.

[3] K. Ohira, K. Kobayashi, K. Toyama et al., "Synaptosomal-associated protein 25 mutation converts dentate granule cells to an immature state in adult mice," *Molecular Brain*, vol. 6, article 12, 2013.

[4] H. Hagihara, H. K. Nakamura, K. Toyama, I. A. Graef, G. R. Crabtree, and T. Miyakawa, "Forebrain-specific calcineurin deficiency causes immaturity of the dentate granule cells in adult mice," SfN meeting 2011 abstract.

[5] K. Kobayashi, Y. Ikeda, A. Sakai et al., "Reversal of hippocampal neuronal maturation by serotonergic antidepressants," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 107, no. 18, pp. 8434–8439, 2010.

[6] R. Shin, K. Kobayashi, H. Hagihara et al., "The immature dentate gyrus represents a common endophenotype of psychiatric disorders and epilepsy," *Bipolar Disorders*, 2013.

[7] N. M. Walton, Y. Zhou, J. H. Kogan et al., "Detection of an immature dentate gyrus feature in human schizophrenia/bipolar patients," *Translational Psychiatry*, vol. 2, no. 7, article e135, 2012.

[8] K. Takao, N. Yamasaki, and T. Miyakawa, "Impact of brain-behavior phenotypying of genetically-engineered mice on research of neuropsychiatric disorders," *Neuroscience Research*, vol. 58, no. 2, pp. 124–132, 2007.

[9] D. J. Gerber, D. Hall, T. Miyakawa et al., "Evidence for association of schizophrenia with genetic variation in the 8p21.3 gene, PPP3CC, encoding the calcineurin gamma subunit," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 100, no. 15, pp. 8993–8998, 2003.

[10] T. Miyakawa, L. M. Leiter, D. J. Gerber et al., "Conditional calcineurin knockout mice exhibit multiple abnormal behaviors related to schizophrenia," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 100, no. 15, pp. 8987–8992, 2003.

[11] D. G. Winder and J. D. Sweatt, "Roles of serine/threonine phosphatases in hippocampal synaptic plasticity," *Nature Reviews Neuroscience*, vol. 2, no. 7, pp. 461–474, 2001.

[12] H. Shoji, H. Hagihara, K. Takao, S. Hattori, and T. Miyakawa, "T-maze forced alternation and left-right discrimination tasks for assessing working and reference memory in mice," *Journal of Visualized Experiments*, no. 60, article e3300, 2012.

[13] N. Matsuo, N. Yamasaki, K. Ohira et al., "Neural activity changes underlying the working memory deficit in alpha-CaMKII heterozygous knockout mice," *Frontiers in Behavioral Neuroscience*, vol. 3, article 20, 2009.

[14] C. E. Young, K. Arima, J. Xie et al., "SNAP-25 deficit and hippocampal connectivity in schizophrenia," *Cerebral Cortex*, vol. 8, no. 3, pp. 261–268, 1998.

[15] C. M. Lewis, D. F. Levinson, L. H. Wise et al., "Genome scan meta-analysis of schizophrenia and bipolar disorder, part II: Schizophrenia," *American Journal of Human Genetics*, vol. 73, no. 1, pp. 34–48, 2003.

[16] C. L. Barr, Y. Feng, K. Wigg et al., "Identification of DNA variants in the SNAP-25 gene and linkage study of these polymorphisms and attention-deficit hyperactivity disorder," *Molecular Psychiatry*, vol. 5, no. 4, pp. 405–409, 2000.

[17] T. K. Choi, H. S. Lee, J. W. Kim et al., "Support for the MnlI polymorphism of SNAP25; a Korean ADHD case-control study," *Molecular Psychiatry*, vol. 12, no. 3, pp. 224–226, 2007.

[18] Y. Zhang, A. P. Vilaythong, D. Yoshor, and J. L. Noebels, "Elevated thalamic low-voltage-activated currents precede the onset of absence epilepsy in the SNAP25-deficient mouse mutant Coloboma," *The Journal of Neuroscience*, vol. 24, no. 22, pp. 5239–5248, 2004.

[19] M. Kataoka, R. Kuwahara, R. Matsuo, M. Sekiguchi, K. Inokuchi, and M. Takahashi, "Development- and activity-dependent regulation of SNAP-25 phosphorylation in rat brain," *Neuroscience Letters*, vol. 407, no. 3, pp. 258–262, 2006.

[20] Y. Horiuchi, H. Ishiguro, M. Koga et al., "Support for association of the PPP3CC gene with schizophrenia," *Molecular Psychiatry*, vol. 12, no. 10, pp. 891–893, 2007.

[21] H. Zeng, S. Chattarji, M. Barbarosie et al., "Forebrain-specific calcineurin knockout selectively impairs bidirectional synaptic plasticity and working/episodic-like memory," *Cell*, vol. 107, no. 5, pp. 617–629, 2001.

[22] H. Hagihara, K. Toyama, N. Yamasaki, and T. Miyakawa, "Dissection of hippocampal dentate gyrus from adult mouse," *Journal of Visualized Experiments*, no. 33, 2009.

[23] L. M. Valor, D. Jancic, R. Lujan, and A. Barco, "Ultrastructural and transcriptional profiling of neuropathological misregulation of CREB function," *Cell Death & Differentiation*, vol. 17, no. 10, pp. 1636–1644, 2010.

[24] K. Merz, S. Herold, and D. C. Lie, "CREB in adult neurogenesis—master and partner in the development of adult-born neurons?" *The European Journal of Neuroscience*, vol. 33, no. 6, pp. 1078–1086, 2011.

[25] K. Ohira and T. Miyakawa, "Chronic treatment with fluoxetine for more than 6 weeks decreases neurogenesis in the subventricular zone of adult mice," *Molecular Brain*, vol. 4, no. 1, article 10, 2011.

[26] S. C. Dulawa, K. A. Holick, B. Gundersen, and R. Hen, "Effects of chronic fluoxetine in animal models of anxiety and depression," *Neuropsychopharmacology*, vol. 29, no. 7, pp. 1321–1330, 2004.

[27] R. Alonso, G. Griebel, G. Pavone, J. Stemmelin, G. Le Fur, and P. Soubrié, "Blockade of CRF1 or V1B receptors reverses stress-induced suppression of neurogenesis in a mouse model of depression," *Molecular Psychiatry*, vol. 9, no. 3, 224 pages, 2004.

[28] D. J. David, B. A. Samuels, Q. Rainer et al., "Neurogenesis-dependent and -independent effects of fluoxetine in an animal model of anxiety/depression," *Neuron*, vol. 62, no. 4, pp. 479–493, 2009.

[29] K. Kobayashi, Y. Ikeda, and H. Suzuki, "Behavioral destabilization induced by the selective serotonin reuptake inhibitor fluoxetine," *Molecular Brain*, vol. 4, no. 1, article 12, 2011.

[30] J. Detour, H. Schroeder, D. Desor, and A. Nehlig, "A 5-month period of epilepsy impairs spatial memory, decreases anxiety, but spares object recognition in the lithium-pilocarpine model in adult rats," *Epilepsia*, vol. 46, no. 4, pp. 499–508, 2005.

[31] I. Sillaber, M. Panhuysen, M. S. H. Henniger et al., "Profiling of behavioral changes and hippocampal gene expression in mice chronically treated with the SSRI paroxetine," *Psychopharmacology*, vol. 200, no. 4, pp. 557–572, 2008.

[32] H. Hagihara, K. Ohira, K. Toyama, and T. Miyakawa, "Expression of the AMPA receptor subunits GluR1 and GluR2 is associated with granule cell maturation in the dentate gyrus," *Frontiers in Neurogenesis*, vol. 5, article 100, 2011.

[33] H. Hagihara, M. Hara, K. Tsunekawa, Y. Nakagawa, M. Sawada, and K. Nakano, "Tonic-clonic seizures induce division of neuronal progenitor cells with concomitant changes in expression of neurotrophic factors in the brain of pilocarpine-treated mice," *Molecular Brain Research*, vol. 139, no. 2, pp. 258–266, 2005.

[34] O. K. Okamoto, L. Janjoppi, F. M. Bonone et al., "Whole transcriptome analysis of the hippocampus: toward a molecular portrait of epileptogenesis," *BMC Genomics*, vol. 11, no. 1, article 230, 2010.

[35] B. H. Cha, C. Akman, D. C. Silveira, X. Liu, and G. L. Holmes, "Spontaneous recurrent seizure following status epilepticus enhances dentate gyrus neurogenesis," *Brain and Development*, vol. 26, no. 6, pp. 394–397, 2004.

[36] N. N. Karpova, A. Pickenhagen, J. Lindholm et al., "Fear erasure in mice requires synergy between antidepressant drugs and extinction training," *Science*, vol. 334, no. 6063, pp. 1731–1734, 2011.

[37] T. K. Hensch, "Critical period plasticity in local cortical circuits," *Nature Reviews Neuroscience*, vol. 6, no. 11, pp. 877–888, 2005.

[38] S. Berretta, "Extracellular matrix abnormalities in schizophrenia," *Neuropharmacology*, vol. 62, no. 3, pp. 1584–1597, 2012.

[39] 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.

[40] N. Gogolla, P. Caroni, A. Lüthi, and C. Herry, "Perineuronal nets protect fear memories from erasure," *Science*, vol. 325, no. 5945, pp. 1258–1261, 2009.

[41] 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.

[42] E. S. Lein, M. J. Hawrylycz, N. Ao et al., "Genome-wide atlas of gene expression in the adult mouse brain," *Nature*, vol. 445, no. 7124, pp. 168–176, 2007.

[43] C. A. Altar, L. W. Jurata, V. Charles et al., "Deficient hippocampal neuron expression of proteasome, ubiquitin, and mitochondrial genes in multiple schizophrenia cohorts," *Biological Psychiatry*, vol. 58, no. 2, pp. 85–96, 2005.

[44] T. M. Hyde, B. K. Lipska, T. Ali et al., "Expression of GABA signaling molecules KCC2, NKCC1, and GAD1 in cortical development and schizophrenia," *The Journal of Neuroscience*, vol. 31, no. 30, pp. 11088–11095, 2011.

[45] P. Blaesse, M. S. Airaksinen, C. Rivera, and K. Kaila, "Cation-chloride cotransporters and neuronal function," *Neuron*, vol. 61, no. 6, pp. 820–838, 2009.

[46] D. A. Lewis, D. A. Cruz, D. S. Melchitzky, and J. N. Pierri, "Lamina-specific deficits in parvalbumin-immunoreactive varicosities in the prefrontal cortex of subjects with schizophrenia: evidence for fewer projections from the thalamus," *The American Journal of Psychiatry*, vol. 158, no. 9, pp. 1411–1422, 2001.

[47] G. P. Reynolds, Z. J. Zhang, and C. L. Beasley, "Neurochemical correlates of cortical GABAergic deficits in schizophrenia: selective losses of calcium binding protein immunoreactivity," *Brain Research Bulletin*, vol. 55, no. 5, pp. 579–584, 2001.

[48] C. L. Beasley, Z. J. Zhang, I. Patten, and G. P. Reynolds, "Selective deficits in prefrontal cortical GABAergic neurons in schizophrenia defined by the presence of calcium-binding proteins," *Biological Psychiatry*, vol. 52, no. 7, pp. 708–715, 2002.

[49] C. L. Beasley and G. P. Reynolds, "Parvalbumin-immunoreactive neurons are reduced in the prefrontal cortex of schizophrenics," *Schizophrenia Research*, vol. 24, no. 3, pp. 349–355, 1997.

[50] A. Y. Wang, K. M. Lohmann, C. K. Yang et al., "Bipolar disorder type 1 and schizophrenia are accompanied by decreased density of parvalbumin- and somatostatin-positive interneurons in the parahippocampal region," *Acta Neuropathologica*, vol. 122, no. 5, pp. 615–626, 2011.

[51] B. W. Okaty, M. N. Miller, K. Sugino, C. M. Hempel, and S. B. Nelson, "Transcriptional and electrophysiological maturation of neocortical fast-spiking GABAergic interneurons," *The Journal of Neuroscience*, vol. 29, no. 21, pp. 7040–7052, 2009.

[52] M. J. Gandal, A. M. Nesbitt, R. M. McCurdy, and M. D. Alter, "Measuring the maturity of the fast-spiking interneuron transcriptional program in autism, schizophrenia, and bipolar disorder," *PLoS ONE*, vol. 7, no. 8, Article ID e41215, 2012.

[53] H. Pantazopoulos, T. U. W. Woo, M. P. Lim, N. Lange, and S. Berretta, "Extracellular matrix-glial abnormalities in the amygdala and entorhinal cortex of subjects diagnosed with schizophrenia," *Archives of General Psychiatry*, vol. 67, no. 2, pp. 155–166, 2010.

[54] H. Pantazopoulos, N. Lange, R. J. Baldessarini, and S. Berretta, "Parvalbumin neurons in the entorhinal cortex of subjects diagnosed with bipolar disorder or schizophrenia," *Biological Psychiatry*, vol. 61, no. 5, pp. 640–652, 2007.

[55] S. Fukuda, Y. Yamasaki, T. Iwaki et al., "Characterization of the biological functions of a transcription factor, c-myc intron binding protein 1 (MIBP1)," *Journal of Biochemistry*, vol. 131, no. 3, pp. 349–357, 2002.

[56] S. M. Purcell, N. R. Wray, J. L. Stone et al., "Common polygenic variation contributes to risk of schizophrenia and bipolar disorder," *Nature*, vol. 460, no. 7256, pp. 748–752, 2009.

[57] J. Shi, D. F. Levinson, J. Duan et al., "Common variants on chromosome 6p22. 1 are associated with schizophrenia," *Nature*, vol. 460, no. 7256, pp. 753–757, 2009.

[58] Y. Shi, Z. Li, Q. Xu et al., "Common variants on 8p12 and 1q24. 2 confer risk of schizophrenia," *Nature Genetics*, vol. 43, no. 12, pp. 1224–1227, 2011.

[59] H. Stefansson, R. A. Ophoff, S. Steinberg et al., "Common variants conferring risk of schizophrenia," *Nature*, vol. 460, no. 7256, pp. 744–747, 2009.

[60] W. H. Yue, H.-F. Wang, L. D. Sun et al., "Genome-wide association study identifies a susceptibility locus for schizophrenia in Han Chinese at 11p11. 2," *Nature Genetics*, vol. 43, no. 12, pp. 1228–1231, 2011.

[61] P. J. van den Elsen, S. J. P. Gobin, M. C. J. A. van Eggermond, and A. Peijnenburg, "Regulation of MHC class I and II gene transcription: differences and similarities," *Immunogenetics*, vol. 48, no. 3, pp. 208–221, 1998.

[62] M. Y. Kimura, H. Hosokawa, M. Yamashita et al., "Regulation of T helper type 2 cell differentiation by murine Schnurri-2," *Journal of Experimental Medicine*, vol. 201, no. 3, pp. 397–408, 2005.

[63] M. Y. Kimura, C. Iwamura, A. Suzuki et al., "Schnurri-2 controls memory Th1 and Th2 cell numbers in vivo," *Journal of Immunology*, vol. 178, no. 8, pp. 4926–4936, 2007.

[64] N. Muller and M. Schwarz, "Schizophrenia as an inflammation-mediated dysbalance of glutamatergic neurotransmission," *Neurotoxicity Research*, vol. 10, no. 2, pp. 131–148, 2006.

[65] B. H. Miller, L. E. Schultz, A. Gulati, M. D. Cameron, and M. T. Pletcher, "Genetic regulation of behavioral and neuronal responses to fluoxetine," *Neuropsychopharmacology*, vol. 33, no. 6, pp. 1312–1322, 2008.

[66] M. Kataoka, S. Yamamori, E. Suzuki et al., "A single amino acid mutation in SNAP-25 induces anxiety-related behavior in mouse," *PLoS ONE*, vol. 6, no. 9, Article ID e25158, 2011.

[67] S. Otsuka, S. Yamamori, S. Watanabe et al., "PKC-dependent phosphorylation of SNAP-25 plays a crucial role in the suppression of epileptogenesis and anxiety-related behavior in postnatal period of mouse," *Neuroscience Research*, vol. 71, supplement, article e296, 2011.

[68] A. Vezzani, "Inflammation and epilepsy," *Epilepsy Currents*, vol. 5, no. 1, pp. 1–6, 2005.

[69] A. Vezzani and T. Granata, "Brain inflammation in epilepsy: experimental and clinical evidence," *Epilepsia*, vol. 46, no. 11, pp. 1724–1743, 2005.

[70] O. Tomkins, O. Friedman, S. Ivens et al., "Blood-brain barrier disruption results in delayed functional and structural alterations in the rat neocortex," *Neurobiology of Disease*, vol. 25, no. 2, pp. 367–377, 2007.

[71] E. A. van Vliet, S. da C. Araújo, S. Redeker, R. Van Schaik, E. Aronica, and J. A. Gorter, "Blood-brain barrier leakage may lead to progression of temporal lobe epilepsy," *Brain*, vol. 130, no. 2, pp. 521–534, 2007.

[72] L. Uva, L. Librizzi, N. Marchi et al., "Acute induction of epileptiform discharges by pilocarpine in the in vitro isolated guinea-pig brain requires enhancement of blood-brain barrier permeability," *Neuroscience*, vol. 151, no. 1, pp. 303–312, 2008.

[73] P. F. Fabene, G. N. Mora, M. Martinello et al., "A role for leukocyte-endothelial adhesion mechanisms in epilepsy," *Nature Medicine*, vol. 14, no. 12, pp. 1377–1383, 2008.

[74] M. Zhou, W. Li, S. Huang et al., "mTOR inhibition ameliorates cognitive and affective deficits caused by Disc1 knockdown in adult-born dentate granule neurons," *Neuron*, vol. 77, no. 4, pp. 647–654, 2013.

[75] D. St Clair, D. Blackwood, W. Muir et al., "Association within a family of a balanced autosomal translocation with major mental illness," *The Lancet*, vol. 336, no. 8706, pp. 13–16, 1990.

[76] N. Burnashev, H. Monyer, P. H. Seeburg, and B. Sakmann, "Divalent ion permeability of AMPA receptor channels is dominated by the edited form of a single subunit," *Neuron*, vol. 8, no. 1, pp. 189–198, 1992.

[77] M. Hollmann, M. Hartley, and S. Heinemann, "Ca2+ permeability of KA-AMPA-gated glutamate receptor channels depends on subunit composition," *Science*, vol. 252, no. 5007, pp. 851–853, 1991.

[78] R. I. Hume, R. Dingledine, and S. F. Heinemann, "Identification of a site in glutamate receptor subunits that controls calcium permeability," *Science*, vol. 253, no. 5023, pp. 1028–1031, 1991.

[79] C. Zhao, W. Deng, and F. H. Gage, "Mechanisms and functional implications of adult neurogenesis," *Cell*, vol. 132, no. 4, pp. 645–660, 2008.

[80] S. Jessberger and G. Kempermann, "Adult-born hippocampal neurons mature into activity-dependent responsiveness," *The European Journal of Neuroscience*, vol. 18, no. 10, pp. 2707–2712, 2003.

[81] P. S. Goldman-Rakic, "Working memory dysfunction in schizophrenia," *Journal of Neuropsychiatry and Clinical Neurosciences*, vol. 6, no. 4, pp. 348–357, 1994.

[82] B. Elvevåg and T. E. Goldberg, "Cognitive impairment in schizophrenia is the core of the disorder," *Critical Reviews in Neurobiology*, vol. 14, no. 1, pp. 1–21, 2000.

[83] D. L. Braff and M. A. Geyer, "Sensorimotor gating and schizophrenia: human and animal model studies," *Archives of General Psychiatry*, vol. 47, no. 2, pp. 181–188, 1990.

[84] American Psychiatric Association, *Diagnostic and Statistical Manual of Mental Disorders*, American Psychiatric Association, Washington, DC, USA, 4th edition, 1994.

[85] R. R. Gainetdinov, A. R. Mohn, L. M. Bohn, and M. G. Caron, "Glutamatergic modulation of hyperactivity in mice lacking the dopamine transporter," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 98, no. 20, pp. 11047–11054, 2001.

[86] P. R. Maycox, F. Kelly, A. Taylor et al., "Analysis of gene expression in two large schizophrenia cohorts identifies multiple changes associated with nerve terminal function," *Molecular Psychiatry*, vol. 14, no. 12, pp. 1083–1094, 2009.

[87] Z. J. Zhang and G. P. Reynolds, "A selective decrease in the relative density of parvalbumin-immunoreactive neurons in the hippocampus in schizophrenia," *Schizophrenia Research*, vol. 55, no. 1-2, pp. 1–10, 2002.

[88] M. B. Knable, B. M. Barci, M. J. Webster, J. Meador-Woodruff, and E. F. Torrey, "Molecular abnormalities of the hippocampus in severe psychiatric illness: postmortem findings from the Stanley Neuropathology Consortium," *Molecular Psychiatry*, vol. 9, no. 6, pp. 609–620, 2004.

[89] F. M. Benes, B. Lim, D. Matzilevich, J. P. Walsh, S. Subburaju, and M. Minns, "Regulation of the GABA cell phenotype in hippocampus of schizophrenics and bipolars," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 104, no. 24, pp. 10164–10169, 2007.

[90] J. N. Pierri, A. S. Chaudry, T. U. W. Woo, and D. A. Lewis, "Alterations in chandelier neuron axon terminals in the prefrontal cortex of schizophrenic subjects," *The American Journal of Psychiatry*, vol. 156, no. 11, pp. 1709–1719, 1999.

[91] J. Gallinat, G. Winterer, C. S. Herrmann, and D. Senkowski, "Reduced oscillatory gamma-band responses in unmedicated schizophrenic patients indicate impaired frontal network processing," *Clinical Neurophysiology*, vol. 115, no. 8, pp. 1863–1874, 2004.

[92] L. V. Moran and L. E. Hong, "High vs low frequency neural oscillations in schizophrenia," *Schizophrenia Bulletin*, vol. 37, no. 4, pp. 659–663, 2011.

[93] S. R. Sponheim, B. A. Clementz, W. G. Iacono, and M. Beiser, "Resting EEG in first-episode and chronic schizophrenia," *Psychophysiology*, vol. 31, no. 1, pp. 37–43, 1994.

[94] A. Abi-Dargham, "Do we still believe in the dopamine hypothesis? New data bring new evidence," *The International Journal of Neuropsychopharmacology*, vol. 7, supplement 1, pp. S1–S5, 2004.

[95] G. Winterer and D. R. Weinberger, "Genes, dopamine and cortical signal-to-noise ratio in schizophrenia," *Trends in Neuroscience*, vol. 27, no. 11, pp. 683–690, 2004.

[96] D. A. Cousins, K. Butts, and A. H. Young, "The role of dopamine in bipolar disorder," *Bipolar Disorders*, vol. 11, no. 8, pp. 787–806, 2009.

[97] G. Novak and P. Seeman, "Hyperactive mice show elevated D2High receptors, a model for schizophrenia: calcium/calmodulin-dependent kinase II alpha knockouts," *Synapse*, vol. 64, no. 10, pp. 794–800, 2010.

[98] C. R. Maxwell, S. J. Kanes, T. Abel, and S. J. Siegel, "Phosphodiesterase inhibitors: a novel mechanism for receptor-independent antipsychotic medications," *Neuroscience*, vol. 129, no. 1, pp. 101–107, 2004.

[99] S. J. Kanes, J. Tokarczyk, S. J. Siegel, W. Bilker, T. Abel, and M. P. Kelly, "Rolipram: a specific phosphodiesterase 4 inhibitor with potential antipsychotic activity," *Neuroscience*, vol. 144, no. 1, pp. 239–246, 2007.

[100] J. Lisman, H. Schulman, and H. Cline, "The molecular basis of CaMKII function in synaptic and behavioural memory," *Nature Reviews Neuroscience*, vol. 3, no. 3, pp. 175–190, 2002.

[101] D. R. Weinberger, "Implications of normal brain development for the pathogenesis of schizophrenia," *Archives of General Psychiatry*, vol. 44, no. 7, pp. 660–669, 1987.

[102] D. R. Weinberger and R. K. McClure, "Neurotoxicity, neuroplasticity, and magnetic resonance imaging morphometry: what is happening in the schizophrenic brain?" *Archives of General Psychiatry*, vol. 59, no. 6, pp. 553–558, 2002.

[103] S. Marenco and D. R. Weinberger, "The neurodevelopmental hypothesis of schizophrenia: following a trail of evidence from cradle to grave," *Development and Psychopathology*, vol. 12, no. 3, pp. 501–527, 2000.

[104] T. D. Cannon, I. M. Rosso, C. E. Bearden, L. E. Sanchez, and T. Hadley, "A prospective cohort study of neurodevelopmental processes in the genesis and epigenesis of schizophrenia," *Development and Psychopathology*, vol. 11, no. 3, pp. 467–485, 1999.

[105] B. K. Lipska, "Using animal models to test a neurodevelopmental hypothesis of schizophrenia," *Journal of Psychiatry and Neuroscience*, vol. 29, no. 4, pp. 282–286, 2004.

[106] C. T. Ekdahl, J.-H. Claasen, S. Bonde, Z. Kokaia, and O. Lindvall, "Inflammation is detrimental for neurogenesis in adult brain," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 100, no. 23, pp. 13632–13637, 2003.

[107] S. Das and A. Basu, "Inflammation: a new candidate in modulating adult neurogenesis," *Journal of Neuroscience Research*, vol. 86, no. 6, pp. 1199–1208, 2008.

[108] R. L. Hunter, N. Dragicevic, K. Seifert et al., "Inflammation induces mitochondrial dysfunction and dopaminergic neurodegeneration in the nigrostriatal system," *Journal of Neurochemistry*, vol. 100, no. 5, pp. 1375–1386, 2007.

[109] M. T. Fischer, R. Sharma, J. L. Lim et al., "NADPH oxidase expression in active multiple sclerosis lesions in relation to oxidative tissue damage and mitochondrial injury," *Brain*, vol. 135, no. 3, pp. 886–899, 2012.

[110] Y. Pang, Z. Cai, and P. G. Rhodes, "Disturbance of oligodendrocyte development, hypomyelination and white matter injury in the neonatal rat brain after intracerebral injection of lipopolysaccharide," *Developmental Brain Research*, vol. 140, no. 2, pp. 205–214, 2003.

[111] W. Bruck, R. Pfortner, T. Pham et al., "Reduced astrocytic NF-κB activation by laquinimod protects from cuprizone-induced demyelination," *Acta Neuropathologica*, vol. 124, no. 3, pp. 411–424, 2012.

[112] A. Reif, S. Fritzen, M. Finger et al., "Neural stem cell proliferation is decreased in schizophrenia, but not in depression," *Molecular Psychiatry*, vol. 11, no. 5, pp. 514–522, 2006.

[113] M. S. Keshavan, H. A. Nasrallah, and R. Tandon, "Schizophrenia, "Just the Facts" 6. Moving ahead with the schizophrenia concept: from the elephant to the mouse," *Schizophrenia Research*, vol. 127, no. 1–3, pp. 3–13, 2011.

[114] H. Nawa and N. Takei, "Recent progress in animal modeling of immune inflammatory processes in schizophrenia: implication of specific cytokines," *Neuroscience Research*, vol. 56, no. 1, pp. 2–13, 2006.

[115] P. H. Patterson, "Immune involvement in schizophrenia and autism: etiology, pathology and animal models," *Behavioural Brain Research*, vol. 204, no. 2, pp. 313–321, 2009.

[116] A. S. Brown and P. H. Patterson, "Maternal infection and schizophrenia: implications for prevention," *Schizophrenia Bulletin*, vol. 37, no. 2, pp. 284–290, 2011.

[117] E. Y. Hsiao, S. W. McBride, J. Chow, S. K. Mazmanian, and P. H. Patterson, "Modeling an autism risk factor in mice leads to permanent immune dysregulation," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 109, no. 31, pp. 12776–12781, 2012.

[118] P. H. Patterson, "Maternal infection: window on neuroimmune interactions in fetal brain development and mental illness," *Current Opinion in Neurobiology*, vol. 12, no. 1, pp. 115–118, 2002.

[119] L. Shi, S. H. Fatemi, R. W. Sidwell, and P. H. Patterson, "Maternal influenza infection causes marked behavioral and pharmacological changes in the offspring," *The Journal of Neuroscience*, vol. 23, no. 1, pp. 297–302, 2003.

[120] J. P. A. Ioannidis, E. E. Ntzani, T. A. Trikalinos, and D. G. Contopoulos-Ioannidis, "Replication validity of genetic association studies," *Nature Genetics*, vol. 29, no. 3, pp. 306–309, 2001.

[121] M. Ayalew, H. Le-Niculescu, D. F. Levey et al., "Convergent functional genomics of schizophrenia: from comprehensive understanding to genetic risk prediction," *Molecular Psychiatry*, vol. 17, no. 9, pp. 887–905, 2012.

[122] K. V. Chowdari, K. Mirnics, P. Semwal et al., "Association and linkage analyses of RGS4 polymorphisms in schizophrenia," *Human Molecular Genetics*, vol. 11, no. 12, pp. 1373–1380, 2002.

[123] K. Mirnics, F. A. Middleton, G. D. Stanwood, D. A. Lewis, and P. Levitt, "Disease-specific changes in regulator of G-protein signaling 4 (RGS4) expression in schizophrenia," *Molecular Psychiatry*, vol. 6, no. 3, pp. 293–301, 2001.

[124] T. L. Petryshen, F. A. Middleton, A. R. Tahl et al., "Genetic investigation of chromosome 5q GABAA receptor subunit genes in schizophrenia," *Molecular Psychiatry*, vol. 10, no. 12, pp. 1074–1088, 2005.

[125] N. C. Allen, S. Bagade, M. B. McQueen et al., "Systematic meta-analyses and field synopsis of genetic association studies in schizophrenia: the SzGene database," *Nature Genetics*, vol. 40, no. 7, pp. 827–834, 2008.

[126] S. J. Huffaker, J. Chen, K. K. Nicodemus et al., "A primate-specific, brain isoform of KCNH2 affects cortical physiology, cognition, neuronal repolarization and risk of schizophrenia," *Nature Medicine*, vol. 15, no. 5, pp. 509–518, 2009.

[127] R. Zakharyan, A. Khoyetsyan, A. Arakelyan et al., "Association of C1QB gene polymorphism with schizophrenia in Armenian population," *BMC Medical Genetics*, vol. 12, article 126, 2011.

[128] J. Ekelund, D. Lichtermann, I. Hovatta et al., "Genome-wide scan for schizophrenia in the Finnish population: evidence for a locus on chromosome 7q22," *Human Molecular Genetics*, vol. 9, no. 7, pp. 1049–1057, 2000.

[129] W. Yan, X.-Y. Guan, E. D. Green et al., "Childhood-onset schizophrenia/autistic disorder and t(1;7) reciprocal translocation: identification of a BAC contig spanning the translocation breakpoint at 7q21," *American Journal of Medical Genetics*, vol. 96, no. 6, pp. 749–753, 2000.

[130] M. Bradford, M. H. Law, A. D. Stewart, D. J. Shaw, I. L. Megson, and J. Wei, "The TGM2 gene is associated with schizophrenia in a british population," *American Journal of Medical Genetics B*, vol. 150, no. 3, pp. 335–340, 2009.

[131] S. W. Flynn, D. J. Lang, A. L. Mackay et al., "Abnormalities of myelination in schizophrenia detected in vivo with MRI, and post-mortem with analysis of oligodendrocyte proteins," *Molecular Psychiatry*, vol. 8, no. 9, pp. 811–820, 2003.

[132] K. Iwamoto, M. Bundo, and T. Kato, "Altered expression of mitochondria-related genes in postmortem brains of patients with bipolar disorder or schizophrenia, as revealed by large-scale DNA microarray analysis," *Human Molecular Genetics*, vol. 14, no. 2, pp. 241–253, 2005.

[133] D. S. Olton and B. C. Papas, "Spatial memory and hippocampal function," *Neuropsychologia*, vol. 17, no. 6, pp. 669–682, 1979.

[134] P. S. Goldman-Rakic, "Regional and cellular fractionation of working memory," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 93, no. 24, pp. 13473–13480, 1996.

[135] R. L. McLamb, W. R. Mundy, and H. A. Tilson, "Intradentate colchicine disrupts the acquisition and performance of a working memory task in the radial arm maze," *NeuroToxicology*, vol. 9, no. 3, pp. 521–528, 1988.

[136] D. F. Emerich and T. J. Walsh, "Selective working memory impairments following intradentate injection of colchicine: attenuation of the behavioral but not the neuropathological effects by gangliosides GM1 and AGF2," *Physiology & Behavior*, vol. 45, no. 1, pp. 93–101, 1989.

[137] A. M. Morris, J. C. Churchwell, R. P. Kesner, and P. E. Gilbert, "Selective lesions of the dentate gyrus produce disruptions in place learning for adjacent spatial locations," *Neurobiology of Learning and Memory*, vol. 97, no. 3, pp. 326–331, 2012.

[138] R. P. Kesner, "Behavioral functions of the CA3 subregion of the hippocampus," *Learning and Memory*, vol. 14, no. 11, pp. 771–781, 2007.

[139] M. L. Shapiro and D. S. Olton, *Memory Systems 1994*, MIT Press, 1994.

[140] J. B. Aimone, J. Wiles, and F. H. Gage, "Potential role for adult neurogenesis in the encoding of time in new memories," *Nature Neuroscience*, vol. 9, no. 6, pp. 723–727, 2006.

[141] M. A. Yassa and C. E. L. Stark, "Pattern separation in the hippocampus," *Trends in Neurosciences*, vol. 34, no. 10, pp. 515–525, 2011.

[142] D. Marr, "Simple memory: a theory for archicortex," *Philosophical Transactions of the Royal Society of London B*, vol. 262, no. 841, pp. 23–81, 1971.

[143] B. L. McNaughton and R. G. M. Morris, "Hippocampal synaptic enhancement and information storage within a distributed memory system," *Trends in Neurosciences*, vol. 10, no. 10, pp. 408–415, 1987.

[144] R. C. O'Reilly and J. L. McClelland, "Hippocampal conjunctive encoding, storage, and recall: avoiding a trade-off," *Hippocampus*, vol. 4, no. 6, pp. 661–682, 1994.

[145] A. Treves, A. Tashiro, M. E. Witter, and E. I. Moser, "What is the mammalian dentate gyrus good for?" *Neuroscience*, vol. 154, no. 4, pp. 1155–1172, 2008.

[146] P. E. Gilbert, R. P. Kesner, and I. Lee, "Dissociating hippocampal subregions: a double dissociation between dentate gyrus and CA1," *Hippocampus*, vol. 11, no. 6, pp. 626–636, 2001.

[147] P. E. Gilbert and R. P. Kesner, "Localization of function within the dorsal hippocampus: the role of the CA3 subregion in paired-associate learning," *Behavioral Neuroscience*, vol. 117, no. 6, pp. 1385–1394, 2003.

[148] I. Lee and R. P. Kesner, "Different contributions of dorsal hippocampal subregios to emory acquisation and retrieval in contextual fear-conditioning," *Hippocampus*, vol. 14, no. 3, pp. 301–310, 2004.

[149] I. Lee and R. P. Kesner, "Encoding versus retrieval of spatial memory: double dissociation between the dentate gyrus and the perforant path inputs into CA3 in the dorsal hippocampus," *Hippocampus*, vol. 14, no. 1, pp. 66–76, 2004.

[150] J. K. Leutgeb, S. Leutgeb, M.-B. Moser, and E. I. Moser, "Pattern separation in the dentate gyrus and CA3 of the hippocampus," *Science*, vol. 315, no. 5814, pp. 961–966, 2007.

[151] T. J. McHugh, M. W. Jones, J. J. Quinn et al., "Dentate gyrus NMDA receptors mediate rapid pattern separation in the hippocampal network," *Science*, vol. 317, no. 5834, pp. 94–99, 2007.

[152] J. B. Aimone, W. Deng, and F. H. Gage, "Resolving new memories: a critical look at the dentate gyrus, adult neurogenesis, and pattern separation," *Neuron*, vol. 70, no. 4, pp. 589–596, 2011.

[153] A. Marín-Burgin, L. A. Mongiat, M. B. Pardi, and A. F. Schinder, "Unique processing during a period of high excitation/inhibition balance in adult-born neurons," *Science*, vol. 335, no. 6073, pp. 1238–1242, 2012.

[154] T. Nakashiba, J. D. Cushman, K. A. Pelkey et al., "Young dentate granule cells mediate pattern separation, whereas old granule cells facilitate pattern completion," *Cell*, vol. 149, no. 1, pp. 188–201, 2012.

[155] W. A. M. Swinkels, J. Kuyk, R. van Dyck, and P. Spinhoven, "Psychiatric comorbidity in epilepsy," *Epilepsy and Behavior*, vol. 7, no. 1, pp. 37–50, 2005.

[156] P. Cifelli and A. A. Grace, "Pilocarpine-induced temporal lobe epilepsy in the rat is associated with increased dopamine neuron activity," *The International Journal of Neuropsychopharmacology*, vol. 15, no. 7, pp. 957–964, 2012.

[157] D. J. Lodge and A. A. Grace, "Hippocampal dysregulation of dopamine system function and the pathophysiology of schizophrenia," *Trends in Pharmacological Sciences*, vol. 32, no. 9, pp. 507–513, 2011.

[158] C. A. Tamminga, S. Southcott, C. Sacco, A. D. Wagner, and S. Ghose, "Glutamate dysfunction in hippocampus: relevance of dentate gyrus and CA3 signaling," *Schizophrenia Bulletin*, vol. 38, no. 5, pp. 927–935, 2012.

[159] C. O. Lacefield, V. Itskov, T. Reardon, R. Hen, and J. A. Gordon, "Effects of adult-generated granule cells on coordinated network activity in the dentate gyrus," *Hippocampus*, vol. 22, no. 1, pp. 106–116, 2012.

[160] D. A. Henze, N. N. Urban, and G. Barrionuevo, "The multifarious hippocampal mossy fiber pathway: a review," *Neuroscience*, vol. 98, no. 3, pp. 407–427, 2000.

[161] J. Song, K. M. Christian, G. Ming, and H. Song, "Modification of hippocampal circuitry by adult neurogenesis," *Developmental Neurobiology*, vol. 72, no. 7, pp. 1032–1043, 2012.

[162] A. A. Grace, S. B. Floresco, Y. Goto, and D. J. Lodge, "Regulation of firing of dopaminergic neurons and control of goal-directed behaviors," *Trends in Neurosciences*, vol. 30, no. 5, pp. 220–227, 2007.

[163] I. I. Gottesman and T. D. Gould, "The endophenotype concept in psychiatry: etymology and strategic intentions," *The American Journal of Psychiatry*, vol. 160, no. 4, pp. 636–645, 2003.

Hindawi Publishing Corporation
Neural Plasticity
Volume 2013, Article ID 698528, 12 pages
http://dx.doi.org/10.1155/2013/698528

Review Article

Neurogenesis, Exercise, and Cognitive Late Effects of Pediatric Radiotherapy

Shaefali P. Rodgers,[1] Melissa Trevino,[1] Janice A. Zawaski,[2] M. Waleed Gaber,[2] and J. Leigh Leasure[1,3]

[1] *Department of Psychology, University of Houston, Houston, TX 77204, USA*
[2] *Department of Pediatrics, Texas Children's Hospital, Baylor College of Medicine, Houston, TX 77030, USA*
[3] *Department of Biology & Biochemistry, University of Houston, Houston, TX 77204, USA*

Correspondence should be addressed to J. Leigh Leasure; jlleasure@uh.edu

Received 15 November 2012; Accepted 20 January 2013

Academic Editor: Chitra D. Mandyam

Brain cancer is a common type of childhood malignancy, and radiotherapy (RT) is a mainstay of treatment. RT is effective for tumor eradication, and survival rates are high. However, RT damages the brain and disrupts ongoing developmental processes, resulting in debilitating cognitive "late" effects that may take years to fully manifest. These late effects likely derive from a long-term decrement in cell proliferation, combined with a neural environment that is hostile to plasticity, both of which are induced by RT. Long-term suppression of cell proliferation deprives the brain of the raw materials needed for optimum cognitive performance (such as new neurons in the hippocampus and new glia in frontal cortex), while chronic inflammation and dearth of trophic substances (such as growth hormone) limit neuroplastic potential in existing circuitry. Potential treatments for cognitive late effects should address both of these conditions. Exercise represents one such potential treatment, since it has the capacity to enhance cell proliferation, as well as to promote a neural milieu permissive for plasticity. Here, we review the evidence that cognitive late effects can be traced to RT-induced suppression of cell proliferation and hostile environmental conditions, as well as emerging evidence that exercise may be effective as an independent or adjuvant therapy.

1. Introduction

Brain tumors are the second most common form of childhood cancer, after acute lymphoblastic leukemia (ALL) [1]. Treatment for both brain tumors and ALL includes cranial RT. Given 5-year survival rates that approach 90% for children treated for ALL and 70% for those treated for brain tumors [2], there are currently a great many survivors of these cancers that suffer from the consequences of RT, including adverse physiological, psychological, and cognitive side effects that manifest both acutely and years later. These so called "late effects" result in lowered quality of life (QOL) [3] in survivors, for which there is at present no effective treatment.

RT for pediatric cancer has long been acknowledged as a primary cause of neurological complications and neurocognitive decline [4–8]. Childhood RT is associated with a significant decrease in IQ scores [8–14], thought to result from deficits in core processing functions impaired by RT, including processing speed [15], attention [15–18], working memory, and other executive functions [7, 19]. In addition to cognitive impairments, adult survivors of childhood RT also experience elevated rates of emotional distress, such as anxiety and/or depression [20, 21] and posttraumatic stress disorder [22]. These cognitive and emotional consequences of RT result in decreased QOL that manifests in a variety of ways. For example, adult survivors of childhood RT are less likely to obtain a college education [23, 24] or marry [5, 20] and more likely to be unemployed [24, 25].

Improving QOL for survivors necessarily involves attenuating the long-term neural consequences of RT. Ionizing radiation damages the brain directly, but in addition, it chronically suppresses cell proliferation, thereby depriving the brain of the raw materials needed for repair. Evidence

indicates that it also creates a milieu that is hostile to regenerative processes. When the brain is irradiated in childhood, there is a further consequence of RT, as suppressed cell proliferation and hostile environmental conditions disrupt ongoing developmental processes. What is needed, therefore, is a treatment that can both "jump-start" cell proliferation and foster a neural environment that is conducive to plasticity. Exercise may represent one such treatment, and its restorative potential for the post-RT brain is discussed.

2. RT Disrupts Brain Development

RT damages the brain regardless of age. However, the brains of children are still developing, and RT profoundly affects ongoing developmental processes. The potential mechanisms underlying this disruption are many, such as perturbations of vasculature [26] and suppression of cell proliferation [27–29]. Damage to the endocrine system [30, 31] has been shown to play a role, in particular, decreased expression of growth hormone (GH). GH deficiency results from the effects of a brain tumor or of therapy such as surgery, RT, or chemotherapy. Merchant et al. [32] report that the peak GH response within 12 months after the initiation of cranial RT depends on hypothalamic dose-volume effects and may be predicted on the basis of a linear model that sums the effects of the entire dose distribution. The rate of decline in the peak GH response may also be influenced by clinical factors indicating the severity of the disease and the type and location of tumor.

Disruption of brain development could also be due in part to cancer treatment effects on food intake. Treatment-induced nausea and vomiting, as well as gastrointestinal toxicity can lead to nutritional deficiency and changes in body composition [34, 35], which may be long-lasting. Indeed, survivors of childhood brain cancer are often underweight [36]. In contrast, survivors of childhood ALL are more likely to be obese, compared with age-matched controls [37]. Thus, treatment effects on hormone levels and nutritional intake, alone or in combination, are likely important contributors to altered neural development and, ultimately, cognitive impairments.

Animal models of pediatric RT enable controlled study of mechanisms that contribute to disrupted development and, ultimately, cognitive late effects. To model the effects of RT on the developing brain, we have treated postnatal day 28 (PND28) rats with whole brain irradiation (WBI), using one of 2 regimens: single dose (20 Gy) or fractionated, in which animals received 20 Gy over the course of 5 days (4 Gy/d). Either regimen results in a profound stunting of brain growth visible to the naked eye (see Figure 1), although the effect is clearly bigger with single dose treatment.

Using this model, we can probe the cellular, chemical, and structural effects of RT that contribute to decreased brain size and cognitive impairments in adulthood. To enhance translational value, we are using imaging techniques to discover RT-induced changes *in vivo* that predict future cognitive impairments before they manifest. For example, we are using magnetic resonance imaging (MRI) and diffusion tensor imaging (DTI) to assess RT-induced structural changes and ^{1}H magnetic resonance spectroscopy (MRS) to assess chemical changes following RT (see Figure 2). DTI has the added advantage of providing information on fractional anisotropy (FA), a measure of the functional integrity of white matter tracts. Our preliminary ^{1}H MRS findings showed changes in glutamate, alanine, and lactate in RT brains, compared to sham controls. In addition, FA analysis showed a significant decline in fimbria volume and mean fimbria FA value in RT brains compared to controls. These changes were observed three months prior to the detected cognitive changes shown in Figure 3, suggesting that imaging changes can be used as early markers of cognitive decline.

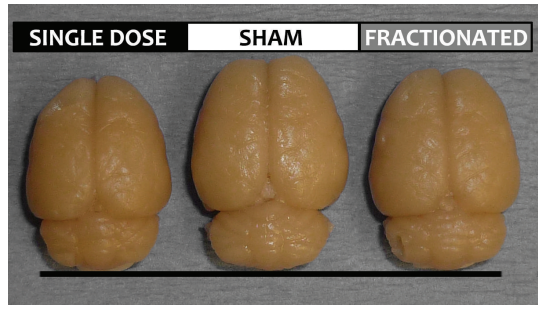


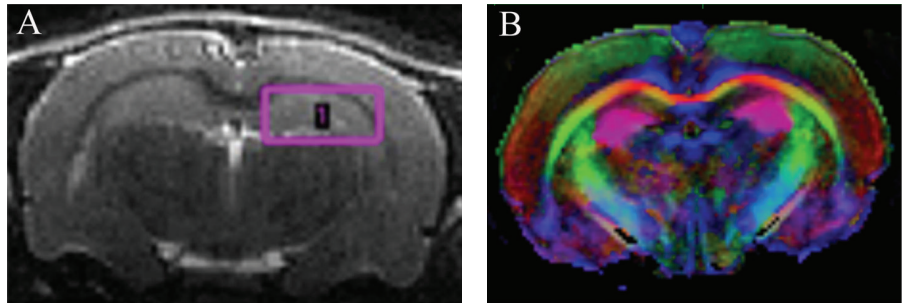
Figure 1: Irradiation of the developing (PND28) rat brain results in visibly decreased brain size in adulthood. Note that a 20 Gy total dose of X-ray radiation resulted in a smaller brain when it was administered as a single dose. A fractionated dose (4 Gy/d for 5 days) was less detrimental to brain size.

3. RT-Induced Suppression of Cell Proliferation Contributes to Cognitive Impairments

Because ionizing radiation kills dividing cells, it is effective at treating cancer, yet devastating to noncancerous tissue in the brain. Although mature neurons are postmitotic and therefore not directly affected by radiation, the brain's actively dividing neural stem cells (NSC) are largely wiped out, even by very low doses [28]. This is problematic, as it decreases the availability of new neurons in neurogenic regions of the brain and of new glia (oligodendrocytes and astrocytes) in nonneurogenic areas.

The dentate gyrus (DG) of the hippocampus, along with the lining of the lateral ventricles (the subventricular zone, or SVZ), is one of the few neurogenic regions of the adult mammalian brain (see [38] for review). Animal studies indicate that ongoing neurogenesis in this region is important for cognition. For example, newly generated neurons are kept alive by effortful learning (for review see [39]) and are needed for the formation of long-term spatial memory [40]. Analysis of postmortem human tissue following cancer treatment shows an almost complete lack of hippocampal neurogenesis [41], the functional importance of which is attested to by the cognitive impairments observed in survivors [42].

Animal models have yielded direct links between RT-induced decrements in hippocampal neurogenesis and



(a) (b)

Figure 2: Data from our group indicates that measurable imaging changes precede cognitive decline. (a) An image of a rat brain acquired using a 9.4 T MRI. The pink box indicates where ^{1}H MRS was performed. Changes in glutamate, alanine, and lactate preceded cognitive impairments. In addition, FA analysis detected a decrease in volume of the fimbria. (b) An FA map of the rat brain.

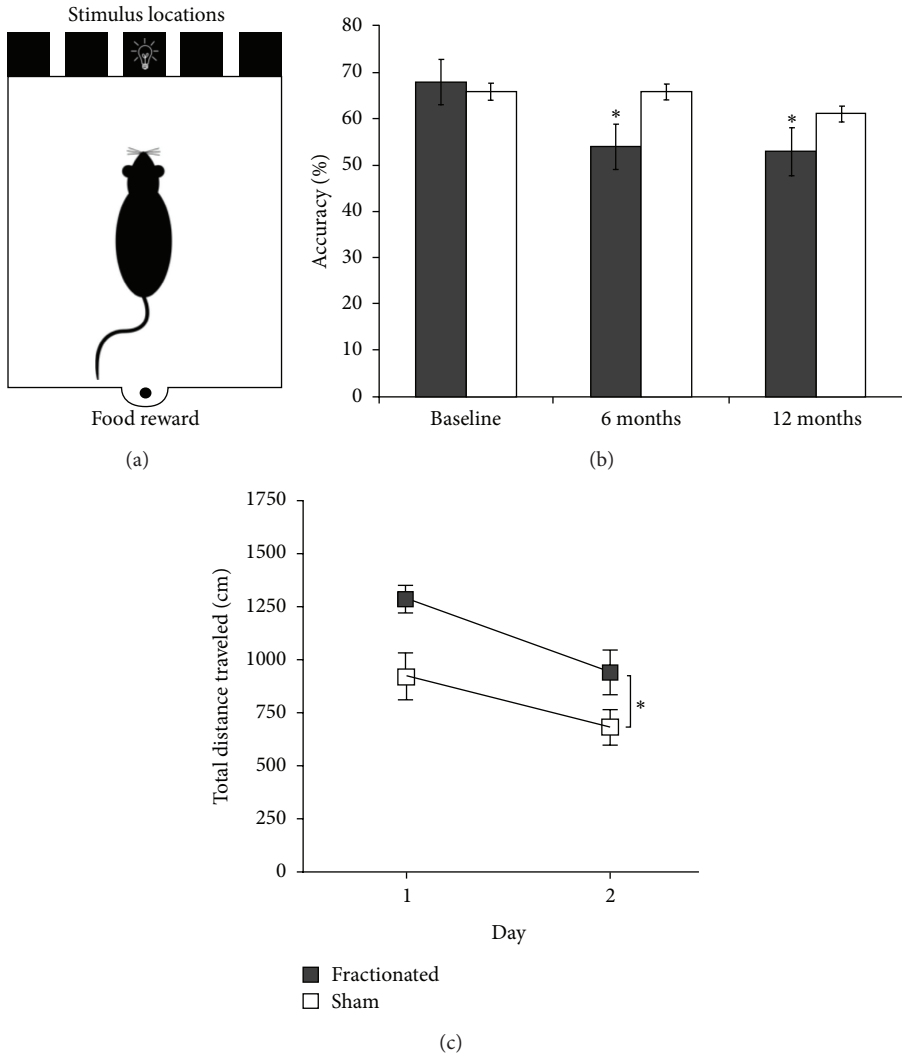


Figure 3: Data from our group showing RT-induced cognitive deficits. (a) Schematic illustration of the 5-CSRTT apparatus. (b) Fractionated X-ray radiation (4 Gy/d for 5 days) restricted to the frontal cortex of young rats significantly reduced choice accuracy on a 5-CSRTT 6 and 9 months following RT ($^{*}P < 0.05$). (c) Fractionated WBI in young rats impaired pliancy on a hippocampus-dependent strategy-switching task in the Morris water maze [33] 3-4 weeks following RT ($^{*}P < 0.05$).

cognitive impairments. Many studies have focused on deficits in spatial performance (place learning or spatial memory) and trace fear conditioning, since these are hippocampus-dependent functions. Spatial impairments have been observed in conjunction with decrements in DG cytogenesis following both fractionated [43, 44] and single-dose WBI [45, 46]. Our group also has noted performance deficits in a spatial task after fractionated irradiation (see Figure 3(c)). Decreased fear conditioning has also been associated with radiation-induced suppression of DG cytogenesis [47–49]. In sum, an increasing body of evidence implicates suppression of hippocampal neurogenesis as a causative factor in cognitive impairments following RT.

However, suppression of hippocampal neurogenesis is likely only part of the story. NSCs in nonneurogenic brain regions, such as the cortex, differentiate into glia [50]. A plentiful supply of glial cells is essential for neuronal health and function [51, 52], so reduced proliferation of NSCs due to RT could contribute to cognitive impairments by reducing the availability of glia. For example, problems with executive functions are widely reported in adult survivors of childhood RT. Executive functions develop linearly during adolescence, in apparent conjunction with myelination of the frontal lobes [19]. Frontal lobe white matter appears particularly vulnerable to RT [53], and RT-induced damage to white matter tracts may, in large part, underlie the neurocognitive deficits experienced by adult survivors of childhood cancer [19, 54]. Myelination is dependent on a ready supply of healthy oligodendrocytes, which is in turn dependent on adequate proliferative activity of NSCs. RT-induced ablation of NSCs in nonneurogenic regions could therefore contribute to cognitive impairments.

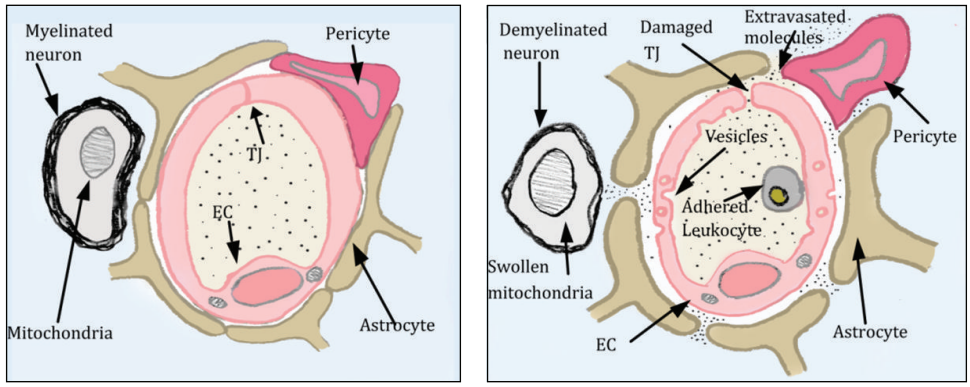
To provide direct evidence that RT-induced suppression of gliogenesis contributes to frontal lobe dysfunction, animal models of frontal lobe-dependent tasks are important. The 5-choice serial reaction time task (5-CSRTT) is a reliable means by which to assess prefrontal cognitive processes in the rodent. This automated task measures several aspects of visual attention, specifically divided, sustained, and selective attention, as well as processing speed and impulsivity [55]. The task requires the animal to detect brief flashes of light that appear in one of five apertures (see Figure 3(a)) and then nose-poke into the aperture that the light appeared in. The animal is given 5 seconds in which to make the nose-poke response. Correct responses are rewarded by a food pellet being dispensed into a magazine at the rear of the testing chamber (see Figure 3(a)). To provide motivation, animals are food restricted. Between stimulus presentations, there is an intertrial interval (ITI), and the animal must inhibit responding during this interval, because premature responses result in a short time-out period during which there are no trials, and thus food reward cannot be obtained. In performing this task, the animal has to sustain attention to all 5 of the apertures in order to constantly monitor where the light stimulus will be presented. Incorrect responses (nose-pokes into an aperture other than that in which the light was presented) indicate impaired attention. Measures of impulsivity are collected through responses that are characterized as perseverative and/or premature. Perseverative responses are defined as continuous nose-pokes in additional apertures. Nose-pokes made before the light is presented are considered premature responses. Processing speed measures are based on various latency times that are collected throughout the task.

We have used the 5-CSRTT to probe for impairment of prefrontal cognitive processes following fractionated irradiation (4 Gy/d for 5 days). Irradiated animals and shams were trained to perform the 5-CSRTT and then tested 6 and 9 months postirradiation. Our preliminary findings indicate that the irradiated animals are significantly less accurate at nose-poking into the correct aperture, suggesting that they have attentional impairments (see Figure 3(b)). Future experiments will focus on replicating these impairments in irradiated animals and determine whether they are linked to reduced gliogenesis in frontal regions.

4. RT Creates a Brain Milieu Hostile to Plasticity

The microenvironment of the brain is regulated and protected by specific barriers, which include the vascular endothelial barrier (also called the blood-brain barrier, or BBB) at the capillary-parenchyma interface and the epithelial barrier (blood-cerebrospinal fluid barrier) at the choroid plexus [56]. The BBB is more than a physical barrier: it plays a fundamental role in regulating the movement of substances between the blood and the CNS (see Figure 4(a)). The microvascular network is also the site of the BBB, and the endothelial cells (ECs) that make up the microvascular network barrier contain few pinocytotic vesicles and adhere to each other via tight junctions [57]. Tight junctions limit paracellular transport of hydrophilic compounds into the CNS as compared to non-CNS vessels [58, 59]. Also, astrocytes in close proximity to the ECs add another impediment to paracellular transport by biochemically conditioning the ECs and strengthening the tight junctions between them [60]. ECs coat, in a single layer, the interior of all blood vessels. Because of this intertwined fate with the circulatory system, ECs play a unique role in maintaining physiological homeostasis, controlling the movement of substances across from the blood compartment into the different tissues and organs with varying demands and function [61]. The ECs also play an important immune function through leukocyte surveillance and extravasation by regulating adhesion integrins and cytokine production [62]. In particular, they have been shown to directly secrete tumor necrosis factor (TNF) [63]. Thus, damage to the ECs compromises the integrity of the BBB.

When the barrier between the vascular supply of the brain and the CNS parenchyma is disrupted, excess extravasation of proteins, biologic-response molecules (e.g., growth factors, cytokines, and clotting factors), inflammatory cells, and therapeutic drugs can damage the brain [56, 64–66]. The disruption of the BBB (see Figure 4(b)) has been identified as a consequence of various diseases/injuries such as multiple sclerosis, ischemia, HIV, hypertension, brain tumors, CNS injury, and radiation exposure [66–70], wherein inflammatory cells are able to penetrate the BBB and destroy the myelin



(a) (b)

Figure 4: This diagram depicts a cross section of brain parenchyma showing the structure of the BBB and the damage induced by RT. (a) Normal BBB showing intact tight junctions (TJ), lack of vesicles, astrocytes and pericytes abutting the EC providing additional barrier support, and a neuron with thick, healthy myelin. (b) Damaged BBB in which astrocytes and pericytes have pulled away from the EC, a leukocyte has adhered to the EC, and there is formation of vesicles and loss of TJ integrity.

surrounding the axons. Demyelination and myelin thinning have been reported in the CNS following RT [71–74]. Felts et al. have also shown that RT-induced BBB permeability prolonged the induced demyelination of neurons [75, 76]. We and others [77–80] have demonstrated that there is an increase in BBB permeability following RT, which is caused in part by EC damage, as expressed by changes in tight junction integrity and by vesicle formation postirradiation. RT-induced EC damage has been investigated [81–83] with the aim of elucidating the effect on initiating and/or sustaining radiation side effects. Eissner et al. [84], as well as others [82, 83], have shown that when irradiated, ECs *in vitro* and *in vivo* undergo apoptosis at a higher percentage than other cells. Our studies using electron microscopy show that RT causes damage to the tight junctions [78], which is also connected to the observed increase in BBB permeability. In addition, several studies, including our own, have shown an increase in BBB permeability and an increase in the number of vesicles following fractionated cranial irradiation [78–80].

Such damage to the microvasculature and breach of the BBB can disturb the delicate brain microenvironment and expose the brain parenchyma and neural cells to noxious substances [70, 78, 85]. This microenvironment imbalance can set into motion a chain of events (such as cytokines release), magnifying the original signal and finally causing measurable late-term tissue damage in the irradiated brain that may play a role in cognitive impairment [86]. We and others have shown that RT induces an inflammatory response as indicated by an increase in TNF-α and intercellular adhesion molecule-1 (ICAM-1) signaling in the brain [87–90]. We have reported activated astrocytes after treatment with single and fractionated RT [78, 91]. Prolonged gliosis can create glial scar sites, which have been theorized to inhibit axonal regeneration or remyelination [92, 93]. We have demonstrated that this inflammation response is related to an increase in BBB permeability following RT and that it is abrogated when treated with antibodies to TNF-α or ICAM-1 [77, 90]. In a histological study on mouse brains we observed significant changes 120 days following fractionated RT: fewer neurons, a significant decrease in myelin suggesting complete destruction of the parts of the white matter at 120 days following RT, and at 90 days following RT, we observed swelling of nerve fibers and increased thickening of the myelin sheaths (see Figure 5(b)) indicative of dying axons.

5. The Restorative Potential of Exercise for the Post-RT Brain

Given its myriad beneficial effects on the brain, exercise has been suggested as a treatment for a wide variety of brain maladies, from aging [94] to alcoholism [95]. In the case of aging, exercise has been shown to have a remarkable restorative effect, encouraging the resurgence of atrophied regions such as white matter tracts [96] and the hippocampus [97], and improving cognition [98]. Such effects are particularly encouraging for the post-RT brain, since it shares many things in common with the aged brain, such as decreased cell proliferation, decreased growth hormone, and increased inflammation. Moreover, in both cases these conditions worsen over time, to ultimately create a neural milieu in which plasticity is suppressed.

In aged rodents, exercise can increase proliferation of NSCs [99], suggesting that it has neurogenic potential even in a system in which cell proliferation is drastically reduced. Encouragingly, exercise has been reported to increase hippocampal neurogenesis in the irradiated brain in rodent models [100, 101]. There are likely multiple mechanisms of this enhanced neurogenesis. Neurogenesis is tightly linked to the microenvironment [102] and is known to be suppressed under conditions in which there is unchecked inflammation [27] or a lack of trophic [103] or hormonal support [104]. Exercise has been shown to increase growth hormone [104] and reduce inflammation [105], two potential ways in which it could counteract the suppressive environment created by RT.

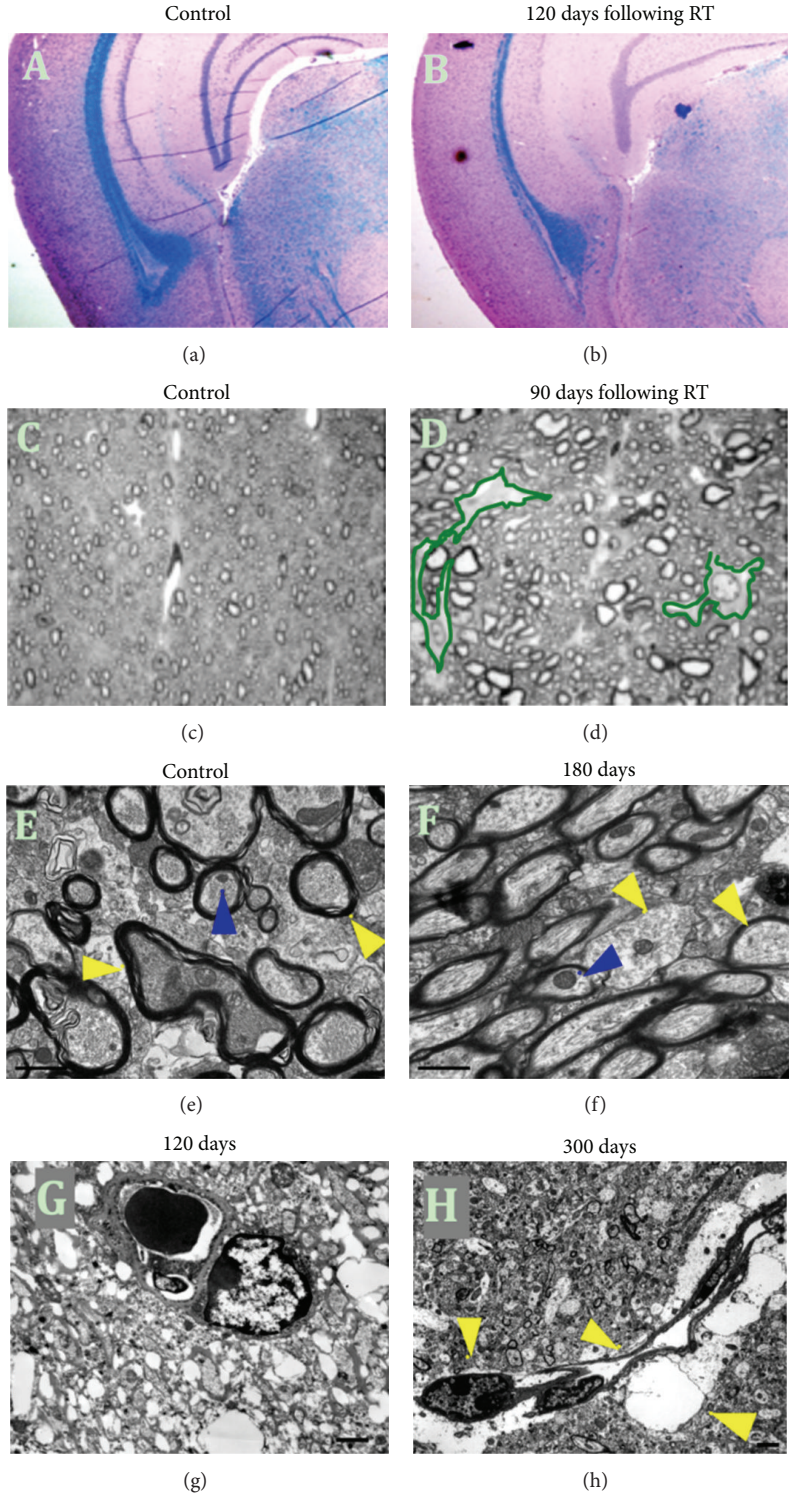


Figure 5: Histological markers of radiation injury in the mouse brain at 90, 120, 180, and 300 days after RT. (a, b) Luxol fast blue staining showing loss of myelin. (c, d) Sections of brain nerve fibers showing structural changes, microglial (outlined in green) inflammation, and myelin sheath thickening indicative of cell death (images at 50x). (e, f) Yellow and blue arrow heads point at myelin sheath surrounding the neurons and at mitochondria, respectively (scale bar = 1 μm). (g) Necrosis detected at 120 days. (h) Yellow arrows point at pericyte pulling away from endothelial cell, a sign of inflammation, with white edematous area causing vascular constriction.

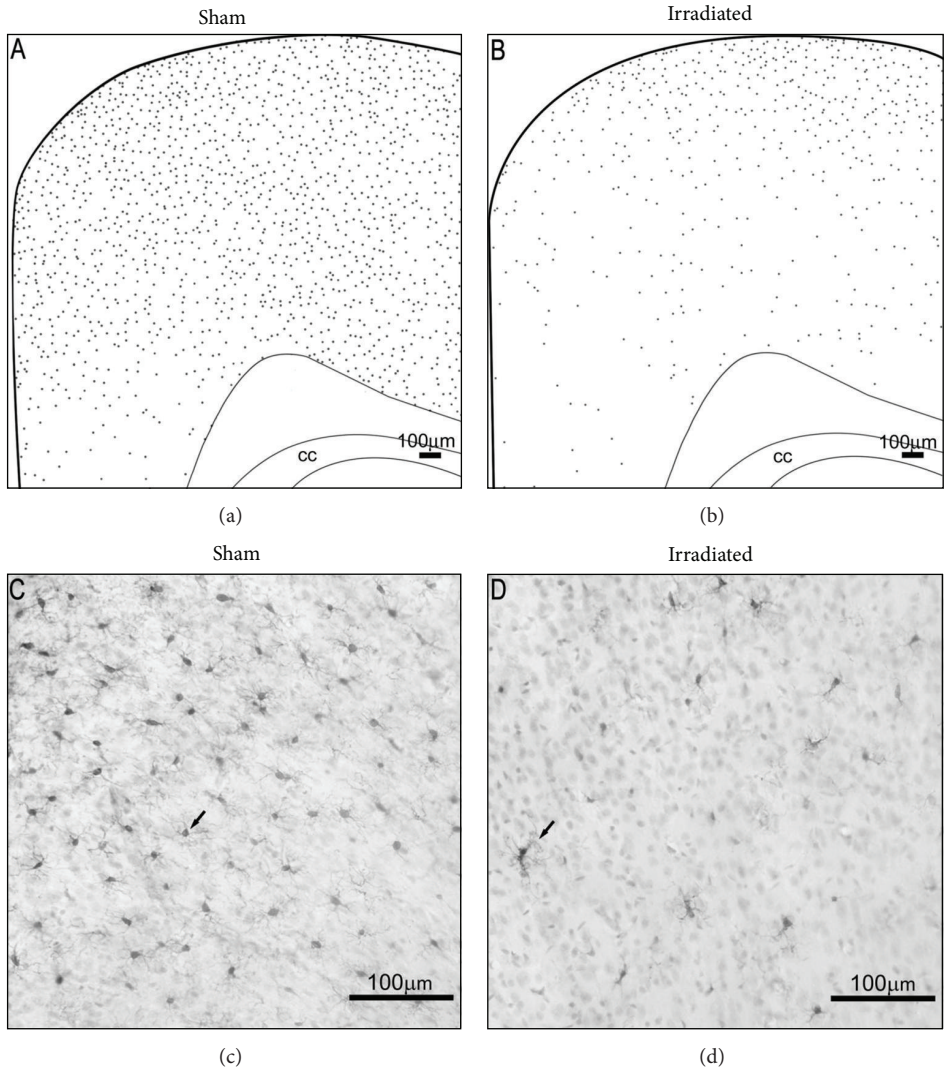


Figure 6: Effect of a single 4 Gy dose of X-ray radiation on microglia in the developing rat brain 24 hr after exposure. (a, b) A schematic representation of microglia distribution (gray dots) in the cerebral cortex showing that RT-induced loss of microglia is more pronounced in inner layers relative to superficial layers. (c, d) Representative 20x images of Iba1+ staining in the retrosplenial cortex showing that RT not only reduces the number of microglia but also alters their morphology.

Recent research has begun to elucidate the important role that microglia have in maintaining the neurogenic niche [106–108]. Unfortunately, radiation severely disrupts microglial distribution, alters their morphology (see Figure 6), and decreases their numbers [109], effects that likely contribute to RT-induced neurogenesis impairment. It has been shown that, after radiation, microglia in the SVZ rebound more quickly than those in the DG. This may explain why neurogenesis recovers better in the SVZ, compared to the dentate [110]. Voluntary exercise has been shown to increase microglia [111], suggesting a further means by which exercise could help to restore a microenvironment conducive to cell proliferation.

Exercise might also have an enhancing effect on gliogenesis in the post-RT brain. As described above, glia are essential for the integrity and function of the cortex. Exercise has been shown to enhance cortical gliogenesis in the intact brain [112], and our future efforts will include determining whether post-RT exercise enhances cortical gliogenesis and ameliorates impairments in the 5-CSRTT.

In addition to these many direct benefits, it is important for survivors of childhood RT to exercise, in order to counteract chronic conditions that arise from cancer treatment, such as impaired pulmonary and cardiac function [113]. Emotional problems like depression and anxiety may also decrease with regular exercise. Unfortunately, treatment-induced fatigue, cardiorespiratory problems, and muscular deconditioning tend to promote sedentary habits during treatment that linger into adulthood, with the result that childhood cancer survivors are much less physically active than their healthy peers (see [113] for review). Cranial RT in particular is associated with sedentary habits in adulthood

[114, 115]. However, recent studies suggest that physical fitness is an achievable goal for childhood cancer survivors [36, 113], so the beneficial effects of exercise observed in animal models can be followed up in human patients.

Fortunately, rodents show no reluctance to exercise after RT, and initial studies suggest that exercise is capable of ameliorating RT-induced deficits in both neurogenesis and cognition. Voluntary running in adulthood has been shown to restore neurogenesis in mice irradiated early in life [100], suggesting that exercise may be a feasible means by which to promote cell proliferation in adult survivors of childhood RT. Furthermore, it may be able to attenuate RT-induced cognitive impairments. A recent study showed that voluntary running ameliorated radiation-induced spatial memory decline 4 months after radiation as well as partially restored neurogenesis in the DG [101].

While these results are encouraging, continued study of the effects of exercise on the RT brain in animal models is essential. For one thing, it is important to better understand the effects of exercise in the context of the RT brain. In particular, it is necessary to determine whether exercise has an adequate neural substrate on which to work. For example, one well-established effect of exercise is its ability to induce angiogenesis [116, 117], an effect that depends upon the brain's capacity to produce new ECs. Given the suppressive effect of RT on cell proliferation, it is possible that the angiogenic effect of exercise would be limited in the post-RT brain. In short, it is possible that the effects of exercise would be dampened in the post-RT brain, reducing its potential as a stand-alone treatment. Further study may indicate that exercise would be most useful as an adjuvant therapy. For example, stem cell replacement shows promise in the irradiated rodent brain [118] and may eventually be possible in humans if the hostile environment created by RT can be made more permissive for growth and repair. Exercise represents a viable means by which to achieve this, and future studies should address the potential of exercise to neutralize the hostile environment created by RT, as a preliminary step in restorative treatments.

6. Conclusions

Both human and animal studies indicate that suppressed cell proliferation and the hostile neural environment induced by cranial RT contribute to subsequent cognitive impairments. These effects of RT may be alleviated, at least to some extent, by exercise. It has been well established that exercise can both promote cell proliferation as well as foster a neural environment permissive for plastic change. Early evidence from animal models indicates that exercise has the capacity to do both in the post-RT brain. However, further studies are needed, in order to determine whether RT-induced perturbations of the microenvironment could limit the plasticity-enhancing effects exercise has to offer.

References

[1] P. Kaatsch, "Epidemiology of childhood cancer," *Cancer Treatment Reviews*, vol. 36, no. 4, pp. 277–285, 2010.

[2] M. A. Smith, N. L. Seibel, S. F. Altekruse et al., "Outcomes for children and adolescents with cancer: challenges for the twenty-first century," *Journal of Clinical Oncology*, vol. 28, no. 15, pp. 2625–2634, 2010.

[3] L. K. Zeltzer, C. Recklitis, D. Buchbinder et al., "Psychological status in childhood cancer survivors: a report from the childhood cancer survivor study," *Journal of Clinical Oncology*, vol. 27, no. 14, pp. 2396–2404, 2009.

[4] B. F. Danoff, F. S. Cowchock, C. Marquette, L. Mulgrew, and S. Kramer, "Assessment of the long-term effects of primary radiation therapy for brain tumors in children," *Cancer*, vol. 49, no. 8, pp. 1580–1586, 1982.

[5] L. Ellenberg, Q. Liu, G. Gioia et al., "Neurocognitive status in long-term survivors of childhood CNS malignancies: a report from the Childhood Cancer Survivor Study," *Neuropsychology*, vol. 23, no. 6, pp. 705–717, 2009.

[6] E. Smibert, V. Anderson, T. Godber, and H. Ekert, "Risk factors for intellectual and educational sequelae of cranial irradiation in childhood acute lymphoblastic leukaemia," *British Journal of Cancer*, vol. 73, no. 6, pp. 825–830, 1996.

[7] B. J. Spiegler, E. Bouffet, M. L. Greenberg, J. T. Rutka, and D. J. Mabbott, "Change in neurocognitive functioning after treatment with cranial radiation in childhood," *Journal of Clinical Oncology*, vol. 22, no. 4, pp. 706–713, 2004.

[8] J. M. Fletcher and D. R. Copeland, "Neurobehavioral effects of central nervous system prophylactic treatment of cancer in children," *Journal of Clinical and Experimental Neuropsychology*, vol. 10, no. 4, pp. 495–537, 1988.

[9] R. J. Packer, L. N. Sutton, T. E. Atkins et al., "A prospective study of cognitive function in children receiving whole-brain radiotherapy and chemotherapy: 2-year results," *Journal of Neurosurgery*, vol. 70, no. 5, pp. 707–713, 1989.

[10] R. K. Mulhern, D. Fairclough, and J. Ochs, "A prospective comparison of neuropsychologic performance of children surviving leukemia who received 18-Gy, 24-Gy, or no cranial irradiation," *Journal of Clinical Oncology*, vol. 9, no. 8, pp. 1348–1356, 1991.

[11] V. Kieffer-Renaux, C. Bulteau, J. Grill, C. Kalifa, D. Viguier, and I. Jambaque, "Patterns of neuropsychological deficits in children with medulloblastoma according to craniospatial irradiation doses," *Developmental Medicine and Child Neurology*, vol. 42, no. 11, pp. 741–745, 2000.

[12] J. Grill, D. Viguier, V. Kieffer et al., "Critical risk factors for intellectual impairment in children with posterior fossa tumors: the role of cerebellar damage," *Journal of Neurosurgery*, vol. 101, no. 2, pp. 152–158, 2004.

[13] C. Eiser, "Neurocognitive sequelae of childhood cancers and their treatment: a comment on Mulhern and Butler," *Pediatric Rehabilitation*, vol. 7, no. 1, pp. 15–16, 2004.

[14] S. Watanabe, Y. Azami, M. Ozawa et al., "Intellectual development after treatment in children with acute leukemia and brain tumor," *Pediatrics International*, vol. 53, no. 5, pp. 694–700, 2011.

[15] V. A. Anderson, T. Godber, E. Smibert, S. Weiskop, and H. Ekert, "Impairments of attention following treatment with cranial irradiation and chemotherapy in children," *Journal of Clinical and Experimental Neuropsychology*, vol. 26, no. 5, pp. 684–697, 2004.

[16] K. A. Lockwood, T. S. Bell, and R. W. Colegrove, "Long-term effects of cranial radiation therapy on attention functioning in survivors of childhood leukemia," *Journal of Pediatric Psychology*, vol. 24, no. 1, pp. 55–66, 1999.

[17] C. B. Reeves, S. L. Palmer, W. E. Reddick et al., "Attention and memory functioning among pediatric patients with medulloblastoma," *Journal of Pediatric Psychology*, vol. 31, no. 3, pp. 272–280, 2006.

[18] R. W. Butler and D. R. Copeland, "Attentional processes and their remediation in children treated for cancer: a literature review and the development of a therapeutic approach," *Journal of the International Neuropsychological Society*, vol. 8, no. 1, pp. 115–124, 2002.

[19] K. R. Wolfe, A. Madan-Swain, and R. K. Kana, "Executive dysfunction in pediatric posterior fossa tumor survivors: a systematic literature review of neurocognitive deficits and interventions," *Developmental Neuropsychology*, vol. 37, no. 2, pp. 153–175, 2012.

[20] B. J. Zebrack, J. G. Gurney, K. Oeffinger et al., "Psychological outcomes in long-term survivors of childhood brain cancer: a report from the childhood cancer survivor study," *Journal of Clinical Oncology*, vol. 22, no. 6, pp. 999–1006, 2004.

[21] B. F. Fuemmeler, T. D. Elkin, and L. L. Mullins, "Survivors of childhood brain tumors: behavioral, emotional, and social adjustment," *Clinical Psychology Review*, vol. 22, no. 4, pp. 547–585, 2002.

[22] M. Bruce, "A systematic and conceptual review of posttraumatic stress in childhood cancer survivors and their parents," *Clinical Psychology Review*, vol. 26, no. 3, pp. 233–256, 2006.

[23] N. E. Langeveld, H. Stam, M. A. Grootenhuis, and B. F. Last, "Quality of life in young adult survivors of childhood cancer," *Supportive Care in Cancer*, vol. 10, no. 8, pp. 579–600, 2002.

[24] J. G. Gurney, K. R. Krull, N. Kadan-Lottick et al., "Social outcomes in the childhood cancer survivor study cohort," *Journal of Clinical Oncology*, vol. 27, no. 14, pp. 2390–2395, 2009.

[25] J. W. Y. Pang, D. L. Friedman, J. A. Whitton et al., "Employment status among adult survivors in the childhood cancer survivor study," *Pediatric Blood and Cancer*, vol. 50, no. 1, pp. 104–110, 2008.

[26] W. R. Brown, R. M. Blair, D. M. Moody et al., "Capillary loss precedes the cognitive impairment induced by fractionated whole-brain irradiation: a potential rat model of vascular dementia," *Journal of the Neurological Sciences*, vol. 257, no. 1-2, pp. 67–71, 2007.

[27] M. L. Monje, H. Toda, and T. D. Palmer, "Inflammatory blockade restores adult hippocampal neurogenesis," *Science*, vol. 302, no. 5651, pp. 1760–1765, 2003.

[28] S. Mizumatsu, M. L. Monje, D. R. Morhardt, R. Rola, T. D. Palmer, and J. R. Fike, "Extreme sensitivity of adult neurogenesis to low doses of X-irradiation," *Cancer Research*, vol. 63, no. 14, pp. 4021–4027, 2003.

[29] M. L. Monje and T. Palmer, "Radiation injury and neurogenesis," *Current Opinion in Neurology*, vol. 16, no. 2, pp. 129–134, 2003.

[30] M. Dennis, B. J. Spiegler, M. C. Obonsawin et al., "Brain tumors in children and adolescents-III. Effects of radiation and hormone status on intelligence and on working, associative and serial-order memory," *Neuropsychologia*, vol. 30, no. 3, pp. 257–275, 1992.

[31] N. Veličković, A. Djordjević, G. Matić, and A. Horvat, "Radiation-induced hyposuppression of the hypothalamic-pituitary-adrenal axis is associated with alterations of hippocampal corticosteroid receptor expression," *Radiation Research*, vol. 169, no. 4, pp. 397–407, 2008.

[32] T. E. Merchant, O. Goloubeva, D. L. Pritchard et al., "Radiation dose-volume effects on growth hormone secretion," *International Journal of Radiation Oncology Biology Physics*, vol. 52, no. 5, pp. 1264–1270, 2002.

[33] S. H. Choi, M. T. Woodlee, J. J. Hong, and T. Schallert, "A simple modification of the water maze test to enhance daily detection of spatial memory in rats and mice," *Journal of Neuroscience Methods*, vol. 156, no. 1-2, pp. 182–193, 2006.

[34] C. Duggan, L. Bechard, K. Donovan et al., "Changes in resting energy expenditure among children undergoing allogeneic stem cell transplantation," *American Journal of Clinical Nutrition*, vol. 78, no. 1, pp. 104–109, 2003.

[35] A. C. White, N. Terrin, K. B. Miller, and H. F. Ryan, "Impaired respiratory and skeletal muscle strength in patients prior to hematopoietic stem-cell transplantation," *Chest*, vol. 128, no. 1, pp. 145–152, 2005.

[36] K. R. Wolfe, G. R. Hunter, A. Madan-Swain, A. T. Reddy, J. Baños, and R. K. Kana, "Cardiorespiratory fitness in survivors of pediatric posterior fossa tumor," *Journal of Pediatric Hematology/Oncology*, vol. 34, no. 6, pp. e222–e227, 2012.

[37] L. Iughetti, P. Bruzzi, B. Predieri, and P. Paolucci, "Obesity in patients with acute lymphoblastic leukemia in childhood," *Italian Journal of Pediatrics*, vol. 38, no. 1, article 4, 2012.

[38] F. H. Gage, "Mammalian neural stem cells," *Science*, vol. 287, no. 5457, pp. 1433–1438, 2000.

[39] T. J. Shors, M. L. Anderson, D. M. Curlik, and M. S. Nokia, "Use it or lose it: how neurogenesis keeps the brain fit for learning," *Behavioural Brain Research*, vol. 227, pp. 450–458, 2012.

[40] W. Deng, M. D. Saxe, I. S. Gallina, and F. H. Gage, "Adult-born hippocampal dentate granule cells undergoing maturation modulate learning and memory in the brain," *Journal of Neuroscience*, vol. 29, no. 43, pp. 13532–13542, 2009.

[41] M. L. Monje, H. Vogel, M. Masek, K. L. Ligon, P. G. Fisher, and T. D. Palmer, "Impaired human hippocampal neurogenesis after treatment for central nervous system malignancies," *Annals of Neurology*, vol. 62, no. 5, pp. 515–520, 2007.

[42] M. Monje and J. Dietrich, "Cognitive side effects of cancer therapy demonstrate a functional role for adult neurogenesis," *Behavioural Brain Research*, vol. 227, pp. 376–379, 2012.

[43] A. A. Nageswara Rao, H. Ye, P. A. Decker, C. L. Howe, and C. Wetmore, "Therapeutic doses of cranial irradiation induce hippocampus-dependent cognitive deficits in young mice," *Journal of Neuro-Oncology*, vol. 105, no. 2, pp. 191–198, 2011.

[44] J. S. Snyder, N. S. Hong, R. J. McDonald, and J. M. Wojtowicz, "A role for adult neurogenesis in spatial long-term memory," *Neuroscience*, vol. 130, no. 4, pp. 843–852, 2005.

[45] A. Barlind, N. Karlsson, T. Björk-Eriksson, J. Isgaard, and K. Blomgren, "Decreased cytogenesis in the granule cell layer of the hippocampus and impaired place learning after irradiation of the young mouse brain evaluated using the IntelliCage platform," *Experimental Brain Research*, vol. 201, no. 4, pp. 781–787, 2010.

[46] Y. Fan, Z. Liu, P. R. Weinstein, J. R. Fike, and J. Liu, "Environmental enrichment enhances neurogenesis and improves functional outcome after cranial irradiation," *European Journal of Neuroscience*, vol. 25, no. 1, pp. 38–46, 2007.

[47] P. Achanta, M. Fuss, and J. L. Martinez, "Ionizing radiation impairs the formation of trace fear memories and reduces hippocampal neurogenesis," *Behavioral Neuroscience*, vol. 123, no. 5, pp. 1036–1045, 2009.

[48] J. M. Wojtowicz, M. L. Askew, and G. Winocur, "The effects of running and of inhibiting adult neurogenesis on learning and memory in rats," *European Journal of Neuroscience*, vol. 27, no. 6, pp. 1494–1502, 2008.

[49] G. Winocur, J. M. Wojtowicz, M. Sekeres, J. S. Snyder, and S. Wang, "Inhibition of neurogenesis interferes with hippocampus-dependent memory function," *Hippocampus*, vol. 16, no. 3, pp. 296–304, 2006.

[50] D. R. Kornack and P. Rakic, "Cell proliferation without neurogenesis in adult primate neocortex," *Science*, vol. 294, no. 5549, pp. 2127–2130, 2001.

[51] A. Barzilai, "The neuro-glial-vascular interrelations in genomic instability symptoms," *Mechanisms of Ageing and Development*, vol. 132, no. 8-9, pp. 395–404, 2011.

[52] E. E. Benarroch, "Neuron-astrocyte interactions: partnership for normal function and disease in the central nervous system," *Mayo Clinic Proceedings*, vol. 80, no. 10, pp. 1326–1338, 2005.

[53] D. Qiu, D. L. W. Kwong, G. C. F. Chan, L. H. T. Leung, and P. L. Khong, "Diffusion tensor magnetic resonance imaging finding of discrepant fractional anisotropy between the frontal and parietal lobes after whole-brain irradiation in childhood medulloblastoma survivors: reflection of regional white matter radiosensitivity?" *International Journal of Radiation Oncology Biology Physics*, vol. 69, no. 3, pp. 846–851, 2007.

[54] R. K. Mulhern, W. E. Reddick, S. L. Palmer et al., "Neurocognitive deficits in medulloblastoma survivors and white matter loss," *Annals of Neurology*, vol. 46, no. 6, pp. 834–841, 1999.

[55] A. Bari, J. W. Dalley, and T. W. Robbins, "The application of the 5-choice serial reaction time task for the assessment of visual attentional processes and impulse control in rats," *Nature Protocols*, vol. 3, no. 5, pp. 759–767, 2008.

[56] H. Davson and M. B. Segal, *Physiology of the CSF and Blood-Brain Barriers*, CRC Press, Boca Raton, Fla, USA, 1996.

[57] A. R. Burns, D. C. Walker, E. S. Brown et al., "Neutrophil transendothelial migration is independent of tight junctions and occurs preferentially at tricellular corners," *Journal of Immunology*, vol. 159, no. 6, pp. 2893–2903, 1997.

[58] C. C. Michel and C. R. Neal, "Openings through endothelial cells associated with increased microvascular permeability," *Microcirculation*, vol. 6, no. 1, pp. 45–54, 1999.

[59] S. Quarmby, P. Kumar, and S. Kumar, "Radiation-induced normal tissue injury: role of adhesion molecules in leukocyte-endothelial cell interactions," *International Journal of Cancer*, vol. 82, no. 3, pp. 385–395, 1999.

[60] L. L. Rubin and J. M. Staddon, "The cell biology of the blood-brain barrier," *Annual Review of Neuroscience*, vol. 22, pp. 11–28, 1999.

[61] C. Michiels, "Endothelial cell functions," *Journal of Cellular Physiology*, vol. 196, no. 3, pp. 430–443, 2003.

[62] C. D. Buckley, G. Ed Rainger, G. B. Nash, and K. Raza, "Endothelial cells, fibroblasts and vasculitis," *Rheumatology*, vol. 44, no. 7, pp. 860–863, 2005.

[63] T. Imaizumi, H. Itaya, K. Fujita et al., "Expression of tumor necrosis factor-α in cultured human endothelial cells stimulated with lipopolysaccharide or interleukin-1α," *Arteriosclerosis, Thrombosis, and Vascular Biology*, vol. 20, no. 2, pp. 410–415, 2000.

[64] W. A. Banks, "Physiology and pathology of the blood-brain barrier: implications for microbial pathogenesis, drug delivery and neurodegenerative disorders," *Journal of NeuroVirology*, vol. 5, no. 6, pp. 538–555, 1999.

[65] W. G. Mayhan, "Regulation of blood-brain barrier permeability," *Microcirculation*, vol. 8, no. 2, pp. 89–104, 2001.

[66] P. Rubin, D. M. Gash, J. T. Hansen, D. F. Nelson, and J. P. Williams, "Disruption of the blood-brain barrier as the primary effect of CNS irradiation," *Radiotherapy and Oncology*, vol. 31, no. 1, pp. 51–60, 1994.

[67] C. F. Brosnan, "Brain microvasculature in multiple sclerosis," in *Introduction To the Blood-Brain Barrier Methodology, Biology, and Pathology*, W. M. Pardridge, Ed., pp. 386–400, Cambridge University Press, Cambridge, UK, 1998.

[68] H. M. Wisniewski, *Introduction to the Blood Brain Barrier Methodology, Biology and Pathology*, Cambridge University Press, New York, NY, USA, 1998.

[69] C. K. Petito, "HIV infection and the blood-brain barrier," in *Introduction To the Blood-Brain Barrier Methodology, Biology, and Pathology*, W. M. Pardridge, Ed., pp. 419–433, Cambridge University Press, Cambridge, UK, 1998.

[70] R. A. Nordal and C. S. Wong, "Molecular targets in radiation-induced blood-brain barrier disruption," *International Journal of Radiation Oncology Biology Physics*, vol. 62, no. 1, pp. 279–287, 2005.

[71] S. Atkinson, Y. Q. Li, and C. S. Wong, "Changes in oligodendrocytes and myelin gene expression after radiation in the rodent spinal cord," *International Journal of Radiation Oncology Biology Physics*, vol. 57, no. 4, pp. 1093–1100, 2003.

[72] C. S. Chiang, K. A. Mason, H. R. Withers, and W. H. McBride, "Alteration in myelin-associated proteins following spinal cord irradiation in guinea pigs," *International Journal of Radiation Oncology Biology Physics*, vol. 24, no. 5, pp. 929–937, 1992.

[73] C. S. Chiang, W. H. McBride, and H. R. Withers, "Myelin-associated changes in mouse brain following irradiation," *Radiotherapy and Oncology*, vol. 27, no. 3, pp. 229–236, 1993.

[74] F. L. Mastaglia, W. I. McDonald, J. V. Watson, and K. Yogendran, "Effects of X radiation on the spinal cord: an experimental study of the morphological changes in central nerve fibres," *Brain*, vol. 99, no. 1, pp. 101–122, 1976.

[75] P. A. Felts and K. J. Smith, "Blood-brain barrier permeability in astrocyte-free regions of the central nervous system remyelinated by Schwann cells," *Neuroscience*, vol. 75, no. 2, pp. 643–655, 1996.

[76] Q. R. Smith and S. I. Rapoport, "Cerebrovascular permeability coefficients to sodium, potassium, and chloride," *Journal of Neurochemistry*, vol. 46, no. 6, pp. 1732–1742, 1986.

[77] H. Yuan, M. W. Gaber, T. McColgan, M. D. Naimark, M. F. Kiani, and T. E. Merchant, "Radiation-induced permeability and leukocyte adhesion in the rat blood-brain barrier: modulation with anti-ICAM-1 antibodies," *Brain Research*, vol. 969, no. 1-2, pp. 59–69, 2003.

[78] H. Yuan, M. W. Gaber, K. Boyd, C. M. Wilson, M. F. Kiani, and T. E. Merchant, "Effects of fractionated radiation on the brain vasculature in a murine model: blood-brain barrier permeability, astrocyte proliferation, and ultrastructural changes," *International Journal of Radiation Oncology Biology Physics*, vol. 66, no. 3, pp. 860–866, 2006.

[79] D. D'Avella, R. Cicciarello, F. Albiero et al., "Quantitative study of blood-brain barrier permeability changes after experimental whole-brain radiation," *Neurosurgery*, vol. 30, no. 1, pp. 30–34, 1992.

[80] P. L. Debbage, S. Seidl, A. Kreczy, P. Hutzler, M. Pavelka, and P. Lukas, "Vascular permeability and hyperpermeability in a murine adenocarcinoma after fractionated radiotherapy: an

ultrastructural tracer study," *Histochemistry and Cell Biology*, vol. 114, no. 4, pp. 259–275, 2000.

[81] C. Belka, W. Budach, R. D. Kortmann, and M. Bamberg, "Radiation induced CNS toxicity—molecular and cellular mechanisms," *British Journal of Cancer*, vol. 85, no. 9, pp. 1233–1239, 2001.

[82] F. Paris, Z. Fuks, A. Kang et al., "Endothelial apoptosis as the primary lesion initiating intestinal radiation damage in mice," *Science*, vol. 293, no. 5528, pp. 293–297, 2001.

[83] L. A. Peña, Z. Fuks, and R. N. Kolesnick, "Radiation-induced apoptosis of endothelial cells in the murine central nervous system: protection by fibroblast growth factor and sphingomyelinase deficiency," *Cancer Research*, vol. 60, no. 2, pp. 321–327, 2000.

[84] G. Eissner, F. Kohlhuber, M. Grell et al., "Critical involvement of transmembrane tumor necrosis factor-α in endothelial programmed cell death mediated by ionizing radiation and bacterial endotoxin," *Blood*, vol. 86, no. 11, pp. 4184–4193, 1995.

[85] M. S. Anscher, L. Chen, Z. Rabbani et al., "Recent progress in defining mechanisms and potential targets for prevention of normal tissue injury after radiation therapy," *International Journal of Radiation Oncology Biology Physics*, vol. 62, no. 1, pp. 255–259, 2005.

[86] V. Sarkissian, "The sequelae of cranial irradiation on human cognition," *Neuroscience Letters*, vol. 382, no. 1-2, pp. 118–123, 2005.

[87] C. S. Chiang, J. H. Hong, A. Stalder, J. R. Sun, H. R. Withers, and W. H. McBride, "Delayed molecular responses to brain irradiation," *International Journal of Radiation Biology*, vol. 72, no. 1, pp. 45–53, 1997.

[88] M. W. Gaber, O. M. Sabek, K. Fukatsu, H. G. Wilcox, M. F. Kiani, and T. E. Merchant, "Differences in ICAM-1 and TNF-α expression between large single fraction and fractionated irradiation in mouse brain," *International Journal of Radiation Biology*, vol. 79, no. 5, pp. 359–366, 2003.

[89] J.-H. Hong, C.-S. Chiang, I. L. Campbell, J.-R. Sun, H. R. Withers, and W. H. McBride, "Induction of acute phase gene expression by brain irradiation," *International Journal of Radiation Oncology Biology Physics*, vol. 33, no. 3, pp. 619–626, 1995.

[90] C. M. Wilson, M. W. Gaber, O. M. Sabek, J. A. Zawaski, and T. E. Merchant, "Radiation-induced astrogliosis and blood-brain barrier damage can be abrogated using anti-TNF treatment," *International Journal of Radiation Oncology Biology Physics*, vol. 74, no. 3, pp. 934–941, 2009.

[91] R. Cicciarello, D. D'Avella, M. E. Gagliardi et al., "Time-related ultrastructural changes in an experimental model of whole brain irradiation," *Neurosurgery*, vol. 38, no. 4, pp. 772–780, 1996.

[92] V. Balasingam, T. Tejada-Berges, E. Wright, R. Bouckova, and Voon Wee Yong, "Reactive astrogliosis in the neonatal mouse brain and its modulation by cytokines," *Journal of Neuroscience*, vol. 14, no. 2, pp. 846–856, 1994.

[93] K. W. Selmaj, M. Farooq, W. T. Norton, C. S. Raine, and C. F. Brosnan, "Proliferation of astrocytes in vitro in response to cytokines. A primary role for tumor necrosis factor," *Journal of Immunology*, vol. 144, no. 1, pp. 129–135, 1990.

[94] C. H. Hillman, K. I. Erickson, and A. F. Kramer, "Be smart, exercise your heart: exercise effects on brain and cognition," *Nature Reviews Neuroscience*, vol. 9, no. 1, pp. 58–65, 2008.

[95] R. A. Brown, A. M. Abrantes, J. P. Read et al., "Aerobic exercise for alcohol recovery: rationale, program description, and preliminary findings," *Behavior Modification*, vol. 33, no. 2, pp. 220–249, 2009.

[96] S. J. Colcombe, K. I. Erickson, P. E. Scalf et al., "Aerobic exercise training increases brain volume in aging humans," *Journals of Gerontology A*, vol. 61, no. 11, pp. 1166–1170, 2006.

[97] K. I. Erickson, M. W. Voss, R. S. Prakash et al., "Exercise training increases size of hippocampus and improves memory," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 7, pp. 3017–3022, 2011.

[98] S. Colcombe and A. F. Kramer, "Fitness effects on the cognitive function of older adults: a meta-analytic study," *Psychological Science*, vol. 14, no. 2, pp. 125–130, 2003.

[99] H. Van Praag, T. Shubert, C. Zhao, and F. H. Gage, "Exercise enhances learning and hippocampal neurogenesis in aged mice," *Journal of Neuroscience*, vol. 25, no. 38, pp. 8680–8685, 2005.

[100] A. S. Naylor, C. Bull, M. K. L. Nilsson et al., "Voluntary running rescues adult hippocampal neurogenesis after irradiation of the young mouse brain," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 38, pp. 14632–14637, 2008.

[101] S. J. E. Wong-Goodrich, M. L. Pfau, C. T. Flores, J. A. Fraser, C. L. Williams, and L. W. Jones, "Voluntary running prevents progressive memory decline and increases adult hippocampal neurogenesis and growth factor expression after whole-brain irradiation," *Cancer Research*, vol. 70, no. 22, pp. 9329–9338, 2010.

[102] C. Zhao, W. Deng, and F. H. Gage, "Mechanisms and functional implications of adult neurogenesis," *Cell*, vol. 132, no. 4, pp. 645–660, 2008.

[103] M. LLorens-Martín, I. Torres-Alemán, and J. L. Trejo, "Exercise modulates insulin-like growth factor 1-dependent and -independent effects on adult hippocampal neurogenesis and behaviour," *Molecular and Cellular Neuroscience*, vol. 44, no. 2, pp. 109–117, 2010.

[104] D. G. Blackmore, M. G. Golmohammadi, B. Large, M. J. Waters, and R. L. Rietze, "Exercise increases neural stem cell number in a growth hormone-dependent manner, augmenting the regenerative response in aged mice," *Stem Cells*, vol. 27, no. 8, pp. 2044–2052, 2009.

[105] T. Archer, A. Fredriksson, E. Schütz, and R. M. Kostrzewa, "Influence of physical exercise on neuroimmunological functioning and health: aging and stress," *Neurotoxicity Research*, vol. 20, no. 1, pp. 69–83, 2011.

[106] Y. Ziv, N. Ron, O. Butovsky et al., "Immune cells contribute to the maintenance of neurogenesis and spatial learning abilities in adulthood," *Nature Neuroscience*, vol. 9, no. 2, pp. 268–275, 2006.

[107] A. Nimmerjahn, F. Kirchhoff, and F. Helmchen, "Neuroscience: resting microglial cells are highly dynamic surveillants of brain parenchyma in vivo," *Science*, vol. 308, no. 5726, pp. 1314–1318, 2005.

[108] A. Sierra, J. M. Encinas, J. J. P. Deudero et al., "Microglia shape adult hippocampal neurogenesis through apoptosis-coupled phagocytosis," *Cell Stem Cell*, vol. 7, no. 4, pp. 483–495, 2010.

[109] M. Kalm, B. Lannering, T. Björk-Eriksson, and K. Blomgren, "Irradiation-induced loss of microglia in the young brain," *Journal of Neuroimmunology*, vol. 206, no. 1-2, pp. 70–75, 2009.

[110] N. A. K. Hellström, O. R. Lindberg, A. Ståhlberg et al., "Unique gene expression patterns indicate microglial contribution to neural stem cell recovery following irradiation," *Molecular and Cellular Neuroscience*, vol. 46, no. 4, pp. 710–719, 2011.

[111] D. Ehninger and G. Kempermann, "Regional effects of wheel running and environmental enrichment on cell genesis and microglia proliferation in the adult murine neocortex," *Cerebral Cortex*, vol. 13, no. 8, pp. 845–851, 2003.

[112] C. D. Mandyam, S. Wee, A. J. Eisch, H. N. Richardson, and G. F. Koob, "Methamphetamine self-administration and voluntary exercise have opposing effects on medial prefrontal cortex gliogenesis," *Journal of Neuroscience*, vol. 27, no. 42, pp. 11442–11450, 2007.

[113] A. F. S. Juan, K. Wolin, and A. Lucía, "Physical activity and pediatric cancer survivorship," *Recent Results in Cancer Research*, vol. 186, pp. 319–347, 2011.

[114] T. A. Florin, G. E. Fryer, T. Miyoshi et al., "Physical inactivity in adult survivors of childhood acute lymphoblastic leukemia: a report from the childhood cancer survivor study," *Cancer Epidemiology Biomarkers and Prevention*, vol. 16, no. 7, pp. 1356–1363, 2007.

[115] K. K. Ness, W. M. Leisenring, S. Huang et al., "Predictors of inactive lifestyle among adult survivors of childhood cancer," *Cancer*, vol. 115, no. 9, pp. 1984–1994, 2009.

[116] J. E. Black, K. R. Isaacs, B. J. Anderson, A. A. Alcantara, and W. T. Greenough, "Learning causes synaptogenesis, whereas motor activity causes angiogenesis, in cerebellar cortex of adult rats," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 87, no. 14, pp. 5568–5572, 1990.

[117] R. A. Swain, A. B. Harris, E. C. Wiener et al., "Prolonged exercise induces angiogenesis and increases cerebral blood volume in primary motor cortex of the rat," *Neuroscience*, vol. 117, no. 4, pp. 1037–1046, 2003.

[118] M. M. Acharya, L. A. Christie, M. L. Lan et al., "Rescue of radiation-induced cognitive impairment through cranial transplantation of human embryonic stem cells," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 45, pp. 19150–19155, 2009.

Hindawi Publishing Corporation
Neural Plasticity
Volume 2013, Article ID 805497, 14 pages
http://dx.doi.org/10.1155/2013/805497

Review Article

The Neural Plasticity Theory of Depression: Assessing the Roles of Adult Neurogenesis and PSA-NCAM within the Hippocampus

Steven R. Wainwright and Liisa A. M. Galea

Program in Neuroscience, Centre for Brain Health, Department of Psychology, University of British Columbia, Vancouver, BC, Canada V6T 1Z4

Correspondence should be addressed to Liisa A. M. Galea; lgalea@psych.ubc.ca

Received 5 February 2013; Accepted 13 March 2013

Academic Editor: Chitra D. Mandyam

Depression is a devastating and prevalent disease, with profound effects on neural structure and function; however the etiology and neuropathology of depression remain poorly understood. Though antidepressant drugs exist, they are not ideal, as only a segment of patients are effectively treated, therapeutic onset is delayed, and the exact mechanism of these drugs remains to be elucidated. Several theories of depression do exist, including modulation of monoaminergic neurotransmission, alterations in neurotrophic factors, and the upregulation of adult hippocampal neurogenesis, and are briefly mentioned in the review. However none of these theories sufficiently explains the pathology and treatment of depression unto itself. Recently, neural plasticity theories of depression have postulated that multiple aspects of brain plasticity, beyond neurogenesis, may bridge the prevailing theories. The term "neural plasticity" encompasses an array of mechanisms, from the birth, survival, migration, and integration of new neurons to neurite outgrowth, synaptogenesis, and the modulation of mature synapses. This review critically assesses the role of adult hippocampal neurogenesis and the cell adhesion molecule, PSA-NCAM (which is known to be involved in many facets of neural plasticity), in depression and antidepressant treatment.

1. Neural Plasticity and Disease

As the fundamental units of the brain, neurons function to integrate and transmit a myriad of signals across vast and complex networks. Neurons by definition are continually forming, eliminating, and modulating (strengthening and weakening) connections in response to the constant flow of information. In mediating and responding to activity, neurons, including their processes and synapses, must be plastic. Thus neural plasticity may then be defined as the ability of neurons and neural elements to adapt in response to intrinsic and extrinsic signals. As such, our ability to process and synthesize information, ultimately producing behavior, is dependent upon this neural plasticity [1]. It is therefore not surprising that dysregulation or disruption of neural plasticity is associated with neuropsychiatric and neurodegenerative disease.

Dynamic processes such as adult neurogenesis, the development of dendritic spines, and synaptic adaptations are included under the umbrella of neural plasticity and are essential to normal functioning. Aberrant neural production, connectivity, or transmission is invariably present under disease states, such as Alzheimer's disease, schizophrenia, or depression [2–5]. Each aspect of neural plasticity can independently and additively cause or contribute to the disease state. Thus, it is of fundamental importance to elucidate the neurological underpinnings of these disease states at the cellular level, in order to understand etiology and better develop effective treatment.

2. Adult Hippocampal Neurogenesis

Neural stem cells were first identified in the adult brain of rodents more than 50 years ago [6] and are found across a variety of species including humans [7]. Interestingly, the production of new neurons is typically limited to two regions: the subventricular zone, which lines the lateral ventricles and sends new neurons to the olfactory bulb via the rostral migratory stream, and the subgranular zone of the hippocampus [8].

It is important to note that neurogenesis, as defined here, requires the proliferation, survival, and differentiation of newly generated cells into neurons. Any number of internal and/or external factors may independently affect the proliferation of progenitor cells, their differentiation into neurons, or their survival rates [9–14]. Thus merely identifying cells as having been produced in the adult brain, without further demonstrating that they survive and become neurons, is insufficient to conclude that neurogenesis has occurred.

Although new neurons can be labelled and observed, the exact role of these neurons in the function of the hippocampus remains to be fully elucidated. New neurons produced in the subgranular zone have been associated with a number of functions [15, 16]. There is solid evidence that new hippocampal neurons are selectively and imperatively involved in spatial learning and memory [17–20]. New neurons in the hippocampus, perhaps more ambiguously, have also been associated with the etiology and treatment of depression [21–23]. Interestingly, the hippocampus is functionally dissociated along the dorsal-ventral axis, wherein the dorsal region plays a larger role in cognitive faculties, while the ventral region is more involved in emotionality [24, 25]. For the purpose this review, however, we will concentrate on the functions of the hippocampus associated with emotionality and stress and refer the reader to reviews that cover the hippocampus' cognitive associations (See: [26, 27]).

3. Cell Adhesion Molecules

Neural plasticity is a complex and varied process; from neural development, the migration of newly generated neurons, dendritic modifications, and synaptic modulation, many proteins facilitate and contribute to the malleability of neural tissue. Cell adhesion molecules (CAMs) are specialized proteins—typically expressed at the cell surface—which are important in synaptic function, synaptic plasticity, and remodeling of neural circuits [28]. The structure and function of CAMs vary widely within the nervous system, and the categorization and function of each CAM protein involved in neural plasticity are beyond the scope of this review (for a more detailed overview see [28, 29]). This review will instead focus on the potential role of a single CAM, the polysialylated form of neural cell adhesion molecule (PSA-NCAM), in the etiology and treatment of depression.

The neural cell adhesion molecule (NCAM) is a member of the immunoglobulin superfamily of cell adhesion molecules and serves to mediate $Ca2^{+}$-independent cell-cell and cell-extracellular matrix (ECM) interactions [30]. Through homo- and heterophilic interactions, NCAM functions in cell migration, neurite outgrowth and targeting, axonal branching, synaptogenesis, and synaptic plasticity [30–32]. Neural plasticity mediated through the NCAM protein is facilitated through posttranslational modifications, the most important and prevalent of which is glycosylation with polysialic acid (PSA) [33]. Polysialic acid is a linear homopolymer of α2,8-linked sialic acid, which bears a negative charge, acting to abate NCAM-NCAM interactions and therefore interfere with cell adhesion [34]. PSA-NCAM serves to regulate cell-cell and cell-ECM interactions during times of plasticity.

In the adult brain PSA-NCAM is expressed on the cell surface of newly generated daughter cells, on neurites during outgrowth and path finding, and at the synapse of mature neurons [30]. The addition of the PSA moiety to NCAM is essential to neural remodelling and synaptic plasticity [10, 35, 36]. Selective cleavage of PSA in the adult brain inhibits activity-induced synaptic plasticity (induction of long-term potentiation (LTP) and long-term depression (LTD)) and alters the normal migration and integration of newly generated neurons within the hippocampus [10, 35], importantly though cleavage of PSA from NCAM does not disrupt normal basal synaptic neurotransmission or alter normal levels of neural proliferation or survival [10, 35]. PSA-NCAM is therefore a particularly interesting protein in that it is one that mediates plasticity at multiple levels, from neural proliferation, integration, differentiation, neuritic outgrowth, synaptogenesis, and modulation of mature synapses.

4. Depression

Depression is a devastating neuropsychiatric disease that is both prevalent—with a lifetime incidence of 20% [37]—and costly, accruing over $83 billion (USD) annually in expense to society in the USA alone [38]. Depression is a spectrum disorder ranging from dysthymia to major depressive disorder (MDD), which also includes seasonal (seasonal affective disorder) and bipolar subtypes; however general symptomology is characterized by a depressed mood and/or anhedonia [39]. The DSM-IV criteria for diagnosis with MDD may include secondary symptoms such as fatigue, insomnia or hypersomnia, changes in body weight, and impaired cognition [39]. Although depression is prevalent and carries a large burden of disease, the pathoetiology of depression is poorly understood. While antidepressant drugs do exist, they are not ideal, as only a segment of patients is effectively treated, and the therapeutic onset is delayed [40–42]. Moreover, the exact mechanism of these drugs remains to be elucidated, though several theories do exist. The existing theories of etiology and treatment of depression will be briefly discussed with an aim of integrating and placing neurogenesis and PSA-NCAM related plasticity within the current state of knowledge.

5. Monoamine Theory of Depression

Much of the current understanding of depression has been built upon the serendipitous discovery of the mood-elevating effects of iproniazid, a monoamine oxidase inhibitor (MAOI), during clinical trials for antituberculosis agents in the early 1950s [43, 44]. Around the same time the first tricyclic antidepressant (TCA), imipramine, a drug also known to modulate monoaminergic neurotransmission, was discovered to relieve depressive symptoms [45]. Both compounds enhance monoaminergic neurotransmitters (primarily serotonin and norepinephrine) function in the synapse and therefore acted as a springboard for the rational design of drugs which specifically enhance the transmission of these neuromodulators. Further, these findings resulted in the development

of the monoamine hypothesis of depression [46, 47]. The monoamine hypothesis of depression postulates that the pathophysiological basis of depression is due to the deficient activity of monoamines in the central nervous system [48].

While first generation antidepressant drugs can indeed be effective, remission rates still typically remain below 60%, and treatment is associated with potentially severe side effects, causing a third of patients to discontinue treatment [49, 50]. Thus new drugs, such as selective serotonin reuptake inhibitors (SSRIs), have been developed in an attempt to enhance efficacy and reduce the side effects observed with the broader-acting first generation compounds. However, the ability of these newer drugs to alleviate depression is, unfortunately, typically lower than that of MAOIs and TCAs [50]. SSRIs are the primary first line treatment for patients with major depressive disorder (MDD), yet only a third of patients will respond to these drugs following initial treatment [41]. Even in patients that are initially responsive to treatment, up to 57% will have depressive symptoms return due to a loss of drug efficacy [40].

While it is known that antidepressants modify monoaminergic neurotransmission, it is not known how antidepressants exert their therapeutic effects, as the influence on monoamines occurs within hours of treatment, but alleviation of depressive symptoms requires weeks of exposure [51–54]. The monoamine theory explains this delayed efficacy of treatment as the time required for the 5-HT1A serotonin autoreceptor to sensitize and normalize serotonergic tone at the synapse [42, 55, 56]. However, disrupting or ablating portions of the serotonergic system fails to induce a depressive phenotype [57], suggesting that the dysregulation of serotonergic neurotransmission is not the only underlying factor in depression. Given the prevalence of treatment-resistant forms of depression and the limited long-term efficacy of current drugs, novel targets are needed for the development of more efficacious pharmacological treatments.

6. Neurogenic Hypothesis of Depression

As noted earlier, neurogenesis in the adult hippocampus is seen across a variety of species, including humans [7]. Adult hippocampal neurogenesis has been hypothesized to play a potentially important role on the pathology and successful treatment of depression. A neurogenic hypothesis of depression has been postulated, which suggests that reduced adult hippocampal neurogenesis may underlie the pathoetiology of depression, while antidepressant efficacy depends on the upregulation of hippocampal neurogenesis [58].

7. Etiology of Depression and Animal Models

As with other neuropsychiatric disorders, depression has a multifaceted and varied etiology, including genetic, epigenetic, and environmental factors. The most prevalent of these factors is stress. Stress is cited as the leading cause of depression by depressed patients [59]. Unipolar depression is associated with abnormal hypothalamic-pituitary adrenal (HPA) axis function such as hypersecretion of cortisol, abnormal diurnal secretion of cortisol resulting in a flattened circadian rhythm and impaired negative feedback [59, 60]. Interestingly, normalization of HPA function is seen after chronic exposure to antidepressants, an effect coincident with, or slightly preceding, behavioural alleviation of depressive symptoms [61]. As such normalization of HPA function has become a major target of novel therapies [59, 62].

Many animal models of depression capitalize on the association of stress and depression; such models have a solid ethological basis. For instance, models using exposure to chronic variable stress (CVS) have shown good face, construct, and predictive validity, making it one of the most commonly used paradigms to model depression [63]. For example, CVS increases immobility in the forced swim test (FST, a putative measure of behavioural despair) [64], reduces sucrose preference (anhedonia) [65], increases novelty-induced hypophagia [21, 64], reduces hippocampal neurogenesis [21, 65], and reduces the expression of proteins associated with neuroplasticity such as PSA-NCAM [66]. Although it is not possible to model all symptoms of depression in rodents, a battery of key endophenotypes can be examined such as anhedonia, body weight changes, behavioural despair, HPA function, and brain alterations (such as reduced hippocampal volume, neurogenesis, and neural plasticity), as have been observed in human patients with depression [67].

8. Stress, Depression, and Reduced Neural Plasticity

The hippocampal formation is an area rich in mineralocorticoid (MRs) and glucocorticoid receptors (GRs). These receptors function in the maintenance of basal HPA tone and in the regulation of negative feedback of glucocorticoid release during a stress response [68]. Given this, it is not surprising that the hippocampus is particularly vulnerable to the effects of stress and depression [69, 70]. Depressed patients have reduced hippocampal volume which varies with the number of episodes and duration of the illness [71–73]. Similarly, postmortem studies of hippocampal tissue collected from depressed patients have shown alterations in gray matter density, reductions in neuropil, and decreased hippocampal neurogenesis [22, 23, 74–77]. Animal models of depression using exposure to chronic stress demonstrate the same changes in hippocampal structure and show reductions in neural plasticity, including reductions in neurogenesis and the expression of proteins associated with neural plasticity [21, 64, 65, 78]. Chronic stress exposure also reduces dendrite length and complexity [79, 80], spine density [81], and NCAM expression [65] and modifies expression of synaptic SNARE proteins [82] in the hippocampus.

Stabilization of the HPA axis by antidepressants is associated with improved mood scores that either precedes or is coincident with the behavioural alleviation of depressive symptoms in humans [61]. Thus the improvements in negative feedback inhibition of the HPA axis that correspond to improvement in depressive symptoms with antidepressant treatment may be attributable to neural adaptation in limbic structures that regulate feedback inhibition of HPA axis such as the hippocampus. Interestingly chronic antidepressant treatment is seen to reverse the negative effects of stress on

hippocampal volume [65, 83], though the exact contribution of neural plastic elements that underlie this enhancement is not fully understood.

9. Neurogenesis in Antidepressant Efficacy

The neurogenic hypothesis of depression is predicated not only on the previous findings that chronic stress decreases hippocampal neurogenesis [21, 65, 84] but also that antidepressant drugs can both prevent [83, 85] and reverse [21, 65, 86] this effect (see Table 1). The enhancement of adult hippocampal neurogenesis is dependent on chronic, but not acute, exposure to antidepressants, occurring in a manner that temporally coincides with the delayed clinical efficacy of these drugs [9, 21]. This delayed enhancement of neurogenesis is mediated through the increased proliferation of neural progenitor cells, while the proportion and the differentiation of surviving cells into neurons remain constant [9]. Several classes of antidepressant including SSRIs, TCAs, MAOIs, SNRIs, and melatonergic antidepressants increase neurogenesis in the hippocampus [87, 88]. Moreover, the nonpharmacological antidepressant treatment of electroconvulsive therapy (ECT) also enhances neurogenesis in the adult hippocampus [89, 90]. ECT is the most effective antidepressant treatment in depressed patients who are treatment resistant [91], and it can upregulate neurogenesis to levels nearly twice that of pharmacological antidepressants [89]. Similarly, recent human studies suggest that the broader acting, typically more efficacious, TCA and MAOI classes of antidepressant increase cell proliferation levels beyond that of SSRIs [23]; further indicating a role for hippocampal neurogenesis in successful antidepressant treatment.

While it is accepted that chronic antidepressant treatment upregulates hippocampal neurogenesis, some studies have suggested that increased neurogenesis is absolutely necessary for antidepressant efficacy. Specifically, if hippocampal neurogenesis is reduced via localized irradiation, antidepressants lose their behavioural efficacy in rodent models of depression [21]. However, use of the cytostatic agent methylazoxymethanol (MAM) to reduce hippocampal neurogenesis failed to disrupt the behavioural efficacy of chronic antidepressant treatment on sucrose preference or in the forced swim test; antidepressant treatment still increased hippocampal volume and NCAM expression, despite the disruption of hippocampal neurogenesis [65]. Nonetheless, neurogenesis does seem to be required for the attenuation of anxiety-like behaviour as measured in the novelty suppressed feeding (NSF) test, suggesting a role for neurogenesis in specific aspects of antidepressant efficacy. Indeed, further research has shown neurogenic-dependent and -independent effects of an antidepressant drug, where hippocampal neurogenesis is only required for the alleviation of some anxiety/depressive-like behaviour [64, 92–94]. These findings point toward multifaceted and pleiotropic effects of antidepressants on neural circuitry. Interestingly anxiolytic drugs, such as benzodiazepines, also show efficacy in the NSF test with acute treatment, while antidepressants require chronic, neurogenesis-enhancing treatment [95]. It is then possible that the reduction in glucocorticoids from anxiolytic treatment mediates the reduction in anxiety-like behaviour in the NSF test; therefore antidepressant-induced neurogenesis may mediate a similar effect on glucocorticoid levels. Indeed this may be the case, as hippocampal neurogenesis buffers the stress response and normalizes glucocorticoid release after stress [96]. Synder and colleagues found that inhibition of hippocampal neurogenesis, either transgenically or via irradiation, attenuated the recovery of basal HPA tone following a stressor and the normal suppression of glucocorticoid release during the dexamethasone suppression test [96]. As such, there is likely a role for hippocampal neurogenesis in reestablishing normal HPA tone and regulating a normal HPA response, possibly through GR-mediated negative feedback. Thus the effectiveness of antidepressant treatment and the importance of ensuing neurogenesis within different behavioural measures of antidepressant efficacy may represent the underlying mechanisms through which antidepressants are exerting their effects. Separate facets of antidepressant-induced neural plasticity may therefore facilitate drug action such as neurogenesis-mediated normalization of the stress response in the NSF test versus modulation of monoaminergic tone through synaptic plasticity in the sucrose preference test and FST. Hence the contribution of broader neural plasticity, beyond neurogenesis, is also of interest and has been proposed as the underlying mechanism of antidepressant efficacy [65].

10. Potential Role for PSA-NCAM in Antidepressant Efficacy

The expression of PSA-NCAM within the adult nervous system is essential in multiple facets of neural plasticity, including neurogenesis, synaptic plasticity, and neurite outgrowth [10, 30, 31, 97, 98]. Importantly, PSA-NCAM functions across all aspects of hippocampal neurogenesis (proliferation, migration, differentiation, and survival), therefore changes in the polysialylation of newly generated neurons alter neurogenesis as a whole [99–104] (see Table 2). For instance, chronic mild stress reduces the expression of both the core NCAM protein [65] and the addition of the PSA moiety [11, 105–108] in the hippocampus; however these changes are dependent on the type of stressor [109] (see Table 3). Alterations in PSA-NCAM expression following stress are also seen in other regions associated with depression including the amygdala and prefrontal cortex [110–113]. Similarly, PSA-NCAM expression is reduced in amygdala of patients with major depressive disorder [2, 114]; however no change is seen in the PFC [115]. Conversely, chronic antidepressant treatment modulates PSA-NCAM expression throughout the limbic system in animal models of depression. [11, 116–119]. Moreover, antidepressant treatment reduces PSA-NCAM expression in the dorsal raphe nucleus [116], implicating PSA-NCAM in antidepressant-induced plasticity related to serotonergic neurotransmission. Taken together these findings suggest PSA-NCAM may play a fundamental role in mediating broad effects of antidepressant treatment across multiple forms of neural plasticity.

Previous studies have shown PSA-NCAM is essential in the activity-dependent induction of hippocampal LTP and

Table 1: Selected publications showing the effect of stress/depression or antidepressant treatment on neurogenesis and the functional importance of neurogenesis in antidepressant efficacy.

(a) Animal models of depression

Reference	Species	Sex	Model	Antidepressant	Method of ablation	Summary of findings
Snyder et al. 2011 [96]	Mouse	M	Restraint	—	X-ray; transgenic	Neurogenesis-dependent regulation of HPA response to stress and behavioural measures (NSF, FST, and SC)
David et al. 2009 [64]	Mouse	M and F	Chronic corticosterone administration	Fluoxetine	X-ray	Neurogenesis-dependent (NSF) and -independent (OFT, FST) aspects of antidepressant efficacy
Bessa et al. 2009 [65]	Rat	M	Chronic mild stress	Imipramine; fluoxetine	MAM	Neurogenesis-dependent (NSF) and -independent (SC, FST) aspects of antidepressant efficacy. Significant alterations in neural plasticity associated with antidepressant efficacy
Surget et al. 2008 [92]	Mouse	M	Chronic unpredictable stress	Imipramine; fluoxetine		Neurogenesis-dependent (NSF, CS, ST) and -independent (A) aspects of antidepressant efficacy
Holick et al. 2008 [93]	Mouse	M	—	Fluoxetine	X-ray	Neurogenesis-independent effects of antidepressant efficacy
Airan et al. 2007 [94]	Rat	F	Chronic mild stress	Imipramine; fluoxetine	X-ray	Neurogenesis-dependent (NSF) and -independent (OFT) aspects of antidepressant efficacy
Alonso et al. 2004 [85]	Mouse	M	Chronic mild stress	Fluoxetine	—	Decreased cell proliferation in the DG, while chronic fluoxetine blocked this effect
Santarelli et al. 2003 [21]	Mouse	M	—	Imipramine; fluoxetine	X-ray	Neurogenesis-dependent (NSF) antidepressant efficacy
Czéh et al. 2002 [78]	Rat	M	Social subordination	—	—	Decreased cell proliferation and survival in the DG
Malberg et al. 2000 [9]	Rat	M	—	Fluoxetine; reboxetine; tranylcypromine; ECS	—	Chronic, but not acute, treatment with monoaminergic antidepressant and ECS increased cell proliferation in the DG

(b) Human studies of depression

Reference	Subjects	Sex	Population assessed	Antidepressant	Effect on neurogenesis
Cobb et al. 2013 [74]	Humans	M and F	Depressed patients postmortem	—	No significant difference in number of granule cells between depressed subjects and controls; decreased hippocampal volume correlating with duration of disease
Boldrini et al. 2013 [75]	Humans	M and F	Depressed patients postmortem	SSRIs; TCAs	Depression is associated with a decreased number of granule neurons, correlated with reduced DG volume. SSRI and TCA treatment increase granule neuron number and DG volume
Boldrini et al. 2012 [22]	Humans	M and F	Depressed patients postmortem	SSRIs; TCAs	Both antidepressant classes increase cell proliferation over untreated depressed patients and controls; NPCs associated with angiogenesis
Boldrini et al. 2009 [23]	Humans	M and F	Depressed patients postmortem	SSRIs; TCAs	Both antidepressant classes increase cell proliferation over untreated depressed patients and controls
Stockmeier et al. 2004 [76]	Humans	M and F	Depressed patients postmortem	—	Increased density of granule cells in the DG of depressed subjects compared to controls

NSF: novelty suppressed feeding; FST: forced swim test; SC: sucrose consumption; OFT: open field test; CS: coat state; ST: splash test; A: actimeter; MAM: methylazoxymethanol acetate (cytostatic agent).

LTD [35], where the polysialylation of NCAM is positively correlated with neural activity of glutamatergic neurons [120, 121]. Moreover, inhibiting the polysialylation of NCAM reduces the rate of dendritic spine formation and decreases the stability of these spines [122]. Interestingly PSA-NCAM is also directly related to monoaminergic neurotransmission, as PSA-NCAM expression is directly modulated by the 5-HT1A receptor [123]. Moreover, serotonergic innervation plays a role in regulating the expression of PSA-NCAM in the hippocampus [124]. Given the interaction of serotonergic

Table 2: Selected publications delineating the role of PSA-NCAM in adult hippocampal neurogenesis.

Authors	Species	Role in neurogenesis	Ablation method	Findings
McCall et al. 2013 [97]	Rat	Neurite outgrowth; survival	EndoN	Cleavage of PSA-NCAM caused expanded dendritic arborization and increased cell death
Burgess et al. 2008 [10]	Rat	Migration; differentiation	EndoN	Cleavage of PSA-NCAM disrupts normal migration and differentiation of newly generated neurons in the DG
Seri et al. 2004 [99]	?	Differentiation	—	PSA-NCAM is highly expressed in the entire cell body and growing processes of D cells (precursors in the generation of new granule neurons in the dentate gyrus)
Ni Dhuill et al. 1999 [100]	Human	Proliferation; neurite outgrowth	—	Hippocampal expression of PSA-NCAM throughout life in humans closely resembles that of the rat. Expression largely contained to granule cells of the dentate gyrus and their mossy fiber axons, with large reductions in expression with age
Seki and Rutishauser 1998 [98]	Mouse	Neurite outgrowth	NCAM KO; EndoN	Aberrant collateral sprouting of mossy fibers and ectopic synaptic bouton formation
Kuhn et al. 1996 [101]	Rat	Migration	—	Age related decline in PSA-NCAM expression in the GCL, reduced migration of PSA-NCAM expressing cells into the GCL
Fox et al. 1995 [102]	Rat	Proliferation	—	PSA-NCAM expression decreases with age, coinciding with decreased cell proliferation
Seki and Arai 1993 [103]	Rat	Proliferation; migration; differentiation	—	Newly generated granule cells in the dentate gyrus express a highly polysialylated form of NCAM, involved in the migration of immature neurons from the subgranular zone into the GCL
Seki and Arai 1991 [104]	Rat	Proliferation	—	Highly polysialylated form of NCAM is persistently expressed in the adult dentate gyrus

EndoN: endoneuraminidase N; GCL: granule cell layer.

neurotransmission with PSA-NCAM and its putative role in regulating synaptic plasticity, it is possible that PSA-NCAM may mediate some of the effects of antidepressant treatment on monoaminergic tone.

The ability of PSA-NCAM to directly alter hippocampal neurogenesis is evidenced by the selective removal of the PSA moiety from NCAM with the bacteriophage enzyme EndoN. Application of EndoN disrupts normal migration of newly produced cells and alters differentiation of these new cells by shifting them toward a neuronal phenotype [10]. Selective cleavage of PSA also produces enhanced dendritic arborization, aberrant mossy fiber sprouting, produces ectopic synaptic bouton formation, and increases cell death within the hippocampus [97, 98].

Given the role of PSA-NCAM in mediating multiple aspects of neural plasticity it appears to function at the confluence of the prevailing theories of depression: monoaminergic, neurotrophic, and neurogenic, thus making it an interesting target for future study. Further research is necessary to elucidate the role of PSA-NCAM and similar proteins, in the etiology of depression and efficacy of antidepressant drugs.

11. Role of Neurotrophic Factors in Antidepressant Efficacy

Brain-derived neurotrophic factor (BDNF) has been implicated in the etiology and treatment of depression. Depressed patients show decreased levels of serum BDNF [125, 126], which is correlated with decreased hippocampal volume and increased ratings of depression [126]. Conversely, increased BDNF levels are associated with antidepressant treatment and alleviation of depression [127]. These findings are mirrored in rodent models of depression [128–132] as chronic antidepressant treatment increases BDNF levels, coinciding with alleviation of depressive-like behaviours [130–132], and intracranial infusion of BDNF produces antidepressant effects [128, 133].

Activation of the TrkB neurotrophin receptor largely mediates the plasticity-enhancing actions of BDNF [134–136], where inhibition of TrkB signalling attenuates the behavioural efficacy of antidepressants [136]. Conversely BDNF, or more specifically proBDNF, via activation of the p75 neurotrophin receptor mediates plasticity-reducing actions such as apoptosis, neural atrophy, and synaptic pruning [137]. It has been postulated that the balance of TrkB and p75 signalling may underlie antidepressant effects of BDNF, suggesting a mechanism through which stress/depression may modulate the effects of BDNF. To this point, knocking out TrkB receptors fails to produce a depressive-like phenotype in neurogenesis-dependent or -independent behavioural measures of antidepressant efficacy [138, 139]; however sex differences exist as female TrkB knockouts do appear to develop a depressive-like behavioural phenotype [140].

Interestingly, PSA-NCAM interacts with BDNF, as removal of PSA from NCAM inhibits the induction of LTP, and the application of exogenous BDNF restores LTP at the affected synapse [141]. Further, disruption of the polysialylation of NCAM also disturbs the effects of BDNF on cortical neuron differentiation and survival, while the application of exogenous BDNF reverses these effects [142].

Table 3: Effects of stress and depression on the expression of PSA-NCAM.

(a) Animal models of stress and depression

Reference	Species	Sex	Model	Antidepressant	Effect on PSA-NCAM
Gilabert-Juan et al. 2012 [110]	Mouse	M	Chronic restraint stress	—	↔ in mPFC
Djordjevic et al. 2012 [111]	Rat	M	Chronic social isolation	—	↑ in HPC; ↓ PFC
Djordjevic et al. 2012 [109]	Rat	M	Chronic social isolation	Fluoxetine	↑ in HPC, ↓ by Flx treatment
Djordjevic et al. 2012 [112]	Rat	M	Chronic social isolation	Fluoxetine	↑ in PFC; ↓ by Flx treatment (with stress)
Gilabert-Juan et al. 2011 [113]	Mouse	M	Chronic restraint stress	—	↓ in CeM; ↔ in BLA; ↔ Me
Wainwright et al. 2011 [11]	Rat	M	Unpredictable chronic mild stress	—	↓ in HPC
Homberg et al. 2011 [116]	Rat	M	—	Fluoxetine	↓ in dRN; ↔ in mPFC; ↑ AMYG (adolescent), ↓ AMYG (adult)
Varea et al. 2007 [117]	Rat	M	—	Fluoxetine	↑ in HPC (str. luc. only); ↓ in Me and BMA; ↔ in BLA
Sairanen et al. 2007 [118]	Rat	M	—	Imipramine	↑ in HPC; ↑ plPFC
Varea et al. 2007 [119]	Rat	M	—	Fluoxetine	↑ in mPFC (whole); ↑ ilPFC; ↔ plPFC
Cordero et al. 2005 [105]	Rat	M	Chronic restraint stress	—	↓ in CeM; ↓ Me
Nacher et al. 2004 [66]	Rat	M	Oral corticosterone administration	—	↓ in HPC
Nacher et al. 2004 [106]	Rat	M	Chronic restraint stress; oral corticosterone administration	—	↓ in piriform cortex (oral CORT); ↑ in piriform cortex (restraint)
Pham et al. 2003 [107]	Rat	M	Chronic restraint stress	—	↑ in HPC (3 weeks), ↔ in HPC (6 weeks)
Sandi et al. 2001 [108]	Rat	M	Chronic restraint stress	—	↑ in HPC

(b) Human studies of depression

Reference	Subjects	Sex	Population assessed	Antidepressant	Effect on PSA-NCAM
Maheu et al. 2013 [114]	Human	?	Depressed patients postmortem	Specific classes not disclosed	↓ in BLA
Gilabert-Juan et al. 2012 [115]	Human	M and F	Depressed patients postmortem	Specific classes not disclosed	↔ in dlPFC
Varea et al. 2012 [2]	Human	M and F	Depressed patients postmortem	Specific classes not disclosed	↓ in BLA; ↓ in BMA

Me: medial amygdala; CeM: centromedial amygdala; BMA: basomedial amygdala; dRN: dorsal raphe nucleus; str. luc.: stratum lucidum; plPFC: prelimbic cortex; ilPFC: infralimbic prefrontal cortex; HPC: hippocampus; AMYG: amygdala; mPFC: medial prefrontal cortex.

These findings suggest that PSA-NCAM may mediate the responsiveness of neurons to BDNF. It is known that PSA-NCAM interacts with, and may regulate, p75 expression in septal neurons [143] and newly generated neurons of the SVZ [144]. Indeed knockout of p75 significantly reduces the expression of PSA-NCAM in SVZ neuroblasts [145]. As previously mentioned the p75 receptor is involved in the regulation of adult hippocampal neurogenesis [146] and regulates neurogenesis stimulated by chronic antidepressant treatment [147]. Given the complex and varied role of BDNF in the etiology of depression and antidepressant treatment more research is needed to further elucidate a mechanism through which the modulation of BDNF exerts its influence.

12. Gonadal Hormones, Depression, and Modulation of Neural Plasticity

Women are twice as likely as men to develop depression [148]. Sex differences are also seen in antidepressant efficacy as men have a better response to TCAs, while women have a better response to SSRIs [149], although these findings remain controversial. Any sex difference observed suggests that gonadal hormone levels are involved, and indeed there is evidence that androgens may protect males from the development of depression. Interestingly, there is an increased incidence of depression in males coinciding with the age related decline in testosterone levels [150–153]. Similarly, young hypogonadal males are more susceptible to developing depression [154], portending protective effects of testosterone against the development of depression. Testosterone has shown some antidepressant action as testosterone replacement therapies are efficacious in alleviating depressive symptoms in hypogonadal men [152, 155, 156]. Testosterone replacement also has efficacy as an adjunct treatment to clinical antidepressants in cases of treatment-resistant depression [155, 157]; however it should be noted that androgen therapies are not always seen to be effective for men suffering depression [158, 159].

Gonadal hormones in females are also likely a factor in the treatment and etiology of depression. Times of dramatic hormone fluctuation, such as during the postpartum and

perimenopause, are associated with an increased incidence in depression [160, 161]. In addition hormonal replacement can show antidepressant effects during the postpartum and in peri- and postmenopausal women [162–164]. Consistent with clinical studies, gonadal hormones are also effective as an adjunct therapy to chronic antidepressant treatment in animal models. For example, chronic imipramine increases hippocampal neurogenesis in intact but not in ovariectomized rats [165]. Another study found that an SSRI decreased immobility in the FST in ovariectomized female rats, but only with adjunct estradiol treatment [166]. Similarly, testosterone potentiates the effects of imipramine in increasing cell proliferation and facilitates the alleviation of depressive-like behavioural phenotypes in castrated socially isolated male rats [167]. Consequently androgens and estrogens may have antidepressant properties which could impede the development of, or ameliorate, extant depressive disorders.

Interestingly gonadal hormones modulate adult neurogenesis, BDNF levels, and PSA-NCAM expression in the hippocampus. Testosterone and its metabolite dihydrotestosterone (DHT) both serve to enhance hippocampal neurogenesis through improved cell survival via an androgen-dependent mechanism within the dentate gyrus [168]. Similarly, estrogens are able to increase adult neurogenesis via proliferation or survival depending on duration of treatment [169]. Estrogens regulate the polysialylation of NCAM across the estrous cycle [170], while the removal of testicular hormones decreases PSA-NCAM expression in the dentate gyrus [11]. Significant interplay exists between gonadal hormones and BDNF, as testosterone and DHT interact with BDNF to modulate synaptic plasticity, dendritic morphology, and neurogenesis in the central nervous system [171–174]. Estrogens and BDNF also share widespread interactions, as estradiol regulates hippocampal BDNF [175, 176] levels possibly through an estrogen-sensitive response element on the BDNF gene [177]. Conversely, BDNF mediates estradiol-induced alterations in hippocampal dendritic spine density [178]. Thus gonadal hormones may play a role in facilitating antidepressant efficacy through the modulation of neural plasticity.

13. Effects of Experimental Manipulation on Neural Plasticity

Many of the studies examining a reduction in neurogenesis have used viral vectors, irradiation, or the cytostatic agent, methylazoxymethanol. It is important to note that the experimental manipulation of neurogenesis also likely produces downstream effects on neural plasticity in a much broader sense, including the expression of cell adhesion molecules (including PSA-NCAM), synaptic proteins, and BDNF. As such, it is difficult to know for sure whether the attenuation of antidepressant efficacy via reductions in neurogenesis is due to changes in neurogenesis levels or other aspects of neural plasticity. For instance, radiation levels as low as 2.5 Gy significantly reduce the density of BDNF-expressing neurons [179] and the expression of PSA-NCAM in the hippocampus [180]. This is important as a 10 Gy dose of radiation is typically used to inhibit neurogenesis in studies assessing attenuated neurogenesis [21, 96, 181], suggesting BDNF and PSA-NCAM expression will also be affected. This suggests that the effects of radiation affect proteins associated with neural remodelling and synaptic plasticity as well as neurogenesis [180]. It is important to address these caveats in future research to further elucidate the role of neurogenesis and neural plasticity in depression and its treatment.

14. Conclusion

Depression is a complex neuropsychiatric disease with a poorly defined etiology. While hosts of antidepressant drugs do exist, they are often inefficacious. Elucidating the neural underpinnings of depression and fully understanding the pleiotropic effects of current antidepressant compounds, beyond their role in modulating monoaminergic neurotransmission, are necessary for the development of more effective drugs. The potential role of neurogenesis, both its decline with the occurrence of depression and its enhancement by chronic treatment with antidepressant drugs and therapies, has provided a promising avenue for research. Though significant findings have been made relating neurogenesis to the effective treatment of depression, large gaps in our understanding still exist. To this end, the role of synaptic proteins and cell adhesion molecules, including PSA-NCAM, in the etiology and treatment of depression should also be investigated. As with neurogenesis, PSA-NCAM is reduced in depressed patients and in models of depression, while chronic antidepressant treatment increases expression of PSA-NCAM. Importantly however, PSA-NCAM mediates multiple facets of neural plasticity, including neurogenesis and synaptic plasticity, in addition to mediating effects of neurotrophic factors, such as BDNF. PSA-NCAM therefore functions at the confluence of many forms of neural plasticity, and its mechanisms bridge several theories of depression: monoamine, neurogenic, and neurotrophic. This review has focused on the potential roles of neurogenesis and PSA-NCAM in depression; however many other proteins are associated with neural plasticity and depression [29, 182, 183]. Given the limitations in the understanding of the genesis of depression and current antidepressant treatments, continued research into the exact contribution of adult neurogenesis and the potential roles of proteins associated with neural plasticity and the continued perusal of a broader, neural plasticity hypothesis of depression are certainly warranted.

Acknowledgment

Liisa A. M. Galea was funded for this research by a grant from the Coast Capital Depression Fund.

References

[1] L. Wilbrecht, A. Holtmaat, N. Wright, K. Fox, and K. Svoboda, "Structural plasticity underlies experience-dependent functional plasticity of cortical circuits," *Journal of Neuroscience*, vol. 30, no. 14, pp. 4927–4932, 2010.

[2] E. Varea, R. Guirado, J. Gilabert-Juan et al., "Expression of PSA-NCAM and synaptic proteins in the amygdala of psychiatric disorder patients," *Journal of Psychiatric Research*, vol. 46, no. 2, pp. 189–197, 2012.

[3] S. T. DeKosky and S. W. Scheff, "Synapse loss in frontal cortex biopsies in Alzheimer's disease: correlation with cognitive severity," *Annals of Neurology*, vol. 27, no. 5, pp. 457–464, 1990.

[4] P. Calabresi, N. B. Mercuri, and M. Di Filippo, "Synaptic plasticity, dopamine and Parkinson's disease: one step ahead," *Brain*, vol. 132, no. 2, pp. 285–287, 2009.

[5] D. A. Lewis and G. González-Burgos, "Neuroplasticity of neocortical circuits in schizophrenia," *Neuropsychopharmacology*, vol. 33, no. 1, pp. 141–165, 2008.

[6] J. Altman, "Are new neurons formed in the brains of adult mammals?" *Science*, vol. 135, no. 3509, pp. 1127–1128, 1962.

[7] P. S. Eriksson, E. Perfilieva, T. Björk-Eriksson et al., "Neurogenesis in the adult human hippocampus," *Nature Medicine*, vol. 4, no. 11, pp. 1313–1317, 1998.

[8] M. S. Kaplan and J. W. Hinds, "Neurogenesis in the adult rat: electron microscopic analysis of light radioautographs," *Science*, vol. 197, no. 4308, pp. 1092–1094, 1977.

[9] J. E. Malberg, A. J. Eisch, E. J. Nestler, and R. S. Duman, "Chronic antidepressant treatment increases neurogenesis in adult rat hippocampus," *Journal of Neuroscience*, vol. 20, no. 24, pp. 9104–9110, 2000.

[10] A. Burgess, S. R. Wainwright, L. S. Shihabuddin, U. Rutishauser, T. Seki, and I. Aubert, "Polysialic acid regulates the clustering, migration, and neuronal differentiation of progenitor cells in the adult hippocampus," *Developmental Neurobiology*, vol. 68, no. 14, pp. 1580–1590, 2008.

[11] S. R. Wainwright, S. E. Lieblich, and L. A. Galea, "Hypogonadism predisposes males to the development of behavioural and neuroplastic depressive phenotypes," *Psychoneuroendocrinology*, vol. 36, no. 9, pp. 1327–1341, 2011.

[12] G. Kempermann, H. G. Kuhn, and F. H. Gage, "Experience-induced neurogenesis in the senescent dentate gyrus," *Journal of Neuroscience*, vol. 18, no. 9, pp. 3206–3212, 1998.

[13] J. M. Barker and L. A. M. Galea, "Repeated estradiol administration alters different aspects of neurogenesis and cell death in the hippocampus of female, but not male, rats," *Neuroscience*, vol. 152, no. 4, pp. 888–902, 2008.

[14] H. van Praag, G. Kempermann, and F. H. Gage, "Running increases cell proliferation and neurogenesis in the adult mouse dentate gyrus," *Nature Neuroscience*, vol. 2, no. 3, pp. 266–270, 1999.

[15] G. Kempermann, "Why new neurons? Possible functions for adult hippocampal neurogenesis," *Journal of Neuroscience*, vol. 22, no. 3, pp. 635–638, 2002.

[16] R. S. Duman, J. Malberg, and S. Nakagawa, "Regulation of adult neurogenesis by psychotropic drugs and stress," *Journal of Pharmacology and Experimental Therapeutics*, vol. 299, no. 2, pp. 401–407, 2001.

[17] J. R. Epp, M. D. Spritzer, and L. A. M. Galea, "Hippocampus-dependent learning promotes survival of new neurons in the dentate gyrus at a specific time during cell maturation," *Neuroscience*, vol. 149, no. 2, pp. 273–285, 2007.

[18] J. S. Snyder, N. S. Hong, R. J. McDonald, and J. M. Wojtowicz, "A role for adult neurogenesis in spatial long-term memory," *Neuroscience*, vol. 130, no. 4, pp. 843–852, 2005.

[19] E. Gould, A. Beylin, P. Tanapat, A. Reeves, and T. J. Shors, "Learning enhances adult neurogenesis in the hippocampal formation," *Nature Neuroscience*, vol. 2, no. 3, pp. 260–265, 1999.

[20] T. J. Shors, G. Miesegaes, A. Beylin, M. Zhao, T. Rydel, and E. Gould, "Neurogenesis in the adult is involved in the formation of trace memories," *Nature*, vol. 410, no. 6826, pp. 372–376, 2001.

[21] L. Santarelli, M. Saxe, C. Gross et al., "Requirement of hippocampal neurogenesis for the behavioral effects of antidepressants," *Science*, vol. 301, no. 5634, pp. 805–809, 2003.

[22] M. Boldrini, R. Hen, M. D. Underwood et al., "Hippocampal angiogenesis and progenitor cell proliferation are increased with antidepressant use in major depression," *Biological Psychiatry*, vol. 72, no. 7, pp. 562–571, 2012.

[23] M. Boldrini, M. D. Underwood, R. Hen et al., "Antidepressants increase neural progenitor cells in the human hippocampus," *Neuropsychopharmacology*, vol. 34, no. 11, pp. 2376–2389, 2009.

[24] M. S. Fanselow and H. W. Dong, "Are the dorsal and ventral hippocampus functionally distinct structures?" *Neuron*, vol. 65, no. 1, pp. 7–19, 2010.

[25] D. M. Bannerman, J. N. Rawlins, S. B. McHugh et al., "Regional dissociations within the hippocampus—memory and anxiety," *Neuroscience & Biobehavioral Reviews*, vol. 28, no. 3, pp. 273–283, 2004.

[26] A. Gärtner and D. Frantz, *Hippocampus: Anatomy, Functions and Neurobiology. Neuroanatomy Research at the Leading Edge Series*, vol. 11, Nova Science, New York, NY, USA, 2010.

[27] M. Koehl and D. N. Abrous, "A new chapter in the field of memory: adult hippocampal neurogenesis," *European Journal of Neuroscience*, vol. 33, no. 6, pp. 1101–1114, 2011.

[28] M. B. Dalva, A. C. McClelland, and M. S. Kayser, "Cell adhesion molecules: signalling functions at the synapse," *Nature Reviews Neuroscience*, vol. 8, no. 3, pp. 206–220, 2007.

[29] C. Sandi, "Stress, cognitive impairment and cell adhesion molecules," *Nature Reviews Neuroscience*, vol. 5, no. 12, pp. 917–930, 2004.

[30] L. Bonfanti, "PSA-NCAM in mammalian structural plasticity and neurogenesis," *Progress in Neurobiology*, vol. 80, no. 3, pp. 129–164, 2006.

[31] L. Bonfanti and D. T. Theodosis, "Polysialic acid and activity-dependent synapse remodeling," *Cell Adhesion and Migration*, vol. 3, no. 1, pp. 43–50, 2009.

[32] O. Senkov, O. Tikhobrazova, and A. Dityatev, "PSA-NCAM: synaptic functions mediated by its interactions with proteoglycans and glutamate receptors," *The International Journal of Biochemistry & Cell Biology*, vol. 44, no. 4, pp. 591–595, 2012.

[33] J. Finne, U. Finne, H. Deagostini-Bazin, and C. Goridis, "Occurrence of α2-8 linked polysialosyl units in a neural cell adhesion molecule," *Biochemical and Biophysical Research Communications*, vol. 112, no. 2, pp. 482–487, 1983.

[34] E. Gascon, L. Vutskits, and J. Z. Kiss, "Polysialic acid-neural cell adhesion molecule in brain plasticity: from synapses to integration of new neurons," *Brain Research Reviews*, vol. 56, no. 1, pp. 101–118, 2007.

[35] D. Muller, C. Wang, G. Skibo et al., "PSA-NCAM is required for activity-induced synaptic plasticity," *Neuron*, vol. 17, no. 3, pp. 413–422, 1996.

[36] U. Rutishauser, M. Watanabe, and J. Silver, "Specific alteration of NCAM-mediated cell adhesion by an endoneuraminidase," *Journal of Cell Biology*, vol. 101, no. 5, pp. 1842–1849, 1985.

[37] T. B. Ustun, J. L. Ayuso-Mateos, S. Chatterji, C. Mathers, and C. J. L. Murray, "Global burden of depressive disorders in the year 2000," *The British Journal of Psychiatry*, vol. 184, pp. 386–392, 2004.

[38] P. E. Greenberg and H. G. Birnbaum, "The economic burden of depression in the US: societal and patient perspectives," *Expert Opinion on Pharmacotherapy*, vol. 6, no. 3, pp. 369–376, 2005.

[39] American Psychiatric Association, *Diagnostic Criteria From DSM-IV-TR*, vol. 12, American Psychiatric Association, Washington, DC, USA, 2000.

[40] S. E. Byrne and A. J. Rothschild, "Loss of antidepressant efficacy during maintenance therapy: possible mechanisms and treatments," *Journal of Clinical Psychiatry*, vol. 59, no. 6, pp. 279–288, 1998.

[41] M. H. Trivedi, A. J. Rush, S. R. Wisniewski et al., "Evaluation of outcomes with citalopram for depression using measurement-based care in STAR∗D: implications for clinical practice," *American Journal of Psychiatry*, vol. 163, no. 1, pp. 28–40, 2006.

[42] G. Piñeyro and P. Blier, "Autoregulation of serotonin neurons: role in antidepressant drug action," *Pharmacological Reviews*, vol. 51, no. 3, pp. 533–591, 1999.

[43] G. E. Crane, "Iproniazid (marsilid) phosphate, a therapeutic agent for mental disorders and debilitating diseases," *Psychiatric Research Reports*, vol. 8, pp. 142–152, 1957.

[44] I. J. Selikoff, E. H. Robitzek, and G. G. Ornstein, "Treatment of pulmonary tuberculosis with hydrazide derivatives of isonicotinic acid," *Journal of the American Medical Association*, vol. 150, no. 10, pp. 973–980, 1952.

[45] R. KUHN, "The treatment of depressive states with G 22355 (imipramine hydrochloride)," *The American Journal of Psychiatry*, vol. 115, no. 5, pp. 459–464, 1958.

[46] P. Fangmann, H. J. Assion, G. Juckel, C. Á. González, and F. López-Muñoz, "Half a century of antidepressant drugs: on the clinical introduction of monoamine oxidase inhibitors, tricyclics, and tetracyclics. Part II: tricyclics and tetracyclics," *Journal of Clinical Psychopharmacology*, vol. 28, no. 1, pp. 1–4, 2008.

[47] F. López-Muñoz, C. Álamo, G. Juckel, and H. J. Assion, "Half a century of antidepressant drugs: on the clinical introduction of monoamine oxidase inhibitors, tricyclics, and tetracyclics. Part I: monoamine oxidase inhibitors," *Journal of Clinical Psychopharmacology*, vol. 27, no. 6, pp. 555–559, 2007.

[48] R. M. A. Hirschfeld, "History and evolution of the monoamine hypothesis of depression," *Journal of Clinical Psychiatry*, vol. 61, supplement 6, pp. 4–6, 2000.

[49] M. E. Thase, M. H. Trivedi, and A. J. Rush, "MAOIs in the contemporary treatment of depression," *Neuropsychopharmacology*, vol. 12, no. 3, pp. 185–219, 1995.

[50] M. E. Thase, "Effectiveness of antidepressants: comparative remission rates," *Journal of Clinical Psychiatry*, vol. 64, supplement 2, pp. 3–7, 2003.

[51] F. M. Quitkin, J. D. Rabkin, and J. M. Markowitz, "Use of pattern analysis to identify true drug response: a replication," *Archives of General Psychiatry*, vol. 44, no. 3, pp. 259–264, 1987.

[52] I. M. Anderson, D. J. Nutt, and J. F. W. Deakin, "Evidence-based guidelines for treating depressive disorders with antidepressants: a revision of the 1993 British Association for Psychopharmacology guidelines," *Journal of Psychopharmacology*, vol. 14, no. 1, pp. 3–20, 2000.

[53] P. Blier, "The pharmacology of putative early-onset antidepressant strategies," *European Neuropsychopharmacology*, vol. 13, no. 2, pp. 57–66, 2003.

[54] F. M. Quitkin, J. G. Rabkin, and D. Ross, "Identification of true drug response to antidepressants. Use of pattern analysis," *Archives of General Psychiatry*, vol. 41, no. 8, pp. 782–786, 1984.

[55] P. Blier, C. De Montigny, and Y. Chaput, "Modifications of the serotonin system by antidepressant treatments: implications for the therapeutic response in major depression," *Journal of Clinical Psychopharmacology*, vol. 7, supplement 6, pp. 24S–35S, 1987.

[56] S. Stahl, "Is serotonin receptor down-regulation linked to the mechanism of action of antidepressant drugs?" *Psychopharmacology Bulletin*, vol. 30, no. 1, pp. 39–43, 1994.

[57] C. K. J. Lieben, H. W. M. Steinbusch, and A. Blokland, "5,7-DHT lesion of the dorsal raphe nuclei impairs object recognition but not affective behavior and corticosterone response to stressor in the rat," *Behavioural Brain Research*, vol. 168, no. 2, pp. 197–207, 2006.

[58] B. L. Jacobs, H. Van Praag, and F. H. Gage, "Adult brain neurogenesis and psychiatry: a novel theory of depression," *Molecular Psychiatry*, vol. 5, no. 3, pp. 262–269, 2000.

[59] C. Schüle, "Neuroendocrinological mechanisms of actions of antidepressant drugs," *Journal of Neuroendocrinology*, vol. 19, no. 3, pp. 213–226, 2007.

[60] K. J. Parker, A. F. Schatzberg, and D. M. Lyons, "Neuroendocrine aspects of hypercortisolism in major depression," *Hormones and Behavior*, vol. 43, no. 1, pp. 60–66, 2003.

[61] M. Ising, S. Horstmann, S. Kloiber et al., "Combined dexamethasone/corticotropin releasing hormone test predicts treatment response in major depression-a potential biomarker?" *Biological Psychiatry*, vol. 62, no. 1, pp. 47–54, 2007.

[62] M. E. Keck, "Corticotropin-releasing factor, vasopressin and receptor systems in depression and anxiety," *Amino Acids*, vol. 31, no. 3, pp. 241–250, 2006.

[63] B. Vollmayr, M. M. Mahlstedt, and F. A. Henn, "Neurogenesis and depression: what animal models tell us about the link," *European Archives of Psychiatry and Clinical Neuroscience*, vol. 257, no. 5, pp. 300–303, 2007.

[64] D. J. David, B. A. Samuels, Q. Rainer et al., "Neurogenesis-dependent and -independent effects of fluoxetine in an animal model of anxiety/depression," *Neuron*, vol. 62, no. 4, pp. 479–493, 2009.

[65] J. M. Bessa, D. Ferreira, I. Melo et al., "The mood-improving actions of antidepressants do not depend on neurogenesis but are associated with neuronal remodeling," *Molecular Psychiatry*, vol. 14, no. 8, pp. 764–773, 2009.

[66] J. Nacher, M. A. Gomez-Climent, and B. McEwen, "Chronic non-invasive glucocorticoid administration decreases polysialylated neural cell adhesion molecule expression in the adult rat dentate gyrus," *Neuroscience Letters*, vol. 370, no. 1, pp. 40–44, 2004.

[67] P. Willner, "Chronic mild stress (CMS) revisited: consistency and behavioural- neurobiological concordance in the effects of CMS," *Neuropsychobiology*, vol. 52, no. 2, pp. 90–110, 2005.

[68] R. M. Sapolsky, M. J. Meaney, and B. S. McEwen, "The development of the glucocorticoid receptor system in the rat limbic brain. III. Negative-feedback regulation," *Brain Research*, vol. 350, no. 1-2, pp. 169–173, 1985.

[69] R. M. Sapolsky, "Glucocorticoid toxicity in the hippocampus: reversal by supplementation with brain fuels," *Journal of Neuroscience*, vol. 6, no. 8, pp. 2240–2244, 1986.

[70] R. M. Sapolsky, D. R. Packan, and W. W. Vale, "Glucocorticoid toxicity in the hippocampus: in vitro demonstration," *Brain Research*, vol. 453, no. 1-2, pp. 367–371, 1988.

[71] Y. I. Sheline, P. W. Wang, M. H. Gado, J. G. Csernansky, and M. W. Vannier, "Hippocampal atrophy in recurrent major depression," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 93, no. 9, pp. 3908–3913, 1996.

[72] Y. I. Sheline, M. H. Gado, and H. C. Kraemer, "Untreated depression and hippocampal volume loss," *American Journal of Psychiatry*, vol. 160, no. 8, pp. 1516–1518, 2003.

[73] M. C. McKinnon, K. Yucel, A. Nazarov, and G. M. MacQueen, "A meta-analysis examining clinical predictors of hippocampal volume in patients with major depressive disorder," *Journal of Psychiatry and Neuroscience*, vol. 34, no. 1, pp. 41–54, 2009.

[74] J. A. Cobb, J. Simpson, G. J. Mahajan et al., "Hippocampal volume and total cell numbers in major depressive disorder," *Journal of Psychiatric Research*, vol. 47, no. 3, pp. 299–306, 2013.

[75] M. Boldrini, A. N. Santiago, R. Hen et al., "Hippocampal granule neuron number and dentate gyrus volume in antidepressant-treated and untreated major depression," *Neuropsychopharmacology*, 2013.

[76] C. A. Stockmeier, G. J. Mahajan, L. C. Konick et al., "Cellular changes in the postmortem hippocampus in major depression," *Biological Psychiatry*, vol. 56, no. 9, pp. 640–650, 2004.

[77] C. Hercher, G. Turecki, and N. Mechawar, "Through the looking glass: examining neuroanatomical evidence for cellular alterations in major depression," *Journal of Psychiatric Research*, vol. 43, no. 11, pp. 947–961, 2009.

[78] B. Czéh, T. Welt, A. K. Fischer et al., "Chronic psychosocial stress and concomitant repetitive transcranial magnetic stimulation: effects on stress hormone levels and adult hippocampal neurogenesis," *Biological Psychiatry*, vol. 52, no. 11, pp. 1057–1065, 2002.

[79] L. A. M. Galea, B. S. McEwen, P. Tanapat, T. Deak, R. L. Spencer, and F. S. Dhabhar, "Sex differences in dendritic atrophy of CA3 pyramidal neurons in response to chronic restraint stress," *Neuroscience*, vol. 81, no. 3, pp. 689–697, 1997.

[80] Y. Watanabe, E. Gould, and B. S. McEwen, "Stress induces atrophy of apical dendrites of hippocampal CA3 pyramidal neurons," *Brain Research*, vol. 588, no. 2, pp. 341–345, 1992.

[81] T. J. Shors, C. Chua, and J. Falduto, "Sex differences and opposite effects of stress on dendritic spine density in the male versus female hippocampus," *Journal of Neuroscience*, vol. 21, no. 16, pp. 6292–6297, 2001.

[82] H. K. Müller, G. Wegener, M. Popoli, and B. Elfving, "Differential expression of synaptic proteins after chronic restraint stress in rat prefrontal cortex and hippocampus," *Brain Research*, vol. 1385, pp. 26–37, 2011.

[83] B. Czéh, T. Michaelis, T. Watanabe et al., "Stress-induced changes in cerebral metabolites, hippocampal volume, and cell proliferation are prevented by antidepressant treatment with tianeptine," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 98, no. 22, pp. 12796–12801, 2001.

[84] E. Gould, B. S. McEwen, P. Tanapat, L. A. M. Galea, and E. Fuchs, "Neurogenesis in the dentate gyrus of the adult tree shrew is regulated by psychosocial stress and NMDA receptor activation," *Journal of Neuroscience*, vol. 17, no. 7, pp. 2492–2498, 1997.

[85] R. Alonso, G. Griebel, G. Pavone, J. Stemmelin, G. Le Fur, and P. Soubrié, "Blockade of CRF1 or V1b receptors reverses stress-induced suppression of neurogenesis in a mouse model of depression," *Molecular Psychiatry*, vol. 9, no. 3, pp. 278–286, 2004.

[86] J. E. Malberg and R. S. Duman, "Cell proliferation in adult hippocampus is decreased by inescapable stress: reversal by fluoxetine treatment," *Neuropsychopharmacology*, vol. 28, no. 9, pp. 1562–1571, 2003.

[87] M. Banasr, A. Soumier, M. Hery, E. Mocaër, and A. Daszuta, "Agomelatine, a new antidepressant, induces regional changes in hippocampal neurogenesis," *Biological Psychiatry*, vol. 59, no. 11, pp. 1087–1096, 2006.

[88] G. Dagyte, A. Trentani, F. Postema et al., "The novel antidepressant agomelatine normalizes hippocampal neuronal activity and promotes neurogenesis in chronically stressed rats," *CNS Neuroscience and Therapeutics*, vol. 16, no. 4, pp. 195–207, 2010.

[89] T. M. Madsen, A. Treschow, J. Bengzon, T. G. Bolwig, O. Lindvall, and A. Tingström, "Increased neurogenesis in a model of electroconvulsive therapy," *Biological Psychiatry*, vol. 47, no. 12, pp. 1043–1049, 2000.

[90] J. Hellsten, M. Wennström, P. Mohapel, C. T. Ekdahl, J. Bengzon, and A. Tingström, "Electroconvulsive seizures increase hippocampal neurogenesis after chronic corticosterone treatment," *European Journal of Neuroscience*, vol. 16, no. 2, pp. 283–290, 2002.

[91] D. Pagnin, V. De Queiroz, S. Pini, and G. B. Cassano, "Efficacy of ECT in depression: a meta-analytic review," *Journal of ECT*, vol. 20, no. 1, pp. 13–20, 2004.

[92] A. Surget, M. Saxe, S. Leman et al., "Drug-dependent requirement of hippocampal neurogenesis in a model of depression and of antidepressant reversal," *Biological Psychiatry*, vol. 64, no. 4, pp. 293–301, 2008.

[93] K. A. Holick, D. C. Lee, R. Hen, and S. C. Dulawa, "Behavioral effects of chronic fluoxetine in BALB/cJ mice do not require adult hippocampal neurogenesis or the serotonin 1A receptor," *Neuropsychopharmacology*, vol. 33, no. 2, pp. 406–417, 2008.

[94] R. D. Airan, L. A. Meltzer, M. Roy, Y. Gong, H. Chen, and K. Deisseroth, "High-speed imaging reveals neurophysiological links to behavior in an animal model of depression," *Science*, vol. 317, no. 5839, pp. 819–823, 2007.

[95] S. R. Bodnoff, B. Suranyi-Cadotte, R. Quirion, and M. J. Meaney, "A comparison of the effects of diazepam versus several typical and atypical anti-depressant drugs in an animal model of anxiety," *Psychopharmacology*, vol. 97, no. 2, pp. 277–279, 1989.

[96] J. S. Snyder, A. Soumier, M. Brewer, J. Pickel, and H. A. Cameron, "Adult hippocampal neurogenesis buffers stress responses and depressive behaviour," *Nature*, vol. 476, no. 7361, pp. 458–461, 2011.

[97] T. McCall, Z. M. Weil, J. Nacher et al., "Depletion of polysialic acid from neural cell adhesion molecule (PSA-NCAM) increases CA3 dendritic arborization and increases vulnerability to excitotoxicity," *Experimental Neurology*, vol. 241, pp. 5–12, 2013.

[98] T. Seki and U. Rutishauser, "Removal of polysialic acid-neural cell adhesion molecule induces aberrant mossy fiber innervation and ectopic synaptogenesis in the hippocampus," *Journal of Neuroscience*, vol. 18, no. 10, pp. 3757–3766, 1998.

[99] B. Seri, J. M. García-Verdugo, L. Collado-Morente, B. S. McEwen, and A. Alvarez-Buylla, "Cell types, lineage, and architecture of the germinal zone in the adult dentate gyrus," *Journal of Comparative Neurology*, vol. 478, no. 4, pp. 359–378, 2004.

[100] C. M. Ni Dhuill, G. B. Fox, S. J. Pittock, A. W. O'Connell, K. J. Murphy, and C. M. Regan, "Polysialylated neural cell adhesion molecule expression in the dentate gyrus of the human hippocampal formation from infancy to old age," *Journal of Neuroscience Research*, vol. 55, no. 1, pp. 99–106, 1999.

[101] H. G. Kuhn, H. Dickinson-Anson, and F. H. Gage, "Neurogenesis in the dentate gyrus of the adult rat: age-related decrease of neuronal progenitor proliferation," *Journal of Neuroscience*, vol. 16, no. 6, pp. 2027–2033, 1996.

[102] G. B. Fox, N. Kennedy, and C. M. Regan, "Polysialylated neural cell adhesion molecule expression by neurons and astroglial processes in the rat dentate gyrus declines dramatically with increasing age," *International Journal of Developmental Neuroscience*, vol. 13, no. 7, pp. 663–672, 1995.

[103] T. Seki and Y. Arai, "Highly polysialylated neural cell adhesion molecule (NCAM-H) is expressed by newly generated granule

cells in the dentate gyrus of the adult rat," *Journal of Neuroscience*, vol. 13, no. 6, pp. 2351–2358, 1993.

[104] T. Seki and Y. Arai, "The persistent expression of a highly polysialylated NCAM in the dentate gyrus of the adult rat," *Neuroscience Research*, vol. 12, no. 4, pp. 503–513, 1991.

[105] M. I. Cordero, J. J. Rodríguez, H. A. Davies, C. J. Peddie, C. Sandi, and M. G. Stewart, "Chronic restraint stress down-regulates amygdaloid expression of polysialylated neural cell adhesion molecule," *Neuroscience*, vol. 133, no. 4, pp. 903–910, 2005.

[106] J. Nacher, K. Pham, V. Gil-Fernandez, and B. S. Mcewen, "Chronic restraint stress and chronic corticosterone treatment modulate differentially the expression of molecules related to structural plasticity in the adult rat piriform cortex," *Neuroscience*, vol. 126, no. 2, pp. 503–509, 2004.

[107] K. Pham, J. Nacher, P. R. Hof, and B. S. McEwen, "Repeated restraint stress suppresses neurogenesis and induces biphasic PSA-NCAM expression in the adult rat dentate gyrus," *European Journal of Neuroscience*, vol. 17, no. 4, pp. 879–886, 2003.

[108] C. Sandi, J. J. Merino, M. I. Cordero, K. Touyarot, and C. Venero, "Effects of chronic stress on contextual fear conditioning and the hippocampal expression of the neural cell adhesion molecule, its polysialylation, and L1," *Neuroscience*, vol. 102, no. 2, pp. 329–339, 2001.

[109] A. Djordjevic, J. Djordjevic, I. Elakovic, M. Adzic, G. Matic, and M. B. Radojcic, "Fluoxetine affects hippocampal plasticity, apoptosis and depressive-like behavior of chronically isolated rats," *Prog Neuropsychopharmacol Biol Psychiatry*, vol. 36, no. 1, pp. 92–100, 2012.

[110] J. Gilabert-Juan, E. Castillo-Gomez, R. Guirado, M. D. Molto, and J. Nacher, "Chronic stress alters inhibitory networks in the medial prefrontal cortex of adult mice," *Brain Structure and Function*, 2012.

[111] J. Djordjevic, A. Djordjevic, M. Adzic, and M. B. Radojcic, "Effects of chronic social isolation on Wistar rat behavior and brain plasticity markers," *Neuropsychobiology*, vol. 66, no. 2, pp. 112–119, 2012.

[112] A. Djordjevic, J. Djordjevic, I. Elakovic, M. Adzic, G. Matic, and M. B. Radojcic, "Effects of fluoxetine on plasticity and apoptosis evoked by chronic stress in rat prefrontal cortex," *European Journal of Pharmacology*, vol. 693, no. 1–3, pp. 37–44, 2012.

[113] J. Gilabert-Juan, E. Castillo-Gomeza, M. Pérez-Randoa, M. Dolores Molt*a*b, and J. Nachera, "Chronic stress induces changes in the structure of interneurons and in the expression of molecules related to neuronal structural plasticity and inhibitory neurotransmission in the amygdala of adult mice," *Experimental Neurology*, vol. 232, no. 1, pp. 33–40, 2011.

[114] M. E. Maheu, M. A. Davoli, G. Turecki, and N. Mechawar, "Amygdalar expression of proteins associated with neuroplasticity in major depression and suicide," *Journal of Psychiatric Research*, vol. 47, no. 3, pp. 384–390, 2013.

[115] J. Gilabert-Juan, E. Varea, R. Guirado, J. M. Blasco-Ibanez, C. Crespo, and J. Nacher, "Alterations in the expression of PSA-NCAM and synaptic proteins in the dorsolateral prefrontal cortex of psychiatric disorder patients," *Neuroscience Letters*, vol. 530, no. 1, pp. 97–102, 2012.

[116] J. R. Homberg, J. D. A. Olivier, T. Blom et al., "Fluoxetine exerts age-dependent effects on behavior and amygdala neuroplasticity in the rat," *PLoS ONE*, vol. 6, no. 1, Article ID e16646, 2011.

[117] E. Varea, E. Castillo-Gómez, M. A. Gómez-Climent et al., "Chronic antidepressant treatment induces contrasting patterns of synaptophysin and PSA-NCAM expression in different regions of the adult rat telencephalon," *European Neuropsychopharmacology*, vol. 17, no. 8, pp. 546–557, 2007.

[118] M. Sairanen, O. F. O'Leary, J. E. Knuuttila, and E. Castrén, "Chronic antidepressant treatment selectively increases expression of plasticity-related proteins in the hippocampus and medial prefrontal cortex of the rat," *Neuroscience*, vol. 144, no. 1, pp. 368–374, 2007.

[119] E. Varea, J. M. Blasco-Ibáñez, M. Á. Gómez-Climent et al., "Chronic fluoxetine treatment increases the expression of PSA-NCAM in the medial prefrontal cortex," *Neuropsychopharmacology*, vol. 32, no. 4, pp. 803–812, 2007.

[120] J. J. Rodríguez, G. M. Dallérac, M. Tabuchi et al., "N-methyl-d-aspartate receptor independent changes in expression of polysialic acid-neural cell adhesion molecule despite blockade of homosynaptic long-term potentiation and heterosynaptic long-term depression in the awake freely behaving rat dentate gyrus," *Neuron Glia Biology*, vol. 4, no. 3, pp. 169–178, 2008.

[121] G. Guiraudie-Capraz, F. A. Chaillan, B. Truchet, J. L. Franc, C. Mourre, and F. S. Roman, "Increase in polysialyltransferase gene expression following LTP in adult rat dentate gyrus," *Hippocampus*, vol. 21, no. 11, pp. 1180–1189, 2011.

[122] D. Muller, P. Mendez, M. DeRoo, P. Klauser, S. Steen, and L. Poglia, "Role of NCAM in Spine dynamics and synaptogenesis," *Advances in Experimental Medicine and Biology*, vol. 663, pp. 245–256, 2010.

[123] M. Grzegorzewska, M. Maćkowiak, K. Wedzony, and G. Hess, "5-HT1A receptors mediate detrimental effects of cocaine on long-term potentiation and expression of polysialylated neural cell adhesion molecule protein in rat dentate gyrus," *Neuroscience*, vol. 166, no. 1, pp. 122–131, 2010.

[124] J. M. Brezun and A. Daszuta, "Serotonergic reinnervation reverses lesion-induced decreases in PSA-NCAM labeling and proliferation of hippocampal cells in adult rats," *Hippocampus*, vol. 10, no. 1, pp. 37–46, 2000.

[125] F. Karege, G. Perret, G. Bondolfi, M. Schwald, G. Bertschy, and J. M. Aubry, "Decreased serum brain-derived neurotrophic factor levels in major depressed patients," *Psychiatry Research*, vol. 109, no. 2, pp. 143–148, 2002.

[126] E. Shimizu, K. Hashimoto, N. Okamura et al., "Alterations of serum levels of brain-derived neurotrophic factor (BDNF) in depressed patients with or without antidepressants," *Biological Psychiatry*, vol. 54, no. 1, pp. 70–75, 2003.

[127] O. Aydemir, A. Deveci, and F. Taneli, "The effect of chronic antidepressant treatment on serum brain-derived neurotrophic factor levels in depressed patients: a preliminary study," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 29, no. 2, pp. 261–265, 2005.

[128] Y. Shirayama, A. C. H. Chen, S. Nakagawa, D. S. Russell, and R. S. Duman, "Brain-derived neurotrophic factor produces antidepressant effects in behavioral models of depression," *Journal of Neuroscience*, vol. 22, no. 8, pp. 3251–3261, 2002.

[129] J. Grønli, C. Bramham, R. Murison et al., "Chronic mild stress inhibits BDNF protein expression and CREB activation in the dentate gyrus but not in the hippocampus proper," *Pharmacology Biochemistry and Behavior*, vol. 85, no. 4, pp. 842–849, 2006.

[130] C. A. Altar, R. E. Whitehead, R. Chen, G. Wörtwein, and T. M. Madsen, "Effects of electroconvulsive seizures and antidepressant drugs on brain-derived neurotrophic factor protein in rat brain," *Biological Psychiatry*, vol. 54, no. 7, pp. 703–709, 2003.

[131] J. Vinet, S. Carra, J. M. C. Blom, N. Brunello, N. Barden, and F. Tascedda, "Chronic treatment with desipramine and fluoxetine

modulate BDNF, CaMKKα and CaMKKβ mRNA levels in the hippocampus of transgenic mice expressing antisense RNA against the glucocorticoid receptor," *Neuropharmacology*, vol. 47, no. 7, pp. 1062–1069, 2004.

[132] A. Russo-Neustadt, R. C. Beard, and C. W. Cotman, "Exercise, antidepressant medications, and enhanced brain derived neurotrophic factor expression," *Neuropsychopharmacology*, vol. 21, no. 5, pp. 679–682, 1999.

[133] J. A. Siuciak, D. R. Lewis, S. J. Wiegand, and R. M. Lindsay, "Antidepressant-like effect of brain-derived neurotrophic factor (BDNF)," *Pharmacology Biochemistry and Behavior*, vol. 56, no. 1, pp. 131–137, 1996.

[134] S. C. Danzer, K. R. C. Crooks, D. C. Lo, and J. O. McNamara, "Increased expression of brain-derived neurotrophic factor induces formation of basal dendrites and axonal branching in dentate granule cells in hippocampal explant cultures," *Journal of Neuroscience*, vol. 22, no. 22, pp. 9754–9763, 2002.

[135] F. J. Seil and R. Drake-Baumann, "TrkB receptor ligands promote activity-dependent inhibitory synaptogenesis," *Journal of Neuroscience*, vol. 20, no. 14, pp. 5367–5373, 2000.

[136] Y. Li, B. W. Luikart, S. Birnbaum et al., "TrkB regulates Hippocampal neurogenesis and governs sensitivity to antidepressive treatment," *Neuron*, vol. 59, no. 3, pp. 399–412, 2008.

[137] E. Castrén and T. Rantamäki, "The role of BDNF and its receptors in depression and antidepressant drug action: reactivation of developmental plasticity," *Developmental Neurobiology*, vol. 70, no. 5, pp. 289–297, 2010.

[138] B. Zörner, D. P. Wolfer, D. Brandis et al., "Forebrain-specific trkB-receptor knockout mice: behaviorally more hyperactive than 'depressive'," *Biological Psychiatry*, vol. 54, no. 10, pp. 972–982, 2003.

[139] T. Saarelainen, P. Hendolin, G. Lucas et al., "Activation of the TrkB neurotrophin receptor is induced by antidepressant drugs and is required for antidepressant-induced behavioral effects," *Journal of Neuroscience*, vol. 23, no. 1, pp. 349–357, 2003.

[140] L. M. Monteggia, B. Luikart, M. Barrot et al., "Brain-derived neurotrophic factor conditional knockouts show gender differences in depression-related behaviors," *Biological Psychiatry*, vol. 61, no. 2, pp. 187–197, 2007.

[141] D. Muller, Z. Djebbara-Hannas, P. Jourdain et al., "Brain-derived neurotrophic factor restores long-term potentiation in polysialic acid-neural cell adhesion molecule-deficient hippocampus," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 8, pp. 4315–4320, 2000.

[142] L. Vutskits, Z. Djebbara-Hannas, H. Zhang et al., "PSA-NCAM modulates BDNF-dependent survival and differentiation of cortical neurons," *European Journal of Neuroscience*, vol. 13, no. 7, pp. 1391–1402, 2001.

[143] A. Burgess and I. Aubert, "Polysialic acid limits choline acetyltransferase activity induced by brain-derived neurotrophic factor," *Journal of Neurochemistry*, vol. 99, no. 3, pp. 797–806, 2006.

[144] E. Gascon, L. Vutskits, B. Jenny, P. Durbec, and J. Z. Kiss, "PSA-NCAM in postnatally generated immature neurons of the olfactory bulb: a crucial role in regulating p75 expression and cell survival," *Development*, vol. 134, no. 6, pp. 1181–1190, 2007.

[145] K. M. Young, T. D. Merson, A. Sotthibundhu, E. J. Coulson, and P. F. Bartlett, "p75 neurotrophin receptor expression defines a population of BDNF-responsive neurogenic precursor cells," *Journal of Neuroscience*, vol. 27, no. 19, pp. 5146–5155, 2007.

[146] V. S. Catts, N. Al-Menhali, T. H. J. Burne, M. J. Colditz, and E. J. Coulson, "The p75 neurotrophin receptor regulates hippocampal neurogenesis and related behaviours," *European Journal of Neuroscience*, vol. 28, no. 5, pp. 883–892, 2008.

[147] M. J. Colditz, V. S. Catts, N. Al-Menhali, G. W. Osborne, P. F. Bartlett, and E. J. Coulson, "P75 neurotrophin receptor regulates basal and fluoxetine-stimulated hippocampal neurogenesis," *Experimental Brain Research*, vol. 200, no. 2, pp. 161–167, 2010.

[148] K. Gutiérrez-Lobos, M. Scherer, P. Anderer, and H. Katschnig, "The influence of age on the female/male ratio of treated incidence rates in depression," *BMC Psychiatry*, vol. 2, no. 1, p. 3, 2002.

[149] S. G. Kornstein, A. F. Schatzberg, M. E. Thase et al., "Gender differences in treatment response to sertraline versus imipramine in chronic depression," *American Journal of Psychiatry*, vol. 157, no. 9, pp. 1445–1452, 2000.

[150] M. M. Shores, K. L. Sloan, A. M. Matsumoto, V. M. Moceri, B. Felker, and D. R. Kivlahan, "Increased incidence of diagnosed depressive illness in hypogonadal older men," *Archives of General Psychiatry*, vol. 61, no. 2, pp. 162–167, 2004.

[151] M. M. Shores, V. M. Moceri, K. L. Sloan, A. M. Matsumoto, and D. R. Kivlahan, "Low testosterone levels predict incident depressive illness in older men: effects of age and medical morbidity," *The Journal of clinical psychiatry*, vol. 66, no. 1, pp. 7–14, 2005.

[152] M. M. Shores, D. R. Kivlahan, T. I. Sadak, E. J. Li, and A. M. Matsumoto, "A randomized, double-blind, placebo-controlled study of testosterone treatment in hypogonadal older men with subthreshold depression (dysthymia or minor depression)," *Journal of Clinical Psychiatry*, vol. 70, no. 7, pp. 1009–1016, 2009.

[153] R. S. McIntyre, D. Mancini, B. S. Eisfeld et al., "Calculated bioavailable testosterone levels and depression in middle-aged men," *Psychoneuroendocrinology*, vol. 31, no. 9, pp. 1029–1035, 2006.

[154] A. B. Veras and A. E. Nardi, "The complex relationship between hypogonadism and major depression in a young male," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 34, no. 2, pp. 421–422, 2010.

[155] S. N. Seidman and J. G. Rabkin, "Testosterone replacement therapy for hypogonadal men with SSRI- refractory depression," *Journal of Affective Disorders*, vol. 48, no. 2-3, pp. 157–161, 1998.

[156] F. A. Zarrouf, S. Artz, J. Griffith, C. Sirbu, and M. Kommor, "Testosterone and depression: systematic review and meta-analysis," *Journal of Psychiatric Practice*, vol. 15, no. 4, pp. 289–305, 2009.

[157] H. G. Pope Jr., G. H. Cohane, G. Kanayama, A. J. Siegel, and J. I. Hudson, "Testosterone gel supplementation for men with refractory depression: a randomized, placebo-controlled trial," *American Journal of Psychiatry*, vol. 160, no. 1, pp. 105–111, 2003.

[158] S. N. Seidman, E. Spatz, C. Rizzo, and S. P. Roose, "Testosterone replacement therapy for hypogonadal men with major depressive disorder: a randomized, placebo-controlled clinical trial," *Journal of Clinical Psychiatry*, vol. 62, no. 6, pp. 406–412, 2001.

[159] H. G. Pope Jr., R. Amiaz, B. P. Brennan et al., "Parallel-group placebo-controlled trial of testosterone gel in men with major depressive disorder displaying an incomplete response to standard antidepressant treatment," *Journal of Clinical Psychopharmacology*, vol. 30, no. 2, pp. 126–134, 2010.

[160] V. Hendrick, L. L. Altshuler, and R. Suri, "Hormonal changes in the postpartum and implications for postpartum depression," *Psychosomatics*, vol. 39, no. 2, pp. 93–101, 1998.

[161] L. S. Cohen, C. N. Soares, A. F. Vitonis, M. W. Otto, and B. L. Harlow, "Risk for new onset of depression during the menopausal transition: the harvard study of moods and cycles," *Archives of General Psychiatry*, vol. 63, no. 4, pp. 385–390, 2006.

[162] N. L. Rasgon, L. L. Altshuler, L. A. Fairbanks et al., "Estrogen replacement therapy in the treatment of major depressive disorder in perimenopausal women," *Journal of Clinical Psychiatry*, vol. 63, supplement 7, pp. 45–48, 2002.

[163] C. De Novaes Soares, O. P. Almeida, H. Joffe, and L. S. Cohen, "Efficacy of estradiol for the treatment of depressive disorders in perimenopausal women: a double-blind, randomized, placebo-controlled trial," *Archives of General Psychiatry*, vol. 58, no. 6, pp. 529–534, 2001.

[164] C. N. Soares, J. R. Poitras, J. Prouty, A. B. Alexander, J. L. Shifren, and L. S. Cohen, "Efficacy of citalopram as a monotherapy or as an adjunctive treatment to estrogen therapy for perimenopausal and postmenopausal women with depression and vasomotor symptoms," *Journal of Clinical Psychiatry*, vol. 64, no. 4, pp. 473–479, 2003.

[165] A. D. Green and L. A. M. Galea, "Adult hippocampal cell proliferation is suppressed with estrogen withdrawal after a hormone-simulated pregnancy," *Hormones and Behavior*, vol. 54, no. 1, pp. 203–211, 2008.

[166] S. L. Sell, R. M. Craft, P. K. Seitz, S. J. Stutz, K. A. Cunningham, and M. L. Thomas, "Estradiol-sertraline synergy in ovariectomized rats," *Psychoneuroendocrinology*, vol. 33, no. 8, pp. 1051–1060, 2008.

[167] N. Carrier and M. Kabbaj, "Testosterone and imipramine have antidepressant effects in socially isolated male but not female rats," *Hormones and Behavior*, vol. 61, no. 5, pp. 678–685, 2012.

[168] M. D. Spritzer and L. A. M. Galea, "Testosterone and dihydrotestosterone, but not estradiol, enhance survival of new hippocampal neurons in adult male rats," *Developmental Neurobiology*, vol. 67, no. 10, pp. 1321–1333, 2007.

[169] L. A. M. Galea, M. D. Spritzer, J. M. Barker, and J. L. Pawluski, "Gonadal hormone modulation of hippocampal neurogenesis in the adult," *Hippocampus*, vol. 16, no. 3, pp. 225–232, 2006.

[170] O. Tan, A. Fadiel, A. Chang et al., "Estrogens regulate posttranslational modification of neural cell adhesion molecule during the estrogen-induced gonadotropin surge," *Endocrinology*, vol. 150, no. 6, pp. 2783–2790, 2009.

[171] Y. Hatanaka, H. Mukai, K. Mitsuhashi et al., "Androgen rapidly increases dendritic thorns of CA3 neurons in male rat hippocampus," *Biochemical and Biophysical Research Communications*, vol. 381, no. 4, pp. 728–732, 2009.

[172] N. J. MacLusky, T. Hajszan, J. A. Johansen, C. L. Jordan, and C. Leranth, "Androgen effects on hippocampal CA1 spine synapse numbers are retained in Tfm male rats with defective androgen receptors," *Endocrinology*, vol. 147, no. 5, pp. 2392–2398, 2006.

[173] S. Rasika, A. Alvarez-Buylla, and F. Nottebohm, "BDNF mediates the effects of testosterone on the survival of new neurons in an adult brain," *Neuron*, vol. 22, no. 1, pp. 53–62, 1999.

[174] L. Y. Yang, T. Verhovshek, and D. R. Sengelaub, "Brain-derived neurotrophic factor and androgen interact in the maintenance of dendritic morphology in a sexually dimorphic rat spinal nucleus," *Endocrinology*, vol. 145, no. 1, pp. 161–168, 2004.

[175] D. T. Solum and R. J. Handa, "Estrogen regulates the development of Brain-derived neurotrophic factor mRNA and protein in the rat hippocampus," *Journal of Neuroscience*, vol. 22, no. 7, pp. 2650–2659, 2002.

[176] T. B. Franklin and T. S. Perrot-Sinal, "Sex and ovarian steroids modulate brain-derived neurotrophic factor (BDNF) protein levels in rat hippocampus under stressful and non-stressful conditions," *Psychoneuroendocrinology*, vol. 31, no. 1, pp. 38–48, 2006.

[177] F. Sohrabji, R. C. G. Miranda, and C. D. Toran-Allerand, "Identification of a putative estrogen response element in the gene encoding brain-derived neurotrophic factor," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 92, no. 24, pp. 11110–11114, 1995.

[178] D. D. Murphy, N. B. Cole, and M. Segal, "Brain-derived neurotrophic factor mediates estradiol-induced dendritic spine formation in hippocampal neurons," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 95, no. 19, pp. 11412–11417, 1998.

[179] Y. Dimberg, M. Vazquez, S. Söderström, and T. Ebendal, "Effects of X-irradiation on nerve growth factor in the developing mouse brain," *Toxicology Letters*, vol. 90, no. 1, pp. 35–43, 1997.

[180] G. Casadesus, B. Shukitt-Hale, H. M. Stellwagen, M. A. Smith, B. M. Rabin, and J. A. Joseph, "Hippocampal neurogenesis and PSA-NCAM expression following exposure to 56Fe particles mimics that seen during aging in rats," *Experimental Gerontology*, vol. 40, no. 3, pp. 249–254, 2005.

[181] J. S. Snyder, J. S. Choe, M. A. Clifford et al., "Adult-born hippocampal neurons are more numerous, faster maturing, and more involved in behavior in rats than in mice," *Journal of Neuroscience*, vol. 29, no. 46, pp. 14484–14495, 2009.

[182] W. N. Marsden, "Synaptic plasticity in depression: molecular, cellular and functional correlates," *Progress in Neuro-Psychopharmacology Biological Psychiatry C*, vol. 43, pp. 168–184, 2012.

[183] C. Sandi and R. Bisaz, "A model for the involvement of neural cell adhesion molecules in stress-related mood disorders," *Neuroendocrinology*, vol. 85, no. 3, pp. 158–176, 2007.

Hindawi Publishing Corporation
Neural Plasticity
Volume 2012, Article ID 854285, 20 pages
doi:10.1155/2012/854285

Review Article

Functional Role of Adult Hippocampal Neurogenesis as a Therapeutic Strategy for Mental Disorders

Heechul Jun,[1] Syed Mohammed Qasim Hussaini,[1] Michael J. Rigby,[2] and Mi-Hyeon Jang[1,3]

[1] *Department of Neurologic Surgery, Mayo College of Medicine, 200 First Street SW, Rochester, MN 55905, USA*
[2] *Summer Undergraduate Research Fellowship Program, Mayo Graduate School, Mayo College of Medicine, 200 First Street SW, Rochester, MN 55905, USA*
[3] *Department of Biochemistry and Molecular Biology, Mayo College of Medicine, 200 First Street SW, Rochester, MN 55905, USA*

Correspondence should be addressed to Mi-Hyeon Jang, jang.mihyeon@mayo.edu

Received 15 September 2012; Revised 30 November 2012; Accepted 30 November 2012

Academic Editor: Chitra D. Mandyam

Adult neurogenesis, the process of generating new neurons from neural stem cells, plays significant roles in synaptic plasticity, memory, and mood regulation. In the mammalian brain, it continues to occur well into adulthood in discrete regions, namely, the hippocampus and olfactory bulb. During the past decade, significant progress has been made in understanding the mechanisms regulating adult hippocampal neurogenesis and its role in the etiology of mental disorders. In addition, adult hippocampal neurogenesis is highly correlated with the remission of the antidepressant effect. In this paper, we discuss three major psychiatric disorders, depression, schizophrenia, and drug addiction, in light of preclinical evidence used in establishing the neurobiological significance of adult neurogenesis. We interpret the significance of these results and pose questions that remain unanswered. Potential treatments which include electroconvulsive therapy, deep brain stimulation, chemical antidepressants, and exercise therapy are discussed. While consensus lacks on specific mechanisms, we highlight evidence which indicates that these treatments may function via an increase in neural progenitor proliferation and changes to the hippocampal circuitry. Establishing a significant role of adult neurogenesis in the pathogenicity of psychiatric disorders may hold the key to potential strategies toward effective treatment.

1. Introduction

Mental disorders are debilitating conditions that significantly impair the function of the central nervous system and degrade the quality of life. About one-quarter of adult Americans are diagnosed with mental disorders such as major depression, anxiety, and schizophrenia [1]. Understanding the neurobiological basis of mental disorders, determining effective treatments, and alleviating the respective symptoms are major forces driving modern psychiatry today.

The hippocampus, an area of the brain important in memory, cognitive function, and mood regulation, is particularly vulnerable to chronic stress and mental disorders [2, 3]. Several landmark clinical studies have demonstrated that major depression is accompanied by a decrease in the volume of hippocampus and consequent deficits in hippocampal function [4, 5]. Similarly, in schizophrenic patients, shape deformations, cell loss, and volume reduction in the hippocampus were found using neuroimaging analysis [6–8]. Reversal of these alterations has successfully improved the behavioral and cognitive symptoms associated with these disorders. Such evidence has encouraged consideration of whether improving hippocampal structure and function could be a potential therapeutic target in treating mental disorders [9, 10].

Since the pioneering discovery of mammalian postnatal neurogenesis in the 1960s [58], adult neurogenesis has been unambiguously investigated in discrete brain regions across mammals including humans. Adult neurogenesis in all mammals, including humans, occurs throughout life within two specialized neurogenic niches, the subventricular zone (SVZ) of the lateral ventricle and the subgranular zone (SGZ) of the hippocampal dentate gyrus [59]. In particular, adult neurogenesis in dentate gyrus has attracted

interest since newborn neurons contribute to enhanced neural plasticity that could sustain specific brain functions such as spatial learning, pattern discrimination, and mood regulation [47, 60, 61]. In addition, adult hippocampal neurogenesis in the mature brain represents a striking example of activity-dependent neural plasticity such as stress, antidepressants, and brain injuries [62]. Extensive studies have shown that voluntary exercise, enriched environments, and antidepressants contribute to overall brain health by robustly promoting adult hippocampal neurogenesis [37, 53, 63]. Decreased neurogenesis in the hippocampus via aging or stress has been implicated in the pathogenesis of cognitive deficits, anxiety and depression [64, 65]. In fact, adult hippocampal neurogenesis not only plays an important role in antidepressant action [47] but also plays a role in ameliorating various pathological disease conditions [25, 46, 66, 67]. Therefore, a better understanding of the molecular and cellular mechanisms that regulate adult hippocampal neurogenesis may offer new therapeutic targets.

In this paper, we will highlight three major psychiatric disorders that have been associated with adult hippocampal neurogenesis. We will present and interpret the significance of the results in regards to the mechanism of cognitive and neurological disorders. Finally, we will lay out some current and potential therapeutic treatments that are used to counter these psychiatric disorders.

2. Adult Neurogenesis and Mental Disorders

2.1. Depression. Major depression is among the most prevalent psychiatric disorders and has high morbidity worldwide. Chronic stress represents a key risk factor in developing depression [68–70]. Despite a tremendous amount of study, the underlying mechanisms associated with the pathophysiology of depression remain poorly understood. The neurogenesis hypothesis of depression originated from studies using animal stress models. Because of the lack of a pathophysiologically reliable animal depression model, stress is primarily used to cause depression in animal models. These studies demonstrate that stress inhibits newborn neuron proliferation in the dentate gyrus of the hippocampus [71, 72]. All antidepressant classes are now known to promote adult hippocampal neurogenesis [38, 47]. The underlying cellular and molecular mechanisms of adult neurogenesis in regulating the suppressive effect of stress have been examined using various stress paradigms including physical and psychosocial stresses (Table 1). For example, physical stress such as repeated restraint [11] and inescapable foot shock [12] inhibits one or more steps of adult neurogenesis in the dentate gyrus. Similarly, chronic psychosocial stress using the social defeat paradigm leads to an inhibitory effect on cell proliferation and survival of newborn granule neurons in rodents [17]. In addition, the effect of depression on hippocampal volume in humans has been studied [4, 5, 73–75]. Early onset depression has been associated with a reduction in hippocampal volume [75], and in patients older than 60, depression has been associated specifically with a reduction in right hippocampal volume [74]. Further, hippocampal grey matter which undergoes reduction in depressed patients could be increased through antidepressive treatment like citalopram [73]. While such changes have been observed, postmortem studies have found no change in cell proliferation between major depression patients and control samples [76, 77]. Intriguingly, antidepressant treatment does increase cell proliferation in the dentate gyrus of major depression patients [77]. Such differential effects between rodents and humans on cell proliferation in the dentate gyrus might be due to biological differences implying different cellular mechanisms [78]. Determining the processes of adult neurogenesis in humans, such as fate determination, survival, differentiation, and integration across both spectrums will be helpful in reconciling the differences.

Preclinical models of depression generally show one or more suppressed steps during the sequential adult neurogenesis process. Such evidence has raised questions regarding the causality between neurogenesis and depression. Using a variety of methods ablating newborn neurons in the adult dentate gyrus, evidence correlates adult hippocampal neurogenesis with depression. However, the results are controversial [47, 48, 76, 79]. For example, ablating adult hippocampal neurogenesis using X-ray irradiation does not affect anxiety- and depressive-like behaviors as measured by novelty-suppressed feeding, open-field, light-dark, and elevated plus maze [47, 48, 79]. Further, pharmacological treatment with an anti-mitotic drug, methylazoxymethanol (MAM) which decreases cell proliferation in the dentate gyrus, does not induce an anhedonic-like state in rats [80]. On the contrary, genetically inhibiting hippocampal neurogenesis using Nestin-rtTA/Tet-Bax bigenic mice does increase anxiety-related behavior but does not affect depressive-like behavior [81]. Also, deletion of adult neurogenesis using GFAP-TK mice influences depressive-like behavioral response as shown by increased immobility time in the tail suspension test and induction of an anhedonic-like response in sucrose-preference test, but shows no effect in novelty-suppressed feeding or elevated plus maze [82]. This group further suggests that suppression of neurogenesis predisposes the animal to stress-induced anxiety/depression-like behavior response and that newborn neurons in the hippocampus buffer this depressive-like behavior [82]. At this juncture, the results do not quite support that decreased neurogenesis is associated with a risk factor in development of depressive behavior. It may be more pertinent to conclude that newborn neurons may be a major contributor in normalizing or ameliorating against disease state rather than being causally involved in the etiology of depression in the animal model.

2.2. Schizophrenia. Schizophrenia is a multifactorial psychiatric disorder resulting from a complex interplay of genetic and environmental susceptibility factors [3]. Establishing the major etiology, neuropathology and pathophysiology of schizophrenia have proven difficult.

Initial studies from postmortem hippocampal tissue have indicated a reduction in neural progenitors and hippocampal volume in patients diagnosed with schizophrenia

Table 1: Select animal studies investigating the effect of depression on adult hippocampal neurogenesis and behavior.

Species	Stress paradigm	Experimental approach	Effects on neurogenesis	Effects on behavior	References
		Physical stress			
Sprague-Dawley rat	Acute restraint stress: either 2 or 6 hours. Chronic restraint stress: 6 hours daily for 21 days. Repeated restraint stress: 6 hours daily for 42 days.	Proliferation: BrdU (1 × 200 mg/kg, ip) at 2 hours pulsing chase Survival: BrdU (100 mg/kg, ip) daily for 4 days and analysis on 12 days to 3 weeks periods after BrdU injection.	Acute stress: no effect on cell proliferation. Chronic stress: Decreased cell proliferation, slightly decreased survival rate with no statistical significance. Repeated stress: decreased survival rate.	Not studied (NS)	[11]
Sprague-Dawley rat	60 inescapable foot shocks	Proliferation: BrdU (1 × 100 mg/kg, ip) at 2 hours pulsing chase. Survival: BrdU (100 mg/kg, ip) on day 9 and analysis 28 days later.	Decreased cell proliferation. No change on survival rate and differentiation.	NS	[12]
ICR mouse	Chronic restraint stress: 6 hours per day for four weeks.	Proliferation: Ki67 immunostaining. Survival: BrdU (3 × 75 mg/kg, ip) at 6-hour intervals and 24 hours before exposure to chronic restraint stress.	Decreased cell proliferation and survival rate.	Impaired context dependent memory.	[13]
Wistar rat	Olfactory bulbectomy (OB).	Proliferation: BrdU (1 × 40 mg/kg, ip) at 2 hours pulsing chase. Survival: BrdU (3 × 200 mg/kg, ip) at 2-hour intervals and analysis 4 weeks after last administration.	Decreased cell proliferation and neuronal differentiation.	Impaired context-dependent memory.	[14]
		Social stress			
Sprague-Dawley rat	Psychosocial stress by introducing intruder animal.	Proliferation: IdU (4 × 57.5 mg/kg, ip) at 12-hour intervals. IdU at 3-day pulsing chase after the first administration. Short-term survival: BrdU (6 × 50 mg/kg, ip), 12-hour intervals. BrdU at 10-day pulsing chase after first administration.	No change on cell proliferation, but decreased short-term survival rate.	NS	[15]
Prairie vole	Social isolation: single-housed for 6 weeks.	Proliferation: Ki67 immunostaining. Survival: BrdU (100 mg/kg, ip) daily for fourteen consecutive days and then 6-week pulsing chase after last administration.	Decreased cell proliferation and neuronal maturation.	Higher levels of anxiety-like and depression-like behaviors	[16]
Wistar rat	Social defeat stress daily for 18 days	Proliferation: BrdU (200 mg/kg, ip) at 24-hour pulsing chase. Survival: BrdU (200 mg/kg, ip) two times per day for 2 days.	Decreased cell proliferation and survival rate.	NS	[17]

[76]. Supporting this, a growing number of studies have identified susceptibility genes associated with schizophrenia that are involved in regulation of adult neurogenesis [22, 83] (Table 2). One such gene is disrupted-in-schizophrenia 1 (DISC1) which was originally identified as a potential susceptibility gene for schizophrenia and related psychiatric disorders in a large Scottish pedigree [84, 85]. It is widely expressed during embryonic neurogenesis and postnatal development with high expression persisting in the adult hippocampus, especially in the dentate gyrus [86, 87]. The expression pattern of DISC1 may implicate a role of DISC1 in neuronal development. Using various approaches including genetic mutants and short-hairpin RNA (shRNA) knockdown, several lines of evidence have converged to indicate that DISC1 function is involved in distinct steps of adult neurogenesis and behavioral response. First, mutant DISC1 mice with selective impairment in working memory showed reduced number of neural progenitors and immature neurons, as well as misoriented apical dendrites of immature neurons in the adult hippocampal dentate gyrus [21]. In the same DISC1 mutant mice, altered axonal targeting and short-term plasticity in the hippocampus were also observed [21]. Second, knockdown of DISC1 via lentiviral-mediated shRNA also led to decreased newborn cells and a depressive-like behavioral response [22]. Third, knockdown of DISC1 via retroviral-mediated shRNA approach to label dividing cells and their progeny results in aberrant dendritic development including an enhancement of dendritic outgrowth, soma hypertrophy, overextended migration, accelerated synapse formation, mistargeted axonal mossy fibers, and presynaptic differentiation of newborn granule neurons in the adult mouse dentate gyrus [18, 19]. Further, the same group also indicated that AKT-mTOR signaling pathway is a critical DISC1 target in regulating dendritic development of newborn granule neurons in the adult dentate gyrus [88]. Taken together, these results suggest that DISC1 orchestrates the tempo of functional integration in the adult brain. It also demonstrates the major roles a susceptibility gene could play in neuronal development and the pathogenesis of the disease.

Other susceptibility genes for schizophrenia linked to adult neurogenesis have been identified. NPAS, a bHLH transcription factor that is broadly expressed in the developing neuroepithelium, was identified as a risk factor for schizophrenia and associated with cognitive deficits in an affected mother and her daughter [89, 90]. Mice lacking *Npas3* show developmental brain abnormalities including a reduction in size of the anterior hippocampus, hypoplasia of the corpus callosum and enlargement of the ventricles [23]. In addition, mice lacking *Npas3* exhibit behavioral abnormalities including locomotor hyperactivity, subtle gait defects, impairment of prepulse inhibition of acoustic startle, deficit in recognition memory, and altered anxiety-related responses [23]. Finally, Npas3-deficent mice also display a significant reduction of adult neurogenesis by 84% relative to their wild-type littermates, which may suggest that adult neurogenesis impairment is involved in the pathogenesis of schizophrenia [22]. However, direct causal evidence linking adult neurogenesis to schizophrenia needs to be examined.

In addition to using genetic animal models for schizophrenia, a prenatal injection of synthetic double-stranded RNA polyriboinosinic polyribocytidylic acid (PolyI:C) into mice has been used as an animal model for schizophrenia [26, 91]. These mice displayed behavioral deficits in the open-field test and prepulse inhibition of the startle response [26]. A recent study demonstrated that these infected animals showed behavioral impairments accompanied by decreased adult hippocampal neurogenesis [25]. Interestingly, these abnormalities were rescued by increasing adult hippocampal neurogenesis via exercise, indicating that enhancing neurogenesis may help aid recovery for schizophrenia [25].

2.3. Drug Addiction. Drug addiction is a chronic relapsing disorder characterized by a loss of the ability to control drug intake and a compulsive drug-seeking and -taking behavior [92]. Generally, the mesolimbic dopaminergic system in the brain is thought to be an important brain area in the neurobiology of addiction [93]. The hippocampus has received great attention because abusive drugs are potent negative regulators of adult hippocampal neurogenesis and as a result may impair cognitive function [27, 94–96] (Table 3).

Cocaine abuse, a powerful and addictive psychostimulant drug, is associated with dynamic regulation of adult neurogenesis in the hippocampal dentate gyrus and its corresponding memory function [30]. Studies have shown that both a high dose of cocaine or chronic cocaine exposure can decrease the proliferation of neural progenitors in the rat dentate gyrus [28] and cause working memory dysfunction [30]. In addition, chronic treatment of cocaine increased adult neurogenesis in mice in some studies [97], but the same effect is not observed in the rat dentate gyrus [27, 31, 98, 99]. While most studies support evidence that chronic administration of cocaine diminishes proliferation of neural progenitors in the adult hippocampus, the effect on survival and maturation of neural progenitors is not always consistent. Possible explanations for the discrepancy might be due to the differences in animal strain, methods, and duration of cocaine treatment. Therefore, more evidence is needed to determine the timing, duration and consequences of impaired adult hippocampal neurogenesis caused by cocaine. Recently, role of adult hippocampal neurogenesis in cocaine-taking and cocaine-seeking behavior was explored. Using X-ray irradiation approach, ablation of adult hippocampal neurogenesis increased cocaine-taking and cocaine-seeking behavior, suggesting that impaired neurogenesis may result in increased susceptibility in the animal model of cocaine addiction [100]. Interestingly, withdrawal from cocaine administration normalizes the reduction of neural progenitors and enhances maturation of neural progenitors in the adult dentate gyrus [27]. This indicates that the normalization of the cocaine-induced neurogenesis deficit may help decrease susceptibility to relapse and related cognitive deficits.

The correlation between alcohol dependence and hippocampal neurogenesis has been extensively studied. Among other anatomic changes, alcoholic patients undergo structural changes in hippocampal volume [101–103]. Animal models have been used to examine the cellular effects and

Table 2: Select animal studies investigating the effect of schizophrenia on adult hippocampal neurogenesis and behavior.

Species	Susceptibility genes (animal model)	Experimental approach	Effects on neurogenesis	Effects on behavior	References
C57BL6/Jmouse	DISC1	Knockdown of *Disc1* via retroviral-mediated shRNA approach.	Enhanced dendritic outgrowth, soma hypertrophy, overextended migration, accelerated synapse formation of newborn granule neurons.	NS	[18]
C57BL6/Jmouse	DISC1	Knockdown of *Disc1* via retroviral-mediated shRNA approach.	Mistargeted axonal mossy fibers and presynaptic differentiation of newborn granule neuron.	NS	[19]
C57BL6/Jmouse	DISC1	Knockdown of *Disc1* via lentiviral-mediated shRNA approach. Proliferation: BrdU (1 × 40 mg/kg, ip) at 2 hours pulsing chase.	Decreased cell proliferation	Hyperlocomotion in a novel environment. Depressive-like behavior.	[20]
129/SvEv	DISC1 ($Disc1^{Tm1Kara}$ mutant)	BrdU (50 mg/kg, ip) for 12 days.	Decreased neural progenitors and immature neurons. Misoriented apical dendrites on immature neurons.	Selective impairment in working memory	[21]
129/SvEvC57BL/6Jmouse	NPAS3 *Npas3* null mouse	Proliferation: BrdU daily (50 mg/kg, ip) for 12 days and analysis on day 13	Decreased cell proliferation in *Npas3* null mouse.	Locomotor hyperactivity, subtle gait defects, impairment of prepulse inhibition of acoustic startle, deficit in recognition memory, and altered anxiety-related responses.	[22, 23]
C57BL6/Jmouse	SREB2 Overexpression of SREB2 transgenic mouse (Tg) SREB2 null mouse	Proliferation: BrdU (3 × 100 mg/kg, ip) interval 2 hours apart and analysis on Day 3 after BrdU injection. Survival: Three BrdU pulses (100 mg/kg, ip) at 2-hour intervals. Analysis on Day 30 following BrdU injection.	SREB2 Tg: decreased cell proliferation and neuronal maturation. Dendritic morphology deficits in newborn granule neurons. SREB2 null mouse: no change in cell proliferation, but increased neuronal survival rate.	SREB2 Tg: decreased discrimination in the spatial pattern separation. SREB2 null mouse: enhanced discrimination in the spatial pattern separation.	[24]
C57BL6/Jmouse	Prenatal injection of synthetic double-stranded RNA polyriboinosinic polyribocytidylic acid (PolyI:C) into mice	BrdU (100 uL/20 g, ip) daily for 3 days and analysis 4 weeks later.	Decreased neural progenitors.	Behavioral deficits in the open field test and prepulse inhibition of the startle response.	[25, 26]

Table 3: Select animal studies investigating the effect of drug addiction on adult hippocampal neurogenesis.

Species	Drug treatment	Experimental approach	Effect on neurogenesis	References
		Cocaine		
Cocaine/rat (Sprague-Dawley)	Self-administration (0.5 mg/kg infusion, iv) for 3 weeks.	Proliferation: BrdU (150 mg/kg, ip) once at 2-hours pulsing chase after the last self-administration session. Survival: BrdU at four-week pulsing chase after last self-administration session.	Cell proliferation: decreased number of BrdU+ cells. Survival: no change.	[27]
Cocaine/rat (Sprague-Dawley)	20 mg/kg (ip) for either 1 or 14 days	BrdU once (100 mg/kg, ip) at 2-hour pulsing chase on either 1 or 14 days during cocaine exposure.	One-day cocaine injection: no change in cell proliferation Fourteen-day cocaine injection: decreased cell proliferation	[28, 29]
Sprague-Dawley rat	Self-administration (0.5 mg/kg/20 s or 1.5 mg/kg/20 s infusion, iv) for 2 weeks.	Proliferation: BrdU (3 × 50 mg/kg, ip) at 4-hour intervals; analysis twenty-four hours after last BrdU injection. Survival: BrdU (3 × 50 mg/kg, ip) at 4-hour intervals; analysis 4 weeks after last BrdU injection.	Proliferation: decreased number of BrdU+ cells. Survival: decreased neurogenesis.	[30]
Cocaine/rat (Wistar)	20 mg/kg (ip) once daily for 8 days.	BrdU once (40 mg/kg, ip) at 2-hour pulsing chase on final day of cocaine exposure.	Decreased number of BrdU+ cells for cell proliferation. Decreased number of Ki67+ cells with no statistical significance.	[31]
Cocaine/rat (Wistar)	20 mg/kg (ip) daily for 24 days.	BrdU (40 mg/kg, ip) daily for 7 days during the first 7 days of cocaine exposure and Ki67 immunostaining.	Decreased number of Ki67+ cells for cell proliferation. No effect on survival.	[31]
Cocaine/rat (Wistar)	20 mg/kg (ip) daily for either 8 or 24 days.	BrdU (2 × 140 mg/kg, ip) at 6-hour intervals and 24 hours before receiving cocaine exposure.	Both 8-and 24-day cocaine injection: no effect on maturation	[31]
		Alcohol		
Sprague-Dawley rat	Acute: EtOH (5 g/kg) by gavage. Chronic: EtOH (5 g/kg) via intragastric catheter on every 8 hours for 4 days.	Proliferation: Acute: BrdU (2 × 100 mg/kg, ip) at 4 hours 15 minutes and 2 hours and 15 minutes pulsing chase. Chronic: BrdU (4 × 100 mg/kg, ip) daily for 4-day binge EtOH Survival: Acute: BrdU (2 × 100 mg/kg, ip) at 4-week pulsing chase. Chronic: BrdU (4 × 50 mg/kg, ip) at 4-week pulsing chase.	Acute: both proliferation and neuronal maturation decreased. Chronic: both proliferation and neuronal maturation decreased.	[32]
Sprague-Dawley rat	EtOH (final concentration, 6.4% vol/vol) by gavage for 6 weeks.	Proliferation: BrdU (7 × 40 mg/kg, ip) at 2-hour intervals; animals were sacrificed 1 hour after last BrdU injection. Survival: BrdU (40 mg/kg, ip) once daily for first 10 days of 6 weeks EtOH binge; animals were sacrificed 32 days after the last dose of EtOH.	Proliferation: decreased cell proliferation. Maturation and survival: decreased neuronal maturation.	[33]

Table 3: Continued.

Species	Drug treatment	Experimental approach	Effect on neurogenesis	References
Wistar rat	Alcohol nondependent: Self-administered alcohol for 3 weeks and exposed air for 10 weeks. Alcohol dependent: Self-administered alcohol for 3 weeks and exposed to intermittent alcohol vapors for 10 weeks.	At week 6, BrdU (150 mg/kg, ip) at 4-week pulsing chase. Immunostaining: Fluoro-Jade C: cell death neuronal degeneration AC-3: apoptotic cells. Ki-67: proliferating cells. DCX: immature neurons. NeuN: mature neurons.	Nondependent drinking: increased cell death and decreased cell proliferation, immature neurons and survival. Dependent drinking: increased cell death and decreased cell proliferation, immature neurons and survival.	[34]
C57BL/6J mouse	EtOH by gavage for 28 days and abstinence for either 1 or 14 days.	BrdU (3×300 mg/kg, ip) at 4 weeks pulsing chase. Immunostaining: DCX: immature neurons PCNA: proliferating cell marker Behavior tests: open-field locomotor activity and forced swimming test.	Abstinence following alcohol drinking caused: (i) no change in survival of neuronal progenitor cells. (ii) decreased proliferative activity of progenitor cells. (iii) decreased neurogenesis. (iv) increased depressive-like behavior. (v) transiently (1 day abstinence) increased locomotor activity.	[35]
Rhesus Monkey	Alcohol induction: EtOH (1%) by gavage for 40 sessions. Alcohol self-administration: EtOH (6%) by gavage for 200 sessions over 11 months. Alcohol abstinence: abstinence for 2–2.5 months.	Immunostaining: Fluoro-Jade C: cell death neuronal degeneration AC-3: apoptotic cells. Ki-67: proliferating cells. GFAP: radial glia-like stem cell and astrocyte marker. SOX2: amplifying neural progenitor cell marker. NeuroD1: immature neurons. PSA-NCAM: mature neurons.	Abstinence following alcohol drinking caused: (i) decreased neurogenesis, (ii) decreased proliferative activity of progenitor cells in initial phases of neuronal development.	[36]

underlying mechanisms. Converging studies have associated alcohol consumption and dependence with a selective decline in adult hippocampal neurogenesis [32, 104–107]. Acute or chronic administration of ethanol treatment in mice inhibits neural progenitor cell proliferation and survival in the adult rat hippocampus [32, 108]. Clinically relevant animal models have been used to confirm this wherein alcohol dependence reduced proliferation of neural progenitors, as well as consequent differentiation and maturation [34]. More recently, chronic alcohol treatment over 11 months in adolescent macaque monkeys produced selective and long-lasting decrease in hippocampal neural progenitor proliferation [36]. Alterations in synaptic plasticity are also associated with chronic alcohol treatment resulting in the reversible inhibition of long-term potentiation (LTP) in the rat hippocampus [109]. Besides LTP changes, functional or behavioral changes as measured via active avoidance, and spatial memory also occur following alcohol consumption indicating learning and memory deficits [110, 111]. An abstinence state following alcohol consumption can contribute to depression-like behavior with a concurrent reduction in the neural progenitor and immature neuron population in dentate gyrus in mice [35]. This was counteracted via desipramine which alleviated both the structural and functional phenotypes associated with this comorbidity [35]. Whether adult neurogenesis can regulate alcohol-drinking or -seeking behavior remains to be studied. Understanding this causality will allow us to develop improved therapeutic intervention in treating the pathological symptoms associated with alcohol dependence.

3. Therapeutic Interventions

Adult neurogenesis is affected by a variety of external stimuli that influence neuronal activity. The therapeutic effects of electroconvulsive therapy, antidepressants, exercise, and others such as deep brain stimulation have been utilized for their therapeutic efficacy. These have been associated with adult hippocampal neurogenesis and will be discussed below.

3.1. Electroconvulsive Therapy and Deep Brain Stimulation. One of the more intense therapeutic treatments for severe psychiatric disorders, especially depression, is electroconvulsive therapy (ECT) [112]. In the 1940s and 50s, ECT was used in extreme cases in humans when patients did not respond to other treatments [112, 113]. Despite extensive research, the mechanism of action of ECT had not been discovered. Recently, it was linked to neurogenesis, providing a possible explanation for the mechanism that helps to relieve symptoms [37, 38] (Table 4).

The first study to demonstrate the influence of ECT on adult hippocampal neurogenesis showed that a single treatment of ECT results in stimulation of adult neural progenitors that survive up to 3 months afterward [37]. A subsequent study confirmed that ECT promotes neural progenitor proliferation [38]. Further, ECT also reversed long-term neurogenesis deficits and hippocampal-dependent fear memory disrupted by X-ray irradiation [114]. This matches clinical observations that ECT is the most effective treatment for depression. It may also suggest that adult hippocampal neurogenesis is a critical neurobiological component underlying the clinical effect of ECT. A detailed characterization of hippocampal progenitors affected by ECT has been reported. At the cellular level, ECT stimulates proliferation of quiescent progenitor cells and at a later phase increases the proliferation of amplifying progenitor cells [39], which may lead to a net increase in the number of mature adult-born neurons [40]. Functionally, electrophysiological analysis shows that chronic treatment of ECT induces long-term-potentiation-(LTP-) like synaptic changes in the adult dentate gyrus [115]. Given that LTP results in increased proliferation of neural progenitors in the dentate gyrus [116], enhanced neurogenesis caused by ECT could potentially alter hippocampal circuitry which may contribute to the functional effects of ECT.

Exactly how ECT stimulates specific niche signals to regulate the sequential process of adult neurogenesis remains unclear. Growing evidence in the past decade has determined a number of factors that regulate adult neurogenesis in response to ECT in the hippocampus. Among those factors, induction of neurotrophic growth factors has been extensively identified. These include brain-derived neurotrophic factor (BDNF), fibroblast growth factor-2 (FGF-2), and vascular endothelial growth factor (VEGF) [105, 106, 117]. BDNF, a member of the nerve growth factor family expressed throughout the brain, is known to be responsible for synaptic strength, survival, and growth of mature neurons via activation of its receptor TrkB. While both acute and chronic treatments of ECT induce BDNF and TrkB gene expression, the level of BDNF gene expression remains prolonged during chronic treatment of ECT [106]. FGF family of growth factors and corresponding receptors are involved in angiogenesis and early stages of neural development [118, 119]. Expression of FGF-2 mRNA in rodent hippocampus is increased in both acute and chronic treatments of ECT [117]. In addition, infusion of BDNF and FGF into rodent hippocampus results in antidepressant-like effects [120, 121], while blockade of BDNF and FGF in the dentate gyrus induces behavioral deficits and decreases adult hippocampal neurogenesis [121, 122]. These findings indicate that such factors can be strong candidates in ECT treatment to mediate antidepressant effects. A recent study demonstrated ECT promoting DNA demethylation in the BDNF and FGF promoter regions and adult neurogenesis in dentate gyrus through growth arrest and DNA-damage-inducible protein 45 beta (Gadd45b) [42]. These results suggest that dynamic epigenetic DNA modifications may serve as an essential mechanism to translate neurogenic niche signals for sustained regulation of adult neurogenesis and antidepressant action of ECT. Lastly, VEGF is known as a regulator for vascular growth and also a stimulator of neurogenesis [123]. The effect of ECT is dependent upon VEGF signaling for induction of quiescent neural progenitor cell proliferation and is sufficient to produce an antidepressant effect [39]. Taken together, ECT is one of the strongest stimuli of hippocampal neurogenesis. It increases the rate of proliferation and maturation of new neurons in

Table 4: Select animal studies investigating the effect of electroconvulsive therapy (ECT) and deep brain stimulation (DBS) on adult hippocampal neurogenesis.

Species	ECT treatment	Experimental approach	Effects on neurogenesis	Molecular target	References
Electroconvulsive therapy					
Wistar rat	Single ECT	BrdU (2 × 37.5 mg/kg, ip) at 12-hour intervals, starting at 0, 3, 5, 7, and 9 days after ECT and analysis 48 hours after the last injection of BrdU.	Increased cell proliferation.	NS	[37]
Sprague-Dawley rat	Chronic ECT: once daily for 10 days	Proliferation: BrdU (4 × 45 mg/kg, ip) with 2-hour intervals, on 4 days after ECT and analysis at 24 hours after last BrdU injection. Survival: BrdU (4 × 45 mg/kg, ip) with 2-hour intervals before ECT treatment and analysis at 28 days after last BrdU injection.	Increased cell proliferation and survival rate.	NS	[38]
Sprague-Dawley rat	Single ECT Chronic ECT: once daily for 7 days	BrdU (150 mg/kg, ip) at 2 or 24 hours pulsing chase.	2-hour BrdU pulse chase after single ECT: increased quiescent neural progenitors (QNPs) and slightly increased amplifying neural progenitors (ANPs) with no statistical significance. At 24-hour BrdU pulse chase after single and chronic ECT: increased QNPs and ANPs.	VEGF	[39]
Wistar rat	ECT every second day for a course of 8 seizures	Starting with seizure number 5, BrdU (1 × 50 mg/kg, ip) at 2-or 3-hour intervals following each ECT treatment. Injections were also given at the same time during the 3 days between treatments, resulting in a total of seven injections.	Increased maturation of newborn neurons.	NS	[40]
Sprague-Dawley rat	ECT two times	NIT-GFP retroviral injection after ECT treatment.	Increased mushroom spine density on newborn granule neuron dendrites.	NS	[41]
C57BL6/J mouse	Single ECT	BrdU (1 × 200 mg/kg, ip) at 2-hour pulsing chase. GFP retroviral injection.	Increased cell proliferation. Increased total dendritic length and dendritic complexity.	Gadd45b	[42]
Bonnet monkey	ECT: three times a week for four weeks	Proliferation: BrdU (100 mg/kg, iv) daily for 4 days. Survival: BrdU (100 mg/kg, iv) daily for 4 days and analysis at 4 weeks after BrdU injection.	Increased cell proliferation and neuronal maturation.	BCL2	[43]

Table 4: Continued.

Species	ECT treatment	Experimental approach	Effects on neurogenesis	Molecular target	References
		Deep Brain Stimulation			
Sprague-Dawley rat	Anterior thalamic nucleus was stimulated at 2.5 V, 90 μse of pulse width, and variable frequencies (10, 50, 130 Hz) for an hour.	BrdU (4 × 50 mg/kg, ip) with 3-hour interval and analysis at 24 hours or 4 weeks after last BrdU injection.	Increased cell proliferation. No change in neuronal maturation	NS	[44]
C57BL/6 mouse and nestin-CFPnuc transgenic mouse	Anterior Thalamic Nucleus was stimulated at 2.5 V, 90 μs of pulse width, and frequencies 10 Hz (low) and 130 Hz (high) for an hour.	BrdU (3 × 50 mg/kg, ip) with 3-hour intervals and analysis at 24 hours or 30 days after last BrdU injection.	Increased cell proliferation in high frequency but not low frequency. Increased proliferation of amplifying neural progenitors (ANPs), but no change in quiescent neural progenitors (QNPs) at high frequency. Increased neuronal maturation at high frequency but not low frequency.	NS	[45]
Mixed genetic background between C57BL/6NTacfBr and 129Svev mice	Entorhinal cortex at at 0–500 μA current, for 30–120 minutes, at 90 μs of pulse width and 130 Hz frequency.	Proliferation: BrdU (1 × 200 mg/kg, ip) injection on day 1, 3, 5, or 7 after stimulation and analysis at 24 hours after BrdU injection. Neuronal maturation: IdU (57 mg/kg, ip) injections during the period of stimulation-induced increased proliferation (postoperative days 3–5), CldU (42.5 mg/kg, i.p.) injections during a similar period of baseline proliferation (postoperative days 7–9), and analysis about 10 weeks later. Survival: BrdU daily (100 mg/kg, ip) with 8-hour intervals for 3 days on 1, 10, or 30 days before stimulation and postoperative analysis 3 weeks later. Dendritic development: GFP-retroviral injection.	Increased cell proliferation after 60 or 120 minutes of stimulation and at 50, 250, and 500 μA. No change in fate determination of newborn cells. Increased dendritic length of newborn granule neurons but no change in nodes per neuron and dendritic spine size.	Improved spatial memory formation measured by water maze.	[46]

the dentate gyrus which could have a significant effect on hippocampal circuitry. ECT's therapeutic efficacy may be a result of this, proving to be an effective treatment for severe depression and other mood disorders.

Another possible treatment for neuropsychiatric disorders is deep brain stimulation (DBS). DBS is an extreme treatment for a wide range of neurological disorders such as Parkinson's disease, dystonia, chronic pain, and tremors [124]. It has proven effective in treating these disorders in initial trials [125, 126] and also in treating major depression especially in patients that do not respond to chemical antidepressants [127]. Growing evidence has focused on multiple anatomical targets in the brain with different stimulation frequencies, pulse width, and amplitude in order to obtain the ideal setting for conferring an antidepressant response (Table 4). The limited number of studies that have been conducted thus far suggests that DBS may proceed via an increase in adult neurogenesis and survival rate of mature neurons integrating into the hippocampal circuitry (Table 4). The first evidence reported by [44] demonstrated that high frequency stimulation of the rat thalamus increased adult neurogenesis and restored experimentally suppressed neurogenesis in the dentate gyrus. One study shows that stimulation of the anterior thalamic nuclei has the effect of promoting proliferation of ANPs, similar to fluoxetine [45]. Similarly, other studies have identified specific stimulation of the entorhinal cortex, a major source of input to the hippocampus, promoting proliferation of progenitors which increase the survival rate and formation of mature neurons integrating into the hippocampal circuitry [46]. This finding was supported by increased performance in the water-maze memory tests. DBS may prove to be a significant approach in combating psychiatric disorders. However, direct involvement of adult neurogenesis conferring antidepressant action of DBS as well as its mechanism of action needs to be determined.

3.2. Chronic Treatment of Chemical Antidepressants. Multiple classes of antidepressants have been shown to positively influence aspects of adult hippocampal neurogenesis in a chronic time course manner (Table 5). Most selective serotonin reuptake inhibitor (SSRI) treatments are associated with a delayed onset of therapeutic efficacy consistent with the time course of maturation of newborn neurons [66]. Stimulation of neurogenesis is required for antidepressant efficacy. Studies in monkeys and rodents confirm that in conferring antidepressant action, chronic fluoxetine treatment stimulates adult hippocampal neurogenesis and mediated depressive-like behavioral effects [38, 47, 116].

The cellular basis of fluoxetine action within the neuronal differentiation cascade has been identified. Using a nestin-cyan fluorescent protein (CFP)nuc mouse line where the reporter is fused to a nuclear localization signal that allows identification and classification of early neuronal progenitors, fluoxetine was shown to increase symmetric divisions of the amplifying neuronal progenitor (ANP) cell class while not affecting division of stem-like cells in the dentate gyrus [49]. These results suggest that the cellular target for fluoxetine's therapeutic action to increase new neurons arises due to a resultant expansion of this ANP cell class. Another study by Wang et al. [48] showed that fluoxetine stimulates dendritic development of newborn neurons and neurogenesis-dependent LTP in the dentate gyrus which results in behavioral alteration induced by fluoxetine. By ablating adult neurogenesis using X-ray irradiation, they indicated that fluoxetine-induced LTP and behavior response both require adult hippocampal neurogenesis [17]. Thus, the effects of chronic fluoxetine administration on the maturation and functional properties of newborn neurons may translate into enhanced synaptic plasticity in the appropriate neural circuits, which subsequently exhibit a behavioral response to antidepressant action. Besides studying fluoxetine, various other classes of antidepressant drugs have been tested. For instance, similar results were seen in rodent administration of SSRI citalopram [50] and tricyclic antidepressant imipramine [51], although this effect was observed in the stressed condition. The mood stabilizers including lithium have been shown to significantly increase both neural progenitors and survival [128]. Although a more detailed characterization of the cellular process needs to be determined, these studies support the notion that regulation of adult neurogenesis may provide potential therapeutic targets for treatment of depression.

What underlies the neurogenic action of antidepressants? Extrinsic factors such as BDNF and VEGF that regulate the microniche of adult neurogenesis may hold the answer. BDNF serves as a key regulator of various aspects in adult hippocampal neurogenesis including proliferation, survival, dendritic growth, maturation, and synaptic plasticity, which could make BDNF a potential mediator of the antidepressant action induced by different chemical antidepressants [129]. Emerging evidence shows that BDNF and TrkB mRNA levels in the hippocampus are dramatically induced by chronic treatment of different chemical antidepressants including fluoxetine, tranylcypromine, sertraline, desipramine, and mianserin [130]. Infusion of exogenous BDNF into the hippocampus exhibits antidepressive-like behavioral responses [120]. Further, behavioral abnormalities found in heterozygous BDNF (BDNF+/−) mice and mice lacking the TrkB are counteracted by chronic antidepressants including fluoxetine [131]. This data coupled with evidence that BDNF-signaling enhances adult neurogenesis in the dentate gyrus [132] led to the suggestion that antidepressants may represent an enhancement of neural plasticity such as adult neurogenesis and behavioral alteration, which in turn could be regulated by increasing the level of BDNF [130]. However, a direct link between adult hippocampal neurogenesis and antidepressive-like behavioral action of antidepressants through BDNF is still lacking.

The other promising molecular target is VEGF, which shows increased mRNA levels with chronic treatment of fluoxetine and desipramine in the hippocampus [133]. VEGF is also sufficient to promote basal level of adult hippocampal neurogenesis [134] and is necessary for the antidepressant action [133]. Conversely, a significant role of VEGF is demonstrated when VEGF signaling diminishes induction of adult neurogenesis and antidepressant action in response to chronic treatment of fluoxetine [133]. Taken together,

Table 5: Select animal studies investigating the effect of chemical antidepressants on adult hippocampal neurogenesis and animal behavior.

Species	Antidepressant treatment	Experimental paradigm	Effect on neurogenesis	Effects on behavior	References
129/SvEv	Fluoxetine (SSRI): 10 mg/kg/day via drinking water for 28 days	Proliferation: BrdU (4 × 75 mg/kg, ip) at 2-hour intervals and analysis at 24 hours after last BrdU injection. Survival: BrdU (4 × 75 mg/kg, ip) at 2-hour intervals and analysis at 28 days after last BrdU injection.	Increased cell proliferation and neuronal maturation.	Less depressive-like behavior in novelty suppressed feeding test.	[47]
Sprague Dawley rat	Fluoxetine (SSRI): 5 mg/kg (ip) 1, 5, 14, 28 d. Tranylcypromine: 7 × 7.5 mg/kg (ip) daily, then 14 × 10 mg/kg (ip) daily. Reboxetine: 20 mg/kg, 2x per day for 21 d	Proliferation: BrdU (4 × 75 mg/kg, ip) at 2-hour intervals, then 24-hour pulsing-chase. Survival: BrdU (4 × 75 mg/kg, ip) at 2-hour intervals, then 28-day pulsing chase.	Fluoxetine (SSRI): increased cell proliferation and neuronal maturation. Tranylcypromine: increased cell proliferation Reboxetine: increased cell proliferation.	NS	[38]
129/SvEv	Fluoxetine (SSRI): 18 mg/kg daily by gavage for behavior test and via drinking water for all experiments for 5 days (subchronic) or 28 days (chronic).	Proliferation: BrdU (1 × 150 mg/kg, ip) at 2-hour pulsing chase. Dendritic maturation, survival, and neuronal maturation: BrdU (4 × 75 mg/kg, ip over 8 hours) on day 0, started fluoxetine treatment on day 1, and analysis on day 21 by BrdU and DCX coimmunostaining.	Increased cell proliferation in chronic treatment of fluoxetine. No change in number of DCX+ immature neurons. Increased dendritic length and dendritic complexity in chronic treatment of fluoxetine, but not subchronic treatment measured by BrdU+ DCX+ colabeled neurons. Increased survival rate and neuronal maturation in chronic treatment of fluoxetine.	Less depressive-like behavior by chronic fluoxetine treatment (but not subchronic) in novelty suppressed feeding test.	[48]
Nestin-CFPnuc mouse	Fluoxetine (SSRI): 10 mg/kg (ip) daily for 15 days.	Proliferation: BrdU (1 × 150 mg/kg, ip) and analysis 1 day later. Survival: BrdU (1 × 150 mg/kg, ip) and analysis 30 days later.	Increased proliferation of amplifying neural progenitors (ANPs), but no change in quiescent neural progenitors (QNPs). Increased survival rate.	NS	[49]
Wistar rat	Escitalopram (SSRI): 5 mg/kg (ip) for 4 weeks.	BrdU (4 × 100 mg/kg, ip) with 2-hour interval during one day and analysis at 16 hours after the last BrdU injection.	Increased cell proliferation in chronic treatment of escitalopram under mild stress condition, but no change under the normal condition.	Chronic treatment of escitalopram shows recovery from anhedonic-like behavior caused by the mild stress condition.	[50]
BALB/c mouse	Fluoxetine (SSRI): 10 mg/kg (ip) daily for 28 days, Imipramine (TCA): 20 mg/kg (ip) daily for 28 days.	BrdU (4 × 75 mg/kg, ip) with 2 hours interval and analysis at 24 hours after last BrdU injection.	Increased cell proliferation in chronic treatment of fluoxetine and imipramine under unpredictable chronic mild stress.	Chronic treatment of fluoxetine and imipramine shows recovery from depressive-like behavior caused by unpredictable chronic mild stress condition.	[51]
Wistar rat	Lithium chloride: 2.5 mEq/kg (ip) for 14 days.	BrdU (50 mg/kg, ip) daily during last 3 days of experimental period	Increased proliferation, neuronal maturation and glial maturation of neural progenitors under normal conditions. Lithium treatment shows recovery from neurogenesis deficits under unpredictable chronic mild stress condition.	Lithium treatment shows less depressive-like behavior under normal conditions. Lithium treatment shows recovery from depressive-like behavior caused by unpredictable chronic mild stress condition.	[52]

*SSRI: Selective serotonin reuptake inhibitor; TCA: Tricyclic Antidepressants; DCX: doublecortin (immature neuronal marker) 13.

an important action of antidepressants may be to increase neurogenesis in the hippocampus through a variety of molecular mechanisms which have an impact on depressive symptoms.

3.3. Exercise Therapy. Physiological stimulation in the form of exercise has been shown to stimulate cell proliferation and adult hippocampal neurogenesis [53, 55, 56, 135] and enhance the learning and memory function in both mice [57, 136] and human [137]. The plastic nature of the mammalian brain, especially neurogenesis continuing in the hippocampus well into adulthood, has allowed for exercise to exert its effects at the cellular level. This not only holds promise for brain diseases such as Alzheimer's or Parkinson's disease, but also for schizophrenia and major depression. A recent clinical trial demonstrated how exercise therapy can improve the mental health and cardiovascular fitness in patients with schizophrenia [138]. The use of exercise in treating depression has also received increased attention in recent years.

The cellular effects of running on hippocampal neurogenesis have been closely investigated in a number of notable studies (Table 6). Using BrdU labeling, one of the earlier studies showed how mice with free access to a running wheel nearly doubled the number of surviving newborn cells [53]. Another study has shown that running actually activates the quiescent radial population in the hippocampus [55]. These activated cells eventually give rise to mature neurons that are functionally integrated into the hippocampal circuitry. Hippocampal structure and function has been closely studied in relation to cognitive or mental function. Running has been shown to improve neurogenesis with a corresponding enhancement in learning and long-term potentiation [57, 136]. Physical exercise has all been implicated in the distinct encoding of spatial information. In one study, young and aged mice undergoing running exercise were compared to each other with the result that running enhanced spatial pattern separation when exercise was correlated with increased hippocampal neurogenesis [56]. The effect of exercise-induced neurogenesis has been studied in humans as well. Cerebral blood volume (CBV) maps in hippocampal formation have been generated in both exercising mice and humans [54]. Similar to mice, exercise specifically targeted the dentate gyrus CBV in humans and was correlated with increased cognitive function.

It is still unclear whether benefits from physical exercise, namely, in cognitive function, are conferred by the increase in neurogenesis. Any causal relationship between the increased neurogenesis and benefits in learning and memory is still being studied, and at least, current evidence is not decisive [139]. BDNF, as previously noted playing a role in both ECT and antidepressants, has again been suggested as a strong contender and possible mediator of the causal relationship being observed. A recent clinical trial demonstrated that exercise training increased hippocampal volume, effectively reversing age-related loss in volume [137]. This increased hippocampal volume was also associated with greater serum levels of BDNF [137]. In the review of Bekinschetein et al., a number of studies are mentioned indicating a strong causal link between BDNF and learning and memory. Physical activity has been associated with increased BDNF mRNA levels in the rat hippocampus, especially the dentate gyrus [140, 141]. Specific deletion of the BDNF-receptor TrkB reduced survival of newborn neurons, impaired neurogenesis-dependent LTP, and increased anxiety-like behavior [142]. Indeed, ablation of TrkB in neural progenitors also prevented behavioral improvements conferred by exercise [132], bolstering the role of BDNF. Other factors, namely, NMDA receptors and downstream effectors such as calcium/calmodulin protein kinase II and mitogen-activated protein kinase, could be involved in the mechanism by which BDNF effects synaptic plasticity [143]. Further evidence may ascertain the mechanism by which BDNF operates.

In an effort to treat neuropsychiatric disorders in relation to aberrant neurogenesis, it is important to study and classify important stimuli that can have a lasting effect on neurogenesis and hippocampal function. ECT, DBS, antidepressants, and physical exercise all seem to have their own effect on neurogenesis, and in combination with proper administration, they could prove vital in discovering potential treatments for psychiatric disorders that continue to disable the population.

4. Conclusion

Significant progress has been made in the past decade documenting the function and regulation of adult neurogenesis. Many offer a neurobiological understanding of the role of adult neurogenesis in psychiatric disorders. These studies demonstrate how these disorders may proceed through an impairment of neural progenitor proliferation in the hippocampus, and how an ablation of neurogenesis may predispose an animal to depressive-like behaviors. A pathophysiologically reliable animal model is, however, still required to confirm data across the cellular and behavioral spectrums. Further, genetic manipulations of susceptibility genes in loss-of-function transgenic models may be used to rescue cognitive deficits and confirm their roles via cellular and behavioral studies. These may bolster a causal link between adult neurogenesis and the disorder.

However, with our current knowledge, several questions remain to be answered. We do not yet have a clear understanding of how external stimuli in current treatments mediate the induction and function of factors in the neurogenic niche that stimulate adult hippocampal neurogenesis. We also do not know how the regulation of neurogenic niche via niche signals and neurotrophic factors may be altered by X-ray irradiation which is used to ablate and study neurogenesis. The role of BDNF has been evidenced in ECT, DBS, antidepressants, and exercise therapy. Going forward, its downstream effects would need to be studied to determine how BDNF mediates synaptic plasticity. Further, in the case of relatively new treatments such as DBS, conclusive evidence of its antidepressant effect and mechanism of action will bolster the significant role it could play in treating psychiatric

Table 6: Select animal studies investigating the effect of exercise on adult hippocampal neurogenesis.

Species	Type of exercise	Experimental paradigm	Effect on neurogenesis	Effects on behavior	References
C57BL/6 mouse	Learners: 2 trials of Morris water maze training per day over 30 days. (with platform). Swimmers: 2 trials of morris water maze training per day over 30 days (without platform). Runners: 1 running wheel for 3-4 mice in rat cage.	Proliferation: BrdU (12 × 50 mg/kg, ip) daily for 12 days, then 24-hour pulsing chase. Survival: BrdU (12 × 50 mg/kg, ip) daily for 12 days, then 40-week pulsing chase.	Learners: decreased cell proliferation and survival of newly generated neurons. Swimmers: decreased cell proliferation and survival of newly generated neurons Runners: increased cell proliferation and survival of newly generated neurons.	NS	[53]
C57BL/6 mouse	Voluntary running.	MRI scan at weeks 0, 2, 4, and 6. Survival: BrdU (7 × 60 mg/kg, ip) during second week of experiment, then 4-weeks pulsing chase.	Exercise: (i) increased cerebral blood volume specifically at dentate gyrus. (ii) increased neuronal maturation.	NS	[54]
Hes5::GFP mouse	Voluntary running.	BrdU (50 mg/kg, ip) either once or 3 consecutive times (2-hour interval); then 2 hour or 5-day pulsing chase.	Exercise activated QNPs in dentate gyrus.	NS	[55]
C57/BL6 mouse	Voluntary running.	Spatial pattern separation. BrdU (5 × 50 mg/kg, ip) daily for 5 days, then 10-week pulsing chase.	Exercise increased neuronal maturation.	Exercise increased spatial pattern separation.	[56]
C57/BL6 mouse	Voluntary running.	Spatial learning: Morris water maze task day 35–39. Proliferation: BrdU (7 × 50 mg/kg, ip) daily for 7 days, then 24-hour pulsing chase. Neuronal morphology analysis: 4-week dpi GFP retrovirus. Blood vessel size analysis: Lectin staining	Proliferation: increased proliferation in both young and old mice. Neuronal morphology analysis: increased dendritic length and branches in young runners. increased blood vessel size in young runners only.	Exercise enhanced spatial learning in both young and old mice.	[57]

disorders. Finally, we are yet to obtain a conclusive causal relationship between adult neurogenesis and depression, as well as adult neurogenesis and cognitive function or learning and memory. How drug abuse may alter these relationships especially in the maturation and integration of newborn neurons in the hippocampus will be useful. Current evidence when coupled to such a characterization may provide evidence of the cellular mechanism at play in many widely used treatments.

Adult neurogenesis is important in synaptic plasticity with overarching roles in memory, learning, and mood. Such roles have been established via a plethora of studies which show that the process is dynamically regulated and demonstrates striking structural plasticity in response to internal and environmental cues. It is strongly regulated by stress signals, and stimulated by antidepressants, which may serve to potentially alter the hippocampal circuitry. Current evidence indicates a significant role for adult neurogenesis in the neurological impairments of psychiatric disorders. At this critical juncture, it is important we distinguish whether adult neurogenesis is causally involved in the etiology or plays a significant role in ameliorating the disease state symptoms. A clear understanding of this relationship in regard to the pathogenesis of the disease will be invaluable in aiding our understanding of the causes at play, and how effective treatments may be designed to alleviate the symptoms.

Authors' Contribution

H. Jun and S. Hussaini contributed equally to this work.

Acknowledgments

The authors would like to thank Dr. Hongjun Song for suggestions. The research in Dr. Jang laboratory was supported by NIH (R00MH090115), NARSAD (Young Investigator Award), start-up funds from Mayo foundation to M. H. Jang, and by a Summer Undergraduate Research Fellowship from Mayo Graduate School to M. J. Rigby.

References

[1] R. C. Kessler, W. T. Chiu, O. Demler, K. R. Merikangas, and E. E. Walters, "Prevalence, severity, and comorbidity of 12-month DSM-IV disorders in the National Comorbidity Survey Replication," *Archives of General Psychiatry*, vol. 62, no. 6, pp. 617–627, 2005.

[2] Y. Li, Y. Mu, and F. H. Gage, "Development of neural circuits in the adult hippocampus," *Current Topics in Developmental Biology*, vol. 87, pp. 149–174, 2009.

[3] D. R. Weinberger, "Cell biology of the hippocampal formation in schizophrenia," *Biological Psychiatry*, vol. 45, no. 4, pp. 395–402, 1999.

[4] Y. I. Sheline, P. W. Wang, M. H. Gado, J. G. Csernansky, and M. W. Vannier, "Hippocampal atrophy in recurrent major depression," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 93, no. 9, pp. 3908–3913, 1996.

[5] J. D. Bremner, M. Narayan, E. R. Anderson, L. H. Staib, H. L. Miller, and D. S. Charney, "Hippocampal volume reduction in major depression," *The American Journal of Psychiatry*, vol. 157, no. 1, pp. 115–118, 2000.

[6] M. D. Nelson, A. J. Saykin, L. A. Flashman, and H. J. Riordan, "Hippocampal volume reduction in schizophrenia as assessed by magnetic resonance imaging: a meta-analytic study," *Archives of General Psychiatry*, vol. 55, no. 5, pp. 433–440, 1998.

[7] J. G. Csernansky, S. Joshi, L. Wang et al., "Hippocampal morphometry in schizophrenia by high dimensional brain mapping," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 95, no. 19, pp. 11406–11411, 1998.

[8] S. Heckers, "Neuroimaging studies of the hippocampus in schizophrenia," *Hippocampus*, vol. 11, no. 5, pp. 520–528, 2001.

[9] V. Dhikav and K. S. Anand, "Is hippocampal atrophy a future drug target?" *Medical Hypotheses*, vol. 68, no. 6, pp. 1300–1306, 2007.

[10] N. A. DeCarolis and A. J. Eisch, "Hippocampal neurogenesis as a target for the treatment of mental illness: a critical evaluation," *Neuropharmacology*, vol. 58, no. 6, pp. 884–893, 2010.

[11] K. Pham, J. Nacher, P. R. Hof, and B. S. McEwen, "Repeated restraint stress suppresses neurogenesis and induces biphasic PSA-NCAM expression in the adult rat dentate gyrus," *European Journal of Neuroscience*, vol. 17, no. 4, pp. 879–886, 2003.

[12] J. E. Malberg and R. S. Duman, "Cell proliferation in adult hippocampus is decreased by inescapable stress: reversal by fluoxetine treatment," *Neuropsychopharmacology*, vol. 28, no. 9, pp. 1562–1571, 2003.

[13] J. Yun, H. Koike, D. Ibi et al., "Chronic restraint stress impairs neurogenesis and hippocampus-dependent fear memory in mice: possible involvement of a brain-specific transcription factor Npas4," *Journal of Neurochemistry*, vol. 114, no. 6, pp. 1840–1851, 2010.

[14] K. Jaako-Movits and A. Zharkovsky, "Impaired fear memory and decreased hippocampal neurogenesis following olfactory bulbectomy in rats," *European Journal of Neuroscience*, vol. 22, no. 11, pp. 2871–2878, 2005.

[15] R. M. Thomas, G. Hotsenpiller, and D. A. Peterson, "Acute psychosocial stress reduces cell survival in adult hippocampal neurogenesis without altering proliferation," *The Journal of Neuroscience*, vol. 27, no. 11, pp. 2734–2743, 2007.

[16] C. Lieberwirth, Y. Liu, X. Jia, and Z. Wang, "Social isolation impairs adult neurogenesis in the limbic system and alters behaviors in female prairie voles," *Hormones and Behavior*, vol. 62, no. 4, pp. 357–366, 2012.

[17] B. Czéh, T. Welt, A. K. Fischer et al., "Chronic psychosocial stress and concomitant repetitive transcranial magnetic stimulation: effects on stress hormone levels and adult hippocampal neurogenesis," *Biological Psychiatry*, vol. 52, no. 11, pp. 1057–1065, 2002.

[18] X. Duan, J. H. Chang, S. Ge et al., "Disrupted-in-schizophrenia 1 regulates integration of newly generated neurons in the adult brain," *Cell*, vol. 130, no. 6, pp. 1146–1158, 2007.

[19] R. L. Faulkner, M. H. Jang, X. B. Liu et al., "Development of hippocampal mossy fiber synaptic outputs by new neurons in the adult brain," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 37, pp. 14157–14162, 2008.

[20] Y. Mao, X. Ge, C. L. Frank et al., "Disrupted in schizophrenia 1 regulates neuronal progenitor proliferation via modulation

of GSK3β/β-catenin signaling," *Cell*, vol. 136, no. 6, pp. 1017–1031, 2009.

[21] M. Kvajo, H. McKellar, L. J. Drew et al., "Altered axonal targeting and short-term plasticity in the hippocampus of Disc1 mutant mice," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 49, pp. E1349–E1358, 2011.

[22] A. A. Pieper, X. Wu, T. W. Han et al., "The neuronal PAS domain protein 3 transcription factor controls FGF-mediated adult hippocampal neurogenesis in mice," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 102, no. 39, pp. 14052–14057, 2005.

[23] E. W. Brunskill, L. A. Ehrman, M. T. Williams et al., "Abnormal neurodevelopment, neurosignaling and behaviour in Npas3-deficient mice," *European Journal of Neuroscience*, vol. 22, no. 6, pp. 1265–1276, 2005.

[24] Q. Chen, J. H. Kogan, A. K. Gross et al., "SREB2/GPR85, a schizophrenia risk factor, negatively regulates hippocampal adult neurogenesis and neurogenesis-dependent learning and memory," *European Journal of Neuroscience*, vol. 36, no. 5, pp. 2597–2608, 2012.

[25] S. A. Wolf, A. Melnik, and G. Kempermann, "Physical exercise increases adult neurogenesis and telomerase activity, and improves behavioral deficits in a mouse model of schizophrenia," *Brain, Behavior, and Immunity*, vol. 25, no. 5, pp. 971–980, 2011.

[26] S. E. P. Smith, J. Li, K. Garbett, K. Mirnics, and P. H. Patterson, "Maternal immune activation alters fetal brain development through interleukin-6," *The Journal of Neuroscience*, vol. 27, no. 40, pp. 10695–10702, 2007.

[27] M. A. Noonan, K. H. Choi, D. W. Self, and A. J. Eisch, "Withdrawal from cocaine self-administration normalizes deficits in proliferation and enhances maturity of adult-generated hippocampal neurons," *The Journal of Neuroscience*, vol. 28, no. 10, pp. 2516–2526, 2008.

[28] M. Yamaguchi, T. Suzuki, T. Seki et al., "Repetitive cocaine administration decreases neurogenesis in adult rat hippocampus," *Annals of the New York Academy of Sciences*, vol. 1025, pp. 351–362, 2004.

[29] M. Yamaguchi, T. Suzuki, T. Seki et al., "Decreased cell proliferation in the dentate gyrus of rats after repeated administration of cocaine," *Synapse*, vol. 58, no. 2, pp. 63–71, 2005.

[30] E. Sudai, O. Croitoru, A. Shaldubina et al., "High cocaine dosage decreases neurogenesis in the hippocampus and impairs working memory," *Addiction Biology*, vol. 16, no. 2, pp. 251–260, 2011.

[31] L. Domínguez-Escribà, V. Hernández-Rabaza, M. Soriano-Navarro et al., "Chronic cocaine exposure impairs progenitor proliferation but spares survival and maturation of neural precursors in adult rat dentate gyrus," *European Journal of Neuroscience*, vol. 24, no. 2, pp. 586–594, 2006.

[32] K. Nixon and F. T. Crews, "Binge ethanol exposure decreases neurogenesis in adult rat hippocampus," *Journal of Neurochemistry*, vol. 83, no. 5, pp. 1087–1093, 2002.

[33] D. G. Herrera, A. G. Yagüe, S. Johnsen-Soriano et al., "Selective impairment of hippocampal neurogenesis by chronic alcoholism: protective effects of an antioxidant," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 100, no. 13, pp. 7919–7924, 2003.

[34] H. N. Richardson, S. H. Chan, E. F. Crawford et al., "Permanent impairment of birth and survival of cortical and hippocampal proliferating cells following excessive drinking during alcohol dependence," *Neurobiology of Disease*, vol. 36, no. 1, pp. 1–10, 2009.

[35] J. R. Stevenson, J. P. Schroeder, K. Nixon, J. Besheer, F. T. Crews, and C. W. Hodge, "Abstinence following alcohol drinking produces depression-like behavior and reduced hippocampal neurogenesis in mice," *Neuropsychopharmacology*, vol. 34, no. 5, pp. 1209–1222, 2009.

[36] M. A. Taffe, R. W. Kotzebue, R. D. Crean, E. F. Crawford, S. Edwards, and C. D. Mandyam, "Long-lasting reduction in hippocampal neurogenesis by alcohol consumption in adolescent nonhuman primates," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 107, no. 24, pp. 11104–11109, 2010.

[37] T. M. Madsen, A. Treschow, J. Bengzon, T. G. Bolwig, O. Lindvall, and A. Tingstrom, "Increased neurogenesis in a model of electroconvulsive therapy," *Biological Psychiatry*, vol. 47, no. 12, pp. 1043–1049, 2000.

[38] J. E. Malberg, A. J. Eisch, E. J. Nestler, and R. S. Duman, "Chronic antidepressant treatment increases neurogenesis in adult rat hippocampus," *The Journal of Neuroscience*, vol. 20, no. 24, pp. 9104–9110, 2000.

[39] E. Segi-Nishida, J. L. Warner-Schmidt, and R. S. Duman, "Electroconvulsive seizure and VEGF increase the proliferation of neural stem-like cells in rat hippocampus," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 32, pp. 11352–11357, 2008.

[40] B. W. Scott, J. M. Wojtowicz, and W. M. Burnham, "Neurogenesis in the dentate gyrus of the rat following electroconvulsive shock seizures," *Experimental Neurology*, vol. 165, no. 2, pp. 231–236, 2000.

[41] C. Zhao, J. Warner-Schmidt, R. S. Duman, and F. H. Gage, "Electroconvulsive seizure promotes spine maturation in newborn dentate granule cells in adult rat," *Developmental Neurobiology*, vol. 72, no. 6, pp. 937–942, 2012.

[42] D. K. Ma, M. H. Jang, J. U. Guo et al., "Neuronal activity-induced Gadd45b promotes epigenetic DNA demethylation and adult neurogenesis," *Science*, vol. 323, no. 5917, pp. 1074–1077, 2009.

[43] T. D. Perera, J. D. Coplan, S. H. Lisanby et al., "Antidepressant-induced neurogenesis in the hippocampus of adult nonhuman primates," *The Journal of Neuroscience*, vol. 27, no. 18, pp. 4894–4901, 2007.

[44] H. Toda, C. Hamani, A. P. Fawcett, W. D. Hutchison, and A. M. Lozano, "The regulation of adult rodent hippocampal neurogenesis by deep brain stimulation: laboratory investigation," *Journal of Neurosurgery*, vol. 108, no. 1, pp. 132–138, 2008.

[45] J. M. Encinas, C. Hamani, A. M. Lozano, and G. Enikolopov, "Neurogenic hippocampal targets of deep brain stimulation," *The Journal of Comparative Neurology*, vol. 519, no. 1, pp. 6–20, 2011.

[46] S. S. Stone, C. M. Teixeira, L. M. Devito et al., "Stimulation of entorhinal cortex promotes adult neurogenesis and facilitates spatial memory," *The Journal of Neuroscience*, vol. 31, no. 38, pp. 13469–13484, 2011.

[47] L. Santarelli, M. Saxe, C. Gross et al., "Requirement of hippocampal neurogenesis for the behavioral effects of antidepressants," *Science*, vol. 301, no. 5634, pp. 805–809, 2003.

[48] J. W. Wang, D. J. David, J. E. Monckton, F. Battaglia, and R. Hen, "Chronic fluoxetine stimulates maturation and synaptic plasticity of adult-born hippocampal granule cells," *The Journal of Neuroscience*, vol. 28, no. 6, pp. 1374–1384, 2008.

[49] J. M. Encinas, A. Vaahtokari, and G. Enikolopov, "Fluoxetine targets early progenitor cells in the adult brain," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 103, no. 21, pp. 8233–8238, 2006.

[50] M. N. Jayatissa, C. Bisgaard, A. Tingström, M. Papp, and O. Wiborg, "Hippocampal cytogenesis correlates to escitalopram-mediated recovery in a chronic mild stress rat model of depression," *Neuropsychopharmacology*, vol. 31, no. 11, pp. 2395–2404, 2006.

[51] A. Surget, M. Saxe, S. Leman et al., "Drug-dependent requirement of hippocampal neurogenesis in a model of depression and of antidepressant reversal," *Biological Psychiatry*, vol. 64, no. 4, pp. 293–301, 2008.

[52] G. Chen, G. Rajkowska, F. Du, N. Seraji-Bozorgzad, and H. K. Manji, "Enhancement of hippocampal neurogenesis by lithium," *Journal of Neurochemistry*, vol. 75, no. 4, pp. 1729–1734, 2000.

[53] H. van Praag, G. Kempermann, and F. H. Gage, "Running increases cell proliferation and neurogenesis in the adult mouse dentate gyrus," *Nature Neuroscience*, vol. 2, no. 3, pp. 266–270, 1999.

[54] A. C. Pereira, D. E. Huddleston, A. M. Brickman et al., "An in vivo correlate of exercise-induced neurogenesis in the adult dentate gyrus," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 104, no. 13, pp. 5638–5643, 2007.

[55] S. Lugert, O. Basak, P. Knuckles et al., "Quiescent and active hippocampal neural stem cells with distinct morphologies respond selectively to physiological and pathological stimuli and aging," *Cell Stem Cell*, vol. 6, no. 5, pp. 445–456, 2010.

[56] D. J. Creer, C. Romberg, L. M. Saksida, H. van Praag, and T. J. Bussey, "Running enhances spatial pattern separation in mice," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 107, no. 5, pp. 2367–2372, 2010.

[57] H. van Praag, T. Shubert, C. Zhao, and F. H. Gage, "Exercise enhances learning and hippocampal neurogenesis in aged mice," *The Journal of Neuroscience*, vol. 25, no. 38, pp. 8680–8685, 2005.

[58] J. Altman and G. D. Das, "Autoradiographic and histological evidence of postnatal hippocampal neurogenesis in rats," *The Journal of Comparative Neurology*, vol. 124, no. 3, pp. 319–335, 1965.

[59] G. L. Ming and H. Song, "Adult neurogenesis in the mammalian central nervous system," *Annual Review of Neuroscience*, vol. 28, pp. 223–250, 2005.

[60] C. D. Clelland, M. Choi, C. Romberg et al., "A functional role for adult hippocampal neurogenesis in spatial pattern separation," *Science*, vol. 325, no. 5937, pp. 210–213, 2009.

[61] A. Sahay, K. N. Scobie, A. S. Hill et al., "Increasing adult hippocampal neurogenesis is sufficient to improve pattern separation," *Nature*, vol. 472, no. 7344, pp. 466–470, 2011.

[62] D. K. Ma, W. R. Kim, G. L. Ming, and H. Song, "Activity-dependent extrinsic regulation of adult olfactory bulb and hippocampal neurogenesis," *Annals of the New York Academy of Sciences*, vol. 1170, pp. 664–673, 2009.

[63] 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.

[64] D. C. Lie, H. Song, S. A. Colamarino, G. L. Ming, and F. H. Gage, "Neurogenesis in the adult brain: new strategies for central nervous system diseases," *Annual Review of Pharmacology and Toxicology*, vol. 44, pp. 399–421, 2004.

[65] C. Mirescu and E. Gould, "Stress and adult neurogenesis," *Hippocampus*, vol. 16, no. 3, pp. 233–238, 2006.

[66] A. Sahay and R. Hen, "Adult hippocampal neurogenesis in depression," *Nature Neuroscience*, vol. 10, no. 9, pp. 1110–1115, 2007.

[67] J. Veena, B. N. Srikumar, K. Mahati, T. R. Raju, and B. S. Shankaranarayana Rao, "Oxotremorine treatment restores hippocampal neurogenesis and ameliorates depression-like behaviour in chronically stressed rats," *Psychopharmacology*, vol. 217, no. 2, pp. 239–253, 2011.

[68] K. S. Kendler, L. M. Karkowski, and C. A. Prescott, "Causal relationship between stressful life events and the onset of major depression," *The American Journal of Psychiatry*, vol. 156, no. 6, pp. 837–841, 1999.

[69] T. L. Bale, "Stress sensitivity and the development of affective disorders," *Hormones and Behavior*, vol. 50, no. 4, pp. 529–533, 2006.

[70] M. V. Schmidt, S. H. Scharf, V. Sterlemann et al., "High susceptibility to chronic social stress is associated with a depression-like phenotype," *Psychoneuroendocrinology*, vol. 35, no. 5, pp. 635–643, 2010.

[71] E. Gould, B. S. McEwen, P. Tanapat, L. A. M. Galea, and E. Fuchs, "Neurogenesis in the dentate gyrus of the adult tree shrew is regulated by psychosocial stress and NMDA receptor activation," *The Journal of Neuroscience*, vol. 17, no. 7, pp. 2492–2498, 1997.

[72] E. Gould, P. Tanapat, B. S. Mcewen, G. Flügge, and E. Fuchs, "Proliferation of granule cell precursors in the dentate gyrus of adult monkeys is diminished by stress," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 95, no. 6, pp. 3168–3171, 1998.

[73] D. Arnone, S. McKie, R. Elliott et al., "State-dependent changes in hippocampal grey matter indepression," *Molecular Psychiatry*. In press.

[74] K. Sawyer, E. Corsentino, N. Sachs-Ericsson, and D. C. Steffens, "Depression, hippocampal volume changes, and cognitive decline in a clinical sample of older depressed outpatients and non-depressed controls," *Aging & Mental Health*, vol. 16, no. 6, pp. 753–762, 2012.

[75] C. E. Sexton, M. Le Masurier, C. L. Allan et al., "Magnetic resonance imaging in late-life depression: vascular and glucocorticoid cascade hypotheses," *The British Journal of Psychiatry*, vol. 201, no. 1, pp. 46–51, 2012.

[76] A. Reif, S. Fritzen, M. Finger et al., "Neural stem cell proliferation is decreased in schizophrenia, but not in depression," *Molecular Psychiatry*, vol. 11, no. 5, pp. 514–522, 2006.

[77] M. Boldrini, M. D. Underwood, R. Hen et al., "Antidepressants increase neural progenitor cells in the human hippocampus," *Neuropsychopharmacology*, vol. 34, no. 11, pp. 2376–2389, 2009.

[78] H. B. van der Worp, D. W. Howells, E. S. Sena et al., "Can animal models of disease reliably inform human studies?" *PLoS Medicine*, vol. 7, no. 3, Article ID e1000245, 2010.

[79] J. Fuss, N. M. B. Ben Abdallah, F. W. Hensley, K. J. Weber, R. Hellweg, and P. Gass, "Deletion of running-induced hippocampal neurogenesis by irradiation prevents development of an anxious phenotype in mice," *PLoS ONE*, vol. 5, no. 9, Article ID 12769, 2010.

[80] M. N. Jayatissa, K. Henningsen, M. J. West, and O. Wiborg, "Decreased cell proliferation in the dentate gyrus does not associate with development of anhedonic-like symptoms in rats," *Brain Research*, vol. 1290, pp. 133–141, 2009.

[81] J. M. Revest, D. Dupret, M. Koehl et al., "Adult hippocampal neurogenesis is involved in anxiety-related behaviors," *Molecular Psychiatry*, vol. 14, no. 10, pp. 959–967, 2009.

[82] J. S. Snyder, A. Soumier, M. Brewer, J. Pickel, and H. A. Cameron, "Adult hippocampal neurogenesis buffers stress responses and depressive behaviour," *Nature*, vol. 476, no. 7361, pp. 458–461, 2011.

[83] G. L. Ming and H. Song, "Adult neurogenesis in the mammalian brain: significant answers and significant questions," *Neuron*, vol. 70, no. 4, pp. 687–702, 2011.

[84] D. H. R. Blackwood, A. Fordyce, M. T. Walker, D. M. St Clair, D. J. Porteous, and W. J. Muir, "Schizophrenia and affective disorders—cosegregation with a translocation at chromosome 1q42 that directly disrupts brain-expressed genes: clinical and P300 findings in a family," *American Journal of Human Genetics*, vol. 69, no. 2, pp. 428–433, 2001.

[85] K. Ishizuka, M. Paek, A. Kamiya, and A. Sawa, "A review of disrupted-in-schizophrenia-1 (DISC1): neurodevelopment, cognition, and mental conditions," *Biological Psychiatry*, vol. 59, no. 12, pp. 1189–1197, 2006.

[86] C. P. Austin, B. Ky, L. Ma, J. A. Morris, and P. J. Shughrue, "Expression of disrupted-in-schizophrenia-1, a schizophrenia-associated gene, is prominent in the mouse hippocampus throughout brain development," *Neuroscience*, vol. 124, no. 1, pp. 3–10, 2004.

[87] I. L. Schurov, E. J. Handford, N. J. Brandon, and P. J. Whiting, "Expression of disrupted in schizophrenia 1 (DISC1) protein in the adult and developing mouse brain indicates its role in neurodevelopment," *Molecular Psychiatry*, vol. 9, no. 12, pp. 1100–1110, 2004.

[88] J. Y. Kim, X. Duan, C. Y. Liu et al., "DISC1 regulates new neuron development in the adult brain via modulation of AKT-mTOR signaling through KIAA1212," *Neuron*, vol. 63, no. 6, pp. 761–773, 2009.

[89] D. Kamnasaran, W. J. Muir, M. A. Ferguson-Smith, and D. W. Cox, "Disruption of the neuronal PAS3 gene in a family affected with schizophrenia," *Journal of Medical Genetics*, vol. 40, no. 5, pp. 325–332, 2003.

[90] B. S. Pickard, A. Christoforou, P. A. Thomson et al., "Interacting haplotypes at the NPAS3 locus alter risk of schizophrenia and bipolar disorder," *Molecular Psychiatry*, vol. 14, no. 9, pp. 874–884, 2009.

[91] M. Nyffeler, U. Meyer, B. K. Yee, J. Feldon, and I. Knuesel, "Maternal immune activation during pregnancy increases limbic GABAA receptor immunoreactivity in the adult offspring: implications for schizophrenia," *Neuroscience*, vol. 143, no. 1, pp. 51–62, 2006.

[92] C. D. Mandyam and G. F. Koob, "The addicted brain craves new neurons: putative role for adult-born progenitors in promoting recovery," *Trends in Neurosciences*, vol. 35, no. 4, pp. 250–260, 2012.

[93] R. C. Pierce and V. Kumaresan, "The mesolimbic dopamine system: the final common pathway for the reinforcing effect of drugs of abuse?" *Neuroscience and Biobehavioral Reviews*, vol. 30, no. 2, pp. 215–238, 2006.

[94] A. J. Eisch, M. Barrot, C. A. Schad, D. W. Self, and E. J. Nestler, "Opiates inhibit neurogenesis in the adult rat hippocampus," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 13, pp. 7579–7584, 2000.

[95] D. N. Abrous, W. Adriani, M. F. Montaron et al., "Nicotine self-administration impairs hippocampal plasticity," *The Journal of Neuroscience*, vol. 22, no. 9, pp. 3656–3662, 2002.

[96] C. D. Mandyam, S. Wee, E. F. Crawford, A. J. Eisch, H. N. Richardson, and G. F. Koob, "Varied access to intravenous methamphetamine self-administration differentially alters adult hippocampal neurogenesis," *Biological Psychiatry*, vol. 64, no. 11, pp. 958–965, 2008.

[97] S. A. Lloyd, Z. R. Balest, F. S. Corotto, and R. J. Smeyne, "Cocaine selectively increases proliferation in the adult murine hippocampus," *Neuroscience Letters*, vol. 485, no. 2, pp. 112–116, 2010.

[98] A. J. Eisch, "Adult neurogenesis: implications for psychiatry," *Progress in Brain Research*, vol. 138, pp. 315–342, 2002.

[99] M. L. Mustroph, D. J. Stobaugh, D. S. Miller, E. K. DeYoung, and J. S. Rhodes, "Wheel running can accelerate or delay extinction of conditioned place preference for cocaine in male C57BL/6J mice, depending on timing of wheel access," *The European Journal of Neuroscience*, vol. 34, no. 7, pp. 1161–1169, 2011.

[100] M. A. Noonan, S. E. Bulin, D. C. Fuller, and A. J. Eisch, "Reduction of adult hippocampal neurogenesis confers vulnerability in an animal model of cocaine addiction," *The Journal of Neuroscience*, vol. 30, no. 1, pp. 304–315, 2010.

[101] I. Agartz, R. Momenan, R. R. Rawlings, M. J. Kerich, and D. W. Hommer, "Hippocampal volume in patients with alcohol dependence," *Archives of General Psychiatry*, vol. 56, no. 4, pp. 356–363, 1999.

[102] O. Bengochea and L. M. Gonzalo, "Effect of chronic alcoholism on the human hippocampus," *Histology and Histopathology*, vol. 5, no. 3, pp. 349–357, 1990.

[103] E. V. Sullivan, L. Marsh, D. H. Mathalon, K. O. Lim, and A. Pfefferbaum, "Anterior hippocampal volume deficits in nonamnesic, aging chronic alcoholics," *Alcoholism*, vol. 19, no. 1, pp. 110–122, 1995.

[104] S. A. Morris, D. W. Eaves, A. R. Smith, and K. Nixon, "Alcohol inhibition of neurogenesis: a mechanism of hippocampal neurodegeneration in an adolescent alcohol abuse model," *Hippocampus*, vol. 20, no. 5, pp. 596–607, 2010.

[105] S. S. Newton, E. F. Collier, J. Hunsberger et al., "Gene profile of electroconvulsive seizures: induction of neurotrophic and angiogenic factors," *The Journal of Neuroscience*, vol. 23, no. 34, pp. 10841–10851, 2003.

[106] M. Nibuya, S. Morinobu, and R. S. Duman, "Regulation of BDNF and trkB mRNA in rat brain by chronic electroconvulsive seizure and antidepressant drug treatments," *The Journal of Neuroscience*, vol. 15, no. 11, pp. 7539–7547, 1995.

[107] K. Nixon, S. A. Morris, D. J. Liput, and M. L. Kelso, "Roles of neural stem cells and adult neurogenesis in adolescent alcohol use disorders," *Alcohol*, vol. 44, no. 1, pp. 39–56, 2010.

[108] M. H. Jang, M. C. Shin, E. H. Kim, and C. J. Kim, "Acute alcohol intoxication decreases cell proliferation and nitric oxide synthase expression in dentate gyrus of rats," *Toxicology Letters*, vol. 133, no. 2-3, pp. 255–262, 2002.

[109] M. Roberto, T. E. Nelson, C. L. Ur, and D. L. Gruol, "Long-term potentiation in the rat hippocampus is reversibly depressed by chronic intermittent ethanol exposure," *Journal of Neurophysiology*, vol. 87, no. 5, pp. 2385–2397, 2002.

[110] L. J. Santín, S. Rubio, A. Begega, and J. L. Arias, "Effects of chronic alcohol consumption on spatial reference and working memory tasks," *Alcohol*, vol. 20, no. 2, pp. 149–159, 2000.

[111] D. W. Walker and G. Freund, "Impairment of shuttle box avoidance learning following prolonged alcohol consumption in rats," *Physiology & Behavior*, vol. 7, no. 5, pp. 773–778, 1971.

[112] M. M. Husain, A. J. Rush, M. Fink et al., "Speed of response and remission in major depressive disorder with acute electroconvulsive therapy (ECT): a consortium for research in ECT (CORE) report," *The Journal of Clinical Psychiatry*, vol. 65, no. 4, pp. 485–491, 2004.

[113] P. G. Janicak, J. M. Davis, R. D. Gibbons et al., "Efficacy of ECT: a meta-analysis," *The American Journal of Psychiatry*, vol. 142, no. 3, pp. 297–302, 1985.

[114] J. L. Warner-Schmidt, T. M. Madsen, and R. S. Duman, "Electroconvulsive seizure restores neurogenesis and hippocampus-dependent fear memory after disruption by irradiation," *European Journal of Neuroscience*, vol. 27, no. 6, pp. 1485–1493, 2008.

[115] C. Stewart and I. Reid, "Electroconvulsive stimulation and synaptic plasticity in the rat," *Brain Research*, vol. 620, no. 1, pp. 139–141, 1993.

[116] E. Bruel-Jungerman, S. Davis, C. Rampon, and S. Laroche, "Long-term potentiation enhances neurogenesis in the adult dentate gyrus," *The Journal of Neuroscience*, vol. 26, no. 22, pp. 5888–5893, 2006.

[117] R. P. Gwinn, A. Kondratyev, and K. Gale, "Time-dependent increase in basic fibroblast growth factor protein in limbic regions following electroshock seizures," *Neuroscience*, vol. 114, no. 2, pp. 403–409, 2002.

[118] A. N. Brooks, E. Kilgour, and P. D. Smith, "Molecular pathways: fibroblast growth factor signaling: a new therapeutic opportunity in cancer," *Clinical Cancer Research*, vol. 18, no. 7, pp. 1855–1862, 2012.

[119] J. M. Hebert, "FGFs: neurodevelopment's Jack-of-all-trades —how do they do it?" *Frontiers in Neuroscience*, vol. 5, p. 133, 2011.

[120] Y. Shirayama, A. C. H. Chen, S. Nakagawa, D. S. Russell, and R. S. Duman, "Brain-derived neurotrophic factor produces antidepressant effects in behavioral models of depression," *The Journal of Neuroscience*, vol. 22, no. 8, pp. 3251–3261, 2002.

[121] M. Elsayed, M. Banasr, V. Duric, N. M. Fournier, P. Licznerski, and R. S. Duman, "Antidepressant effects of fibroblast growth factor-2 in behavioral and cellular models of depression," *Biological Psychiatry*, vol. 72, no. 4, pp. 258–265, 2012.

[122] D. Taliaz, N. Stall, D. E. Dar, and A. Zangen, "Knockdown of brain-derived neurotrophic factor in specific brain sites precipitates behaviors associated with depression and reduces neurogenesis," *Molecular Psychiatry*, vol. 15, no. 1, pp. 80–92, 2010.

[123] M. H. Maurer, W. K. Tripps, R. E. Feldmann Jr., and W. Kuschinsky, "Expression of vascular endothelial growth factor and its receptors in rat neural stem cells," *Neuroscience Letters*, vol. 344, no. 3, pp. 165–168, 2003.

[124] A. M. Lozano and C. Hamani, "The future of deep brain stimulation," *Journal of Clinical Neurophysiology*, vol. 21, no. 1, pp. 68–69, 2004.

[125] A. M. Lozano, H. S. Mayberg, P. Giacobbe, C. Hamani, R. C. Craddock, and S. H. Kennedy, "Subcallosal cingulate gyrus deep brain stimulation for treatment-resistant depression," *Biological Psychiatry*, vol. 64, no. 6, pp. 461–467, 2008.

[126] H. S. Mayberg, A. M. Lozano, V. Voon et al., "Deep brain stimulation for treatment-resistant depression," *Neuron*, vol. 45, no. 5, pp. 651–660, 2005.

[127] B. H. Bewernick, R. Hurlemann, A. Matusch et al., "Nucleus accumbens deep brain stimulation decreases ratings of depression and anxiety in treatment-resistant depression," *Biological Psychiatry*, vol. 67, no. 2, pp. 110–116, 2010.

[128] R. Silva, A. R. Mesquita, J. Bessa et al., "Lithium blocks stress-induced changes in depressive-like behavior and hippocampal cell fate: the role of glycogen-synthase-kinase-3β," *Neuroscience*, vol. 152, no. 3, pp. 656–669, 2008.

[129] M. Sairanen, G. Lucas, P. Ernfors, M. Castrén, and E. Castrén, "Brain-derived neurotrophic factor and antidepressant drugs have different but coordinated effects on neuronal turnover, proliferation, and survival in the adult dentate gyrus," *The Journal of Neuroscience*, vol. 25, no. 5, pp. 1089–1094, 2005.

[130] F. L. Neto, G. Borges, S. Torres-Sanchez, J. A. Mico, and E. Berrocoso, "Neurotrophins role in depression neurobiology: a review of basic and clinical evidence," *Current Neuropharmacology*, vol. 9, no. 4, pp. 530–552, 2011.

[131] T. Saarelainen, P. Hendolin, G. Lucas et al., "Activation of the TrkB neurotrophin receptor is induced by antidepressant drugs and is required for antidepressant-induced behavioral effects," *The Journal of Neuroscience*, vol. 23, no. 1, pp. 349–357, 2003.

[132] Y. Li, B. W. Luikart, S. Birnbaum et al., "TrkB regulates hippocampal neurogenesis and governs sensitivity to antidepressive treatment," *Neuron*, vol. 59, no. 3, pp. 399–412, 2008.

[133] J. L. Warner-Schmidt and R. S. Duman, "VEGF is an essential mediator of the neurogenic and behavioral actions of antidepressants," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 104, no. 11, pp. 4647–4652, 2007.

[134] T. D. Palmer, E. A. Markakis, A. R. Willhoite, F. Safar, and F. H. Gage, "Fibroblast growth factor-2 activates a latent neurogenic program in neural stem cells from diverse regions of the adult CNS," *The Journal of Neuroscience*, vol. 19, no. 19, pp. 8487–8497, 1999.

[135] H. van Praag, B. R. Christie, T. J. Sejnowski, and F. H. Gage, "Running enhances neurogenesis, learning, and long-term potentiation in mice," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 96, no. 23, pp. 13427–13431, 1999.

[136] K. van der Borght, R. Havekes, T. Bos, B. J. L. Eggen, and E. A. van der Zee, "Exercise improves memory acquisition and retrieval in the Y-maze task: relationship with hippocampal neurogenesis," *Behavioral Neuroscience*, vol. 121, no. 2, pp. 324–334, 2007.

[137] K. I. Erickson, M. W. Voss, R. S. Prakash et al., "Exercise training increases size of hippocampus and improves memory," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 108, no. 7, pp. 3017–3022, 2011.

[138] T. W. Scheewe, F. J. Backx, T. Takken et al., "Exercise therapy improves mental and physical health in schizophrenia: a randomised controlled trial," *Acta Psychiatrica Scandinavica*. In press.

[139] P. Bekinschtein, C. A. Oomen, L. M. Saksida, and T. J. Bussey, "Effects of environmental enrichment and voluntary exercise on neurogenesis, learning and memory, and pattern separation: BDNF as a critical variable?" *Seminars in Cell & Developmental Biology*, vol. 22, no. 5, pp. 536–542, 2011.

[140] J. Farmer, X. Zhao, H. van Praag, K. Wodtke, F. H. Gage, and B. R. Christie, "Effects of voluntary exercise on synaptic plasticity and gene expression in the dentate gyrus of adult male sprague-dawley rats in vivo," *Neuroscience*, vol. 124, no. 1, pp. 71–79, 2004.

[141] S. A. Neeper, F. Gómez-Pinilla, J. Choi, and C. W. Cotman, "Physical activity increases mRNA for brain-derived neurotrophic factor and nerve growth factor in rat brain," *Brain Research*, vol. 726, no. 1-2, pp. 49–56, 1996.

[142] M. Bergami, R. Rimondini, S. Santi, R. Blum, M. Götz, and M. Canossa, "Deletion of TrkB in adult progenitors alters newborn neuron integration into hippocampal circuits and increases anxiety-like behavior," *Proceedings of the National*

Academy of Sciences of the United States of America, vol. 105, no. 40, pp. 15570–15575, 2008.

[143] S. Vaynman, Z. Ying, and F. Gomez-Pinilla, "Interplay between brain-derived neurotrophic factor and signal transduction modulators in the regulation of the effects of exercise on synaptic-plasticity," *Neuroscience*, vol. 122, no. 3, pp. 647–657, 2003.

Hindawi Publishing Corporation
Neural Plasticity
Volume 2012, Article ID 283829, 9 pages
doi:10.1155/2012/283829

Research Article

Increases in Doublecortin Immunoreactivity in the Dentate Gyrus following Extinction of Heroin-Seeking Behavior

Megan P. Hicks,[1] Kelly C. Wischerath,[2] Amber L. Lacrosse,[2] and M. Foster Olive[2,3]

[1] *Department of Psychiatry and Behavioral Sciences, Medical University of South Carolina, Charleston, SC 29425, USA*
[2] *Department of Psychology, Arizona State University, 950 S. McAllister Avenue, P.O. Box 871104, Tempe, AZ 85287, USA*
[3] *Interdisciplinary Graduate Program in Neuroscience, Arizona State University, Tempe, AZ 85287, USA*

Correspondence should be addressed to M. Foster Olive, foster.olive@asu.edu

Received 16 June 2012; Revised 3 October 2012; Accepted 6 October 2012

Academic Editor: Chitra D. Mandyam

Adult-generated neurons in the dentate gyrus (DG) of the hippocampus play a role in various forms of learning and memory. However, adult born neurons in the DG, while still at an immature stage, exhibit unique electrophysiological properties and are also functionally implicated in learning and memory processes. We investigated the effects of extinction of drug-seeking behavior on the formation of immature neurons in the DG as assessed by quantification of doublecortin (DCX) immunoreactivity. Rats were allowed to self-administer heroin (0.03 mg/kg/infusion) for 12 days and then subjected either to 10 days of extinction training or forced abstinence. We also examined extinction responding patterns following heroin self-administration in glial fibrillary acidic protein thymidine kinase (GFAP-tk) transgenic mice, which have been previously demonstrated to show reduced formation of immature and mature neurons in the DG following treatment with ganciclovir (GCV). We found that extinction training increased DCX immunoreactivity in the dorsal DG as compared with animals undergoing forced abstinence, and that GCV-treated GFAP-tk mice displayed impaired extinction learning as compared to saline-treated mice. Our results suggest that extinction of drug-seeking behavior increases the formation of immature neurons in the DG and that these neurons may play a functional role in extinction learning.

1. Introduction

Adult-born neurons in the dentate gyrus (DG) of the adult hippocampus are generated from neural progenitor cells (NPCs) located in the subgranular layer of this brain region, which proliferate and differentiate first into immature neurons and subsequently migrate into the granule cell layer as mature neurons where they integrate into existing hippocampal circuits [1]. There are numerous lines of evidence to support the notion that mature adult-born neurons contribute to several forms of hippocampus-dependent learning and memory processes such as spatial navigation, contextual learning, and pattern separation (reviewed in [2–7]). Various neuropsychiatric disorders such as depression, schizophrenia, and drug addiction are characterized by alterations in neurogenesis in the DG [1, 8–10]. In the drug addiction literature, there is general agreement that most drugs of abuse, including opiates, ethanol, and psychostimulants, suppress neurogenesis in the DG [11–16], and it has been suggested that diminished DG neurogenesis may play a causative role in certain cognitive deficits frequently observed in drug addicts, such as maladaptive contextual and episodic memory formation, altered spatial abilities, cognitive inflexibility, and susceptibility to relapse [17–19].

While the majority of research on neurogenesis in the adult DG has focused on factors influencing the proliferation, differentiation, and survival of NPCs, as well as the role of mature adult-born neurons in various cognitive functions, relatively little is known about the function of adult-born neurons that have not yet fully developed into mature granule cells. There are known species-specific time periods (approximately 3–6 weeks in rodents) between NPC proliferation and formation of mature dentate granule cells when adult-born neurons are considered immature. During

this time period, immature adult-born neurons undergo a substantial amount of morphological and physiological maturation and exhibit enhanced synaptic plasticity and lower thresholds for the induction of long-term potentiation (LTP) [20, 21]. During this stage, immature neurons transiently express the microtubule-associated protein doublecortin (DCX) [22, 23], which allows for immunohistochemical detection and quantification of the number of immature neurons in the DG. Recent studies using novel transgenic approaches to selectively ablate immature neurons in the DG have revealed that in fact these neurons mediate spatial memory as well as the extinction of conditioned fear [24]. However, the role of immature DG neurons in extinction of addiction-related behaviors has not yet been explored.

Given that extinction is an established form of new and active learning [25–28], the goal of the present study was designed to explore the possibility that immature DG neurons are involved in the extinction of drug-seeking behavior. In Experiment 1, we sought to determine if extinction of drug-seeking behavior following heroin self-administration would alter DCX immunoreactivity in the DG as compared to animals undergoing forced abstinence. In Experiment 2, we sought to determine if suppression of the generation of immature neurons in the DG would affect extinction responding following heroin self-administration.

2. Material and Methods

2.1. Subjects. Animals were maintained on a 12-hour light-dark cycle (lights off at 07:00 hr) in a temperature- and humidity-controlled room. Animals were given ad libitum access to food and water during all phases of the experiment except during behavioral testing. All experimental procedures were conducted with the approval of the Institutional Animal Care and Use Committee of the Medical University of South Carolina and in accordance with the Principles of Laboratory Animal Care and the Guidelines for the Care and Use of Mammals in Neuroscience and Behavioral Research (National Research Council, 2003). For Experiment 1, male Sprague-Dawley rats (Harlan Laboratories, Livermore, CA, USA) weighing approximately 250–275 g were individually housed upon arrival. For Experiment 2, male GFAP-tk mice (B6.Cg-Tg(Gfap-Tk) 7.1 Mvs/J, stock #005698; Jackson Laboratories, Bar Harbor, ME, USA) weighing approximately 20–25 g were individually housed upon arrival. This mouse strain was generated as described elsewhere [29, 30] to express a herpes simplex virus thymidine kinase (tk) exclusively in GFAP-positive cells, and proliferating cells expressing the transgene in the presence of the antiviral agent ganciclovir (GCV) produce toxic nucleotide analogues that promote cell death. Thus, GFAP-tk mice have been used as a tool for investigating the function of adult-generated neuronal precursors [31]. This strain has been backcrossed from the founder line onto a C57BL/6 background strain for more than 12 generations. Prior to shipment, all animals were implanted with jugular vein catheters by Harlan or Jackson Laboratories Surgical Services. Catheters were filled with a Hep-Lock solution, tunneled subcutaneously to exit the dorsum between the scapulae, plugged with 1 cm segments of stainless steel tubing and secured to skin on the dorsum with wound clips.

2.2. Drugs. Heroin (diacetylmorphine hydrochloride), 5-bromodeoxyuridine (BrDU), and sodium pentobarbital were obtained from Sigma-Aldrich (St. Louis, MO, USA). Heroin was dissolved in sterile saline for intravenous (i.v.) self-administration. BrDU was dissolved in sterile saline at a concentration of 50 mg/mL for intraperitoneal (i.p.) administration and was administered in a volume of 1 mL/kg to yield a final dose of 50 mg/kg per injection. Sodium pentobarbital was dissolved in sterile saline containing 10% v/v ethanol and 40% v/v propylene glycol at a concentration of 150 mg/mL for i.p. administration. Ganciclovir (GCV) was obtained from Waterstone Technology (St. Carmel, IN, USA) and dissolved at a concentration of 10 mg/mL in sterile saline for subcutaneous administration via osmotic minipumps.

2.3. Apparatus. Drug self-administration and extinction training were conducted in operant self-administration chambers (ENV-008 for rats, ENV-307 for mice; Med Associates, St. Albans, VT, USA). Each self-administration chamber was located inside a sound-attenuating cubicle equipped with a house light and an exhaust fan designed to mask external noise and odors and was interfaced to a PC computer. Positioned above each lever was a 2.5 cm diameter white stimulus light. Syringe pumps were located outside self-administration chambers and were interfaced to a PC. Drug solutions were delivered via the syringe pump through a single-channel liquid swivel mounted atop of the chamber, which was connected to a vascular access port.

2.4. Surgical Procedures. Upon arrival, animals were allowed one day of acclimation before surgical procedures. For Experiment 1, rats were anesthetized with isoflurane (2% v/v) vaporized in medical-grade oxygen at a flow rate of 2 L/min. Staples surrounding catheter tubing were removed and a one-inch longitudinal incision was made for implantation of a threaded vascular access port (Plastics One; Roanoke, VA, USA). Access ports were attached to a mesh collar that was sutured underneath the surrounding tissue within the incision and were sealed with a piece of Tygon tubing closed at one end and a protective cap. Following surgical procedures, rats were given 5 days of postoperative care during which they received daily intravenous infusions of 70 U/mL heparin (0.2 mL volume) to maintain catheter patency and 100 mg/mL cefazolin (0.1 mL volume) to protect against infection. Rats also received daily subcutaneous injections of 2.5 mg/mL of meloxicam (0.15 mL volume) for surgery-related discomfort. The surgery site was also treated with topical lidocaine and triple antibiotic ointment to facilitate healing of the wound.

For Experiment 2, mice were anesthetized as described above, the catheter exit wound was enlarged to 1 cm in length, and mice were implanted with sterile osmotic minipumps (Model 1004, pump rate 0.11 μL/hr; Alzet; Cupertino, CA, USA) containing either sterile saline or

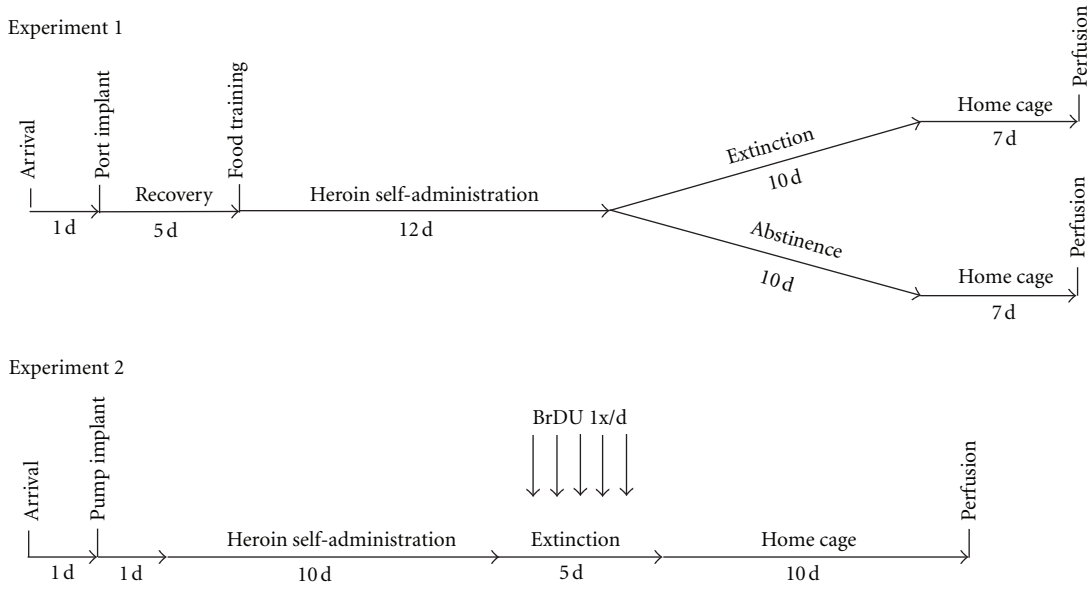


Figure 1: Timelines of procedures in Experiments 1 and 2.

GCV (10 mg/mL) into the dorsum. The catheter was trimmed to extend 1 cm from the exit wound. Following minipump implantation, mice were treated with 10 U/mL heparin (0.05 mL volume) to maintain catheter patency and 10 mg/mL cefazolin (0.05 mL volume) to protect against infection. The surgery site was treated with topical lidocaine and triple antibiotic ointment to facilitate healing of the wound, and mice were allowed 1 day to recover prior to initiation of drug self-administration procedures.

2.5. Heroin Self-Administration. All self-administration procedures were conducted during the dark phase of the light-dark cycle. A timeline of both experiments is provided in Figure 1. In Experiment 1, to initiate operant responding, rats were placed in the self-administration chambers for a single 16 hr overnight training sessions whereby each press on the designated active lever delivered a 45 mg food pellet (Test-diet, Richmond, IN, USA) into a pellet receptacle on a fixed-ratio 1 (FR1) schedule of reinforcement. Each lever press was accompanied by a concurrent illumination of a stimulus light located above the active lever for 2 sec. Each food pellet delivery was followed by a 20 sec time-out period, during which additional active lever presses were recorded but produced no programmed consequences. Presses on the designated inactive lever were recorded but produced no consequences at any time during the experiment. Approximately 24 hr following the initial overnight training session, 3 hr daily heroin self-administration sessions were initiated, whereby presses on the active lever resulted in delivery of heroin (0.03 mg/kg/infusion, delivered in a volume of 0.06 mL over a 2 sec period) on a FR1 schedule of reinforcement. Each drug infusion was followed by a 20 sec time-out period, during which additional active lever presses were recorded but produced no programmed consequences. Each drug infusion was accompanied by concurrent illumination of a stimulus light located above the active lever for 2 sec. Self-administration sessions were conducted 6 consecutive days per week for a total of 12 days. Body weights were assessed every 5–7 days in order to assess potential weight loss as a result of heroin self-administration and to adjust the concentration of heroin in the infusion syringe as needed.

In Experiment 2, heroin self-administration in GFAP-tk mice was commenced on the day following minipump implantation. Procedures were similar to those of Experiment 1, with the following exceptions: (1) food pellet training was not conducted since mouse self-administration chambers were not equipped with pellet dispensers or receptacles; (2) heroin was delivered in a volume of 0.02 mL over a 1 sec period; (3) self-administration sessions were conducted for a total of 10 days. Group body weights were assessed on the day following minipump implantation and again during each of the 5 extinction days in order to assess potential weight loss as a result of heroin self-administration and/or GCV treatment and to adjust the concentration of heroin in the infusion syringe as needed. BrDU was administered at a dose of 50 mg/kg within 1 minute of the end of each of the 5 extinction sessions.

2.6. Extinction Procedures. Extinction sessions were 3 hr in length during which responding on the lever that previously delivered heroin no longer produced any programmed consequences. In addition, the light cue that previously accompanied reinforcer delivery was not available during extinction sessions. Animals were subjected to extinction training to achieve a target level of extinction responding equivalent to 25% of the number of active lever presses that were emitted on the average of the last 2 days of active heroin self-administration. Since the majority of extinction of responding on the active lever in Experiment 1 was observed during the first 5 extinction training sessions,

only 5 extinction sessions were conducted in Experiment 2. Immediately following each of the 5 extinction training sessions, mice were injected with BrDU (50 mg/kg i.p.) to provide an indicator of suppression of the formation of immature neurons.

In Experiment 1, animals undergoing forced abstinence instead of extinction training were transported to a neutral room at approximately the same time of day as when extinction sessions were conducted, handled briefly, and left undisturbed in the home cage for 3 hr (equivalent to the length of extinction sessions). Rats were then returned to the colony room. This process was repeated daily for a total of 10 days to parallel the number of days that extinction training was conducted.

2.7. Assessment of DCX Immunoreactivity. In Experiment 1, rats were anesthetized with sodium pentobarbital (150 mg/kg i.p.) 7 days following the final extinction training session (or on the same day for animals undergoing forced abstinence—see Figure 1) and perfused transcardially with 100 mL of phosphate-buffered saline (PBS, pH = 7.4) containing 0.1% w/v heparin followed by 200 mL of 4% w/v paraformaldehyde (pH = 7.4) in 0.1 M PBS. Brains were removed, postfixed in the fixative solution for 24 hr at 4°C, cryoprotected in 30% w/v sucrose in PBS for 48 hr at 4°C, cut into 30 μm coronal sections along the hippocampal neuraxis), and processed for free-floating DCX immunohistochemistry. Sections were preblocked for 1 hr in PBS containing 0.1% Tween 20, 1 M glycine, and 5% w/v donkey serum, followed by overnight incubation with rabbit anti-DCX primary antisera (1 : 500; Abcam; Cambridge, MA, USA) at 4°C. On the next day, sections were washed, incubated with DyLight 594-conjugated donkey anti-rabbit secondary antisera (1 : 500; Jackson ImmunoResearch; West Grove, PA, USA), washed, mounted onto microscope slides, and coverslipped with Prolong antifade mounting medium (Invitrogen; Carlsbad, CA, USA). Slides were stored in darkness until quantification, which was performed under epifluorescence microscopy at 400x magnification (Leica Microsystems; Bannockburn, IL, USA) by an investigator blind to experimental condition. Manual rotation of the fine focus in the z-place was performed to verify the presence of DCX-positive cell bodies. The number of DCX immunoreactive neurons in the subgranular layer of the DG was quantified in one hemisphere from −1.8 to −6.4 mm posterior to bregma to the nearest 0.2 mm according to [32]. Total DCX counts throughout the hippocampal neuraxis were calculated by summation of all counts at each coronal level of section for each individual animal. Images of DCX immunoreactivity in the DG that are presented in Figure 2 were obtained on a Zeiss LSM 410 confocal microscope.

Immunohistochemical staining of tissue from GFAP-tk mice for DCX immunoreactivity yielded inadequate and unsatisfactory staining patterns, as evidenced by a lack of staining of dendritic processes in the granule cell layer. Use of primary antisera from a different vendor also produced unsatisfactory results. We therefore attempted to verify reductions in neuron production in the DG of these mice induced by GCV using previously published procedures for BrDU immunohistochemistry [33] throughout the mouse hippocampal neuraxis.

2.8. Statistical Analyses. Active and inactive lever press data were analyzed by a one-way ANOVA followed by the Holm-Sidak post hoc tests. DCX immunoreactivity quantified along the hippocampal neuraxis was analyzed by a two-way ANOVA (with the treatment group as the intersubject variable and plane of section as the intrasubject variable) followed by the Holm-Sidak post hoc tests. Total DCX counts in animals undergoing extinction versus abstinence, as well as body weights of mice treated with either saline or GCV, were analyzed by one-way ANOVA. The level of statistical significance was set to $P < 0.05$ for all tests.

3. Results

The timelines for behavioral procedures in Experiments 1 and 2 are presented in Figure 1. Heroin self-administration data and the effects of extinction versus abstinence on DCX immunoreactivity are depicted in Figure 2. No differences in the number of active or inactive lever presses during active heroin self-administration were observed between animals that subsequently underwent extinction or abstinence ($P > 0.05$). In addition, the number of heroin infusions obtained in each session did not differ between groups ($P > 0.05$, data not shown), indicating that heroin intake was equivalent prior to extinction. Animals subjected to extinction training displayed significant changes in active lever pressing behavior across extinction sessions ($F(10, 133) = 30.2$, $P < 0.001$; Figure 2(a)). Presses on the active lever were significantly increased on the first day of extinction as compared to the average of the last 2 days of active heroin self-administration, characteristic of an expected "extinction burst" following removal of heroin as the reinforcer. After the 2nd day of extinction, lever pressing declined and was significantly lower during the 3rd through 10th extinction sessions as compared to the average of the last two days of self-administration. The number of inactive lever presses did not differ across experimental groups and also did not change across extinction sessions (P's > 0.05; Figure 2(b)). The lack of reduction of inactive lever presses produced by extinction training was likely due to a floor effect since the mean number of inactive lever presses per session during the final two days of heroin self-administration was less than 5 for the two groups.

Analysis of DCX immunoreactivity along the hippocampal neuraxis in animals that underwent either extinction training or abstinence following heroin self-administration revealed that extinction-trained animals showed significantly higher levels of DCX immunoreactivity ($F(1, 208) = 21.95$, $P < 0.001$; Figures 2(c)–2(e)) in dorsal regions of the DG ranging from 3.0 to 4.0 mm posterior to bregma. No group differences were detected in ventral/posterior regions of the DG. In addition, total DCX counts throughout all sections analyzed were higher ($F(1, 13) = 8.19$, $P < 0.05$) in animals

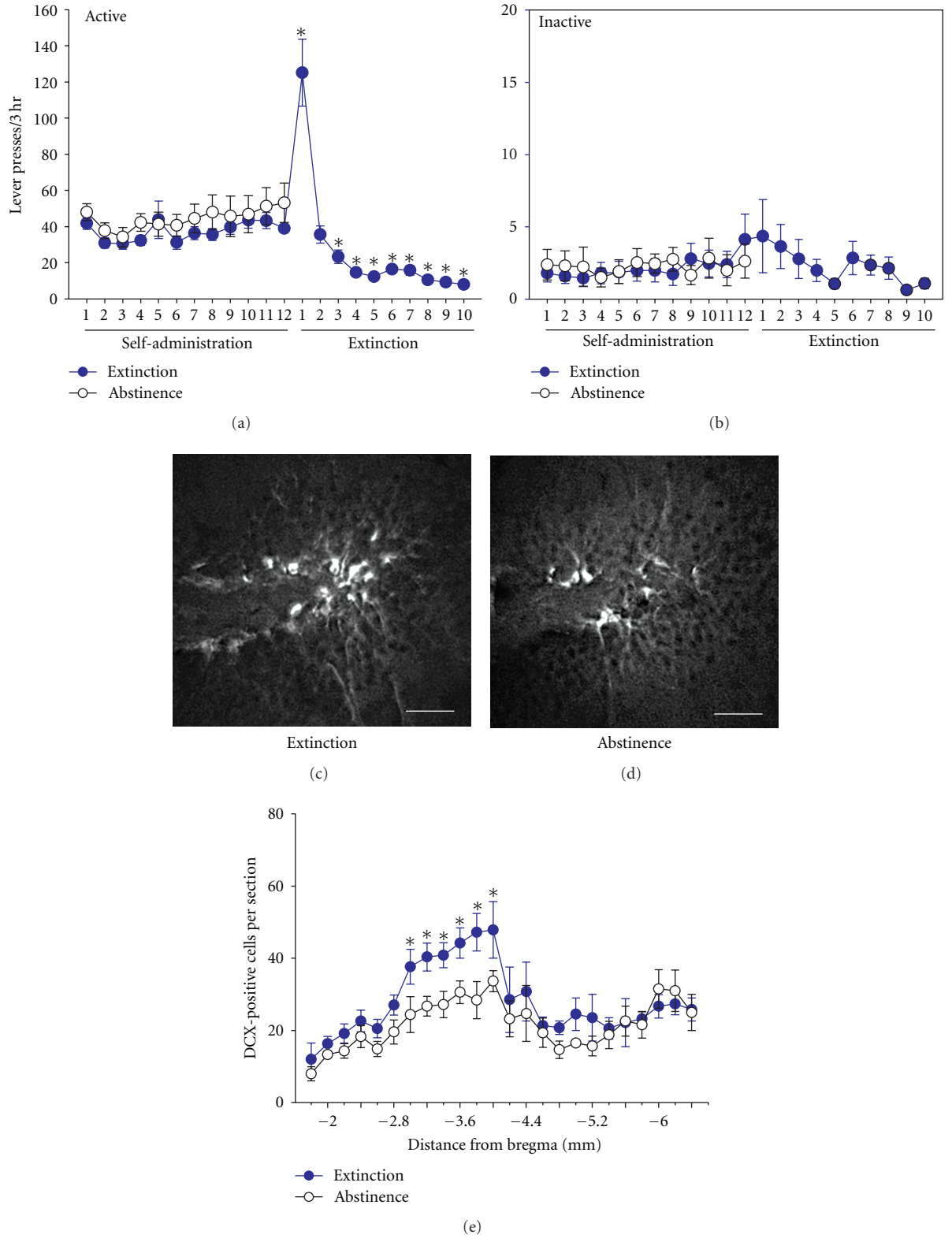


Figure 2: Extinction training increases DCX immunoreactivity in the DG following heroin self-administration (Experiment 1). (a) Active and (b) inactive lever presses during heroin self-administration and extinction. Animals undergoing extinction ($n = 15$) were subjected to 10 days of extinction training following the last day of heroin self-administration, while animals undergoing forced abstinence ($n = 13$) remained in the home cage. $^*P < 0.05$ versus the average of the last two days of heroin self-administration. (c) and (d) Representative DCX immunostaining at 400x magnification in the DG (~3.8 mm posterior to bregma) from animals subjected to extinction (c) or abstinence (d). Scale bar = 35 μm. (e) Quantification of DCX immunoreactivity along the DG neuraxis in a subset of animals subjected to extinction or abstinence ($n = 8$ per group). $^*P < 0.05$ versus the abstinence group at the same coronal plane relative to bregma.

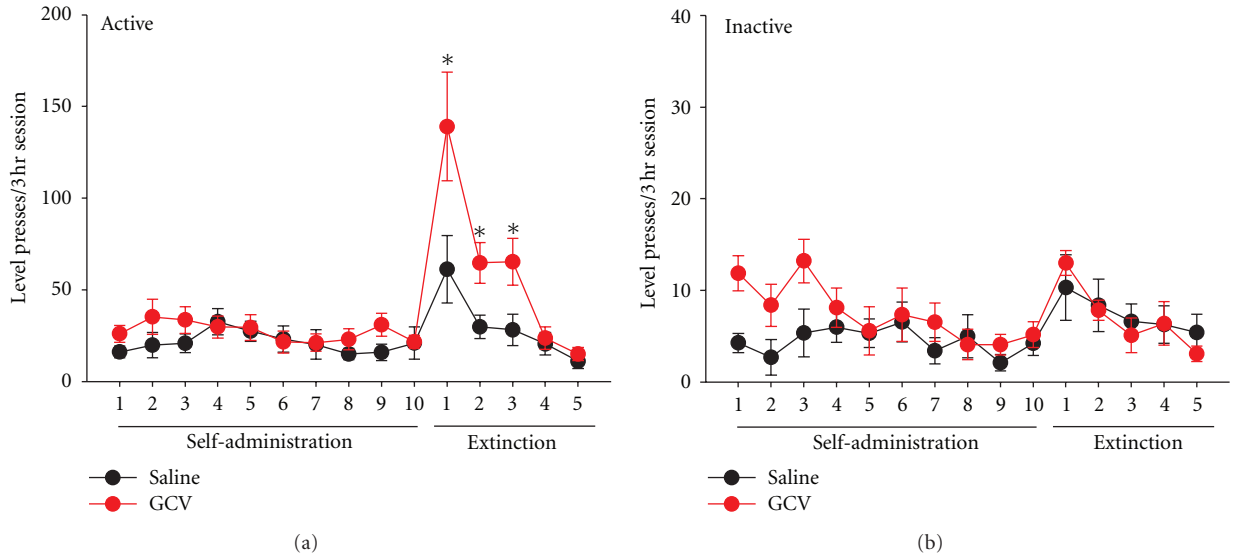


FIGURE 3: Heroin self-administration and extinction in GFAP-tk mice implanted with minipumps containing either saline or GCV. (a) Active lever presses during 10 days of heroin self-administration followed by 5 days of extinction. Mice treated with GCV ($n = 11$) displayed increased extinction responding during the first 3 extinction days (E1–E3) as compared to mice that were treated with saline ($n = 9$). $^*P < 0.05$ versus saline-treated mice on the same day of extinction. (b) Inactive lever presses during 10 days of heroin self-administration followed by 5 days of extinction.

that underwent extinction (502.4 ± 45.6, mean ± SEM) than those that underwent abstinence (357.2 ± 25.8, mean ± SEM).

The results of Experiment 2 are depicted in Figure 3. No differences in lever pressing during the 10 days of active heroin self-administration were noted in GFAP-tk mice receiving saline or GCV ($P > 0.05$; Figure 3(a)) or in the number of heroin infusions obtained ($P > 0.05$, data not shown) indicating equal levels of heroin intake between the two groups. Levels of heroin intake in these GFAP-tk mice were similar to those reported previously in Balb/c mice [34]. On the first 3 days of extinction, GCV-treated mice displayed an increased number of presses on the lever that previously delivered heroin ($F(1, 226) = 5.8$, $P < 0.05$; Figure 3(a)), suggesting an impairment in extinction learning. No differences between treatment groups were noted for inactive lever presses across any phase of the experiment ($P > 0.05$; Figure 3(b)). Similarly, analysis of body weight data before and after heroin self-administration revealed neither weight loss nor weight gain ($P > 0.05$), suggesting that heroin self-administration and/or GCV treatment did not produce nonspecific effects on food intake in GFAP-tk mice.

As mentioned earlier, use of primary antisera against DCX from two different commercial vendors produced inadequate and unsatisfactory staining patterns. Therefore, to verify the GCV-induced reduction of newly proliferating cells in the DG of GFAP-tk mice, immunohistochemistry for BrDU was performed. The number of BrDU-positive cells per section was significantly lower in mice treated with GCV (5.7 ± 0.6, mean ± SEM) as compared with those treated with saline (14.1 ± 0.8, mean ± SEM) ($F(1, 45) = 74.2$, $P < 0.001$).

4. Discussion

In Experiment 1, we demonstrated that extinction training procedures following heroin self-administration produced increased levels of DCX immunoreactivity in the DG, primarily in more rostral/dorsal regions, as compared to animals that underwent abstinence. Extinction of drug-seeking behavior in rodent models of addiction is typically produced by withholding drug reinforcers and associated cues when animals are performing the same operant task that previously resulted in drug infusions and cue presentation. Although we did not assess the effects of extinction on the production of mature neurons in the DG, our findings are in line with those of Rapanelli and colleagues [35], who found that subjecting rats to an operant conditioning task resulted in increased numbers of mature neurons in the dorsal DG.

In Experiment 2, we found that impairment of cell proliferation in the DG by administration of GCV to GFAP-tk mice resulted in increases in extinction responding during the first 3 sessions following cessation of heroin availability. Similarly, Noonan and colleagues [36] have previously demonstrated that ablation of both mature and immature neurons in the DG by hippocampal irradiation resulted in increased responding during the first 3 extinction sessions following cocaine self-administration. Unfortunately, however, the present study did not assess the impact of GCV treatment on formation of immature versus mature neurons, and only nonspecific quantification of the number of BrDU-positive cells as an indicator of cell proliferation was performed. Nonetheless the short time frame of GCV administration (10 days followed by commencement of extinction training on the following day) and the known

amount of time required for the formation of mature neurons in the adult mouse DG (>4 weeks, see [37, 38]), our findings suggest that immature neurons in the DG may play a role in the process of extinction learning. Additional studies using transgenic lines of mice that more precisely allow the examination of the function on DG immature neurons, such as nestin-tk mice [24], are clearly warranted to confirm this.

We noted that extinction training following heroin self-administration increased DCX immunoreactivity primarily in dorsal regions of the DG, while no such effects were observed in the ventral DG. Numerous lines of evidence suggest that dorsal regions of the hippocampus mediate drug-seeking behavior [39–41], although the precise role of adult-born neurons specifically in the DG, both immature and mature, has not yet been fully defined [17–19]. In contrast to our findings, other investigators have reported that cocaine self-administration followed by either continued drug intake or withdrawal results in preferential increases in DCX immunoreactivity in posterior/ventral regions of the DG [42]. The reasons underlying our observations of extinction-induced increases in DCX immunoreactivity in dorsal DG regions are currently unclear but may be related to the functional dichotomy of dorsal versus ventral areas of the hippocampus, with more dorsal regions mediating spatial information processing and more posterior/ventral regions mediating limbic functions such as emotionality [43]. Future studies employing methods to manipulate immature DG neurons with precise anatomical specificity are needed to resolve this issue.

There are several limitations of the present study. First, the number of DCX-positive neurons per coronal section in Experiment 1 is lower than those that have been reported by others [36, 42, 44]. There are numerous factors that could have contributed to these differences, such as opiate-induced suppression of NPC proliferation or survival [12, 45–47], different antigens to which DCX primary antisera were raised, and different methods of quantification (stereology versus manual counting). Second, due to technical problems with utilizing DCX antisera in mouse tissue, we were unable to quantify the degree to which GCV suppressed the formation of DCX-immunoreactive immature neurons in the DG, and only a nonspecific assessment of the number of proliferating cells by BrDU immunohistochemistry was performed. Clearly, future studies should be conducted to assess changes in a range of markers of NPC proliferation, differentiation, maturation, and cell death in the DG that occur as a result of extinction training. Third, both mice and rats self-administered heroin under limited access conditions (3 hr/day) and were not likely opiate dependent, which is typically achieved under conditions of extended daily access (6+ hr/day). The effects of extinction training on DCX immunoreactivity in the DG in heroin-dependent animals is an avenue worthy of further exploration.

In summary, we present evidence that extinction training following heroin self-administration increases the number of DCX-positive immature neurons selectively in the dorsal regions of the DG. We also present evidence that inhibition of cell proliferation in the DG increases responding during extinction, which is indicative of impaired extinction learning. These findings lend further support to the notion that immature neurons in the DG may play a functional role in certain learning and memory processes, including the extinction of drug-seeking behavior.

Acknowledgment

This work was supported by the Public Health Service Grant DA024355 from the National Institute on Drug Abuse.

References

[1] D. T. Balu and I. Lucki, "Adult hippocampal neurogenesis: regulation, functional implications, and contribution to disease pathology," *Neuroscience and Biobehavioral Reviews*, vol. 33, no. 3, pp. 232–252, 2009.

[2] G. L. Ming and H. Song, "Adult neurogenesis in the mammalian brain: significant answers and significant questions," *Neuron*, vol. 70, no. 4, pp. 687–702, 2011.

[3] M. Koehl and D. N. Abrous, "A new chapter in the field of memory: adult hippocampal neurogenesis," *European Journal of Neuroscience*, vol. 33, no. 6, pp. 1101–1114, 2011.

[4] W. Deng, J. B. Aimone, and F. H. Gage, "New neurons and new memories: how does adult hippocampal neurogenesis affect learning and memory?" *Nature Reviews Neuroscience*, vol. 11, no. 5, pp. 339–350, 2010.

[5] B. Leuner, E. Gould, and T. J. Shors, "Is there a link between adult neurogenesis and learning?" *Hippocampus*, vol. 16, no. 3, pp. 216–224, 2006.

[6] C. Zhao, W. Deng, and F. H. Gage, "Mechanisms and functional implications of adult neurogenesis," *Cell*, vol. 132, no. 4, pp. 645–660, 2008.

[7] M. F. Olive, *Drug Addiction and Adult Neurogenesis*, Research Signpost/Transworld Research Network Publishers, Karala, India, 2011.

[8] A. J. Eisch, H. A. Cameron, J. M. Encinas, L. A. Meltzer, G. L. Ming, and L. S. Overstreet-Wadiche, "Adult neurogenesis, mental health, and mental illness: hope or hype?" *Journal of Neuroscience*, vol. 28, no. 46, pp. 11785–11791, 2008.

[9] N. A. DeCarolis and A. J. Eisch, "Hippocampal neurogenesis as a target for the treatment of mental illness: a critical evaluation," *Neuropharmacology*, vol. 58, no. 6, pp. 884–893, 2010.

[10] G. Kempermann, J. Krebs, and K. Fabel, "The contribution of failing adult hippocampal neurogenesis to psychiatric disorders," *Current Opinion in Psychiatry*, vol. 21, no. 3, pp. 290–295, 2008.

[11] A. Kubesova, V. Bubenikova-Valesova, M. Mertlova, T. Palenicek, and J. Horacek, "Impact of psychotropic drugs on adult hippocampal neurogenesis," *Neuroscience Research*, vol. 73, no. 2, pp. 93–98, 2012.

[12] A. J. Eisch, M. Barrot, C. A. Schad, D. W. Self, and E. J. Nestler, "Opiates inhibit neurogenesis in the adult rat hippocampus," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 97, no. 13, pp. 7579–7584, 2000.

[13] A. J. Eisch and G. C. Harburg, "Opiates, psychostimulants, and adult hippocampal neurogenesis: insights for addiction and stem cell biology," *Hippocampus*, vol. 16, no. 3, pp. 271–286, 2006.

[14] K. Nixon, "Alcohol and adult neurogenesis: roles in Neurodegeneration and recovery in chronic alcoholism," *Hippocampus*, vol. 16, no. 3, pp. 287–295, 2006.

[15] T. A. Powrozek, Y. Sari, R. P. Singh, and F. C. Zhou, "Neurotransmitters and substances of abuse: effects on adult neurogenesis," *Current Neurovascular Research*, vol. 1, no. 3, pp. 251–260, 2004.

[16] A. Venkatesan, A. Nath, G. L. Ming, and H. Song, "Adult hippocampal neurogenesis: regulation by HIV and drugs of abuse," *Cellular and Molecular Life Sciences*, vol. 64, no. 16, pp. 2120–2132, 2007.

[17] J. J. Canales, "Adult neurogenesis and the memories of drug addiction," *European Archives of Psychiatry and Clinical Neuroscience*, vol. 257, no. 5, pp. 261–270, 2007.

[18] J. J. Canales, "Comparative neuroscience of stimulant-induced memory dysfunction: role for neurogenesis in the adult hippocampus," *Behavioural Pharmacology*, vol. 21, no. 5-6, pp. 379–393, 2010.

[19] C. D. Mandyam and G. F. Koob, "The addicted brain craves new neurons: putative role for adult-born progenitors in promoting recovery," *Trends in Neurosciences*, vol. 35, no. 4, pp. 250–260, 2012.

[20] C. Schmidt-Hieber, P. Jones, and J. Bischofberger, "Enhanced synaptic plasticity in newly generated granule cells of the adult hippocampus," *Nature*, vol. 429, no. 6988, pp. 184–187, 2004.

[21] S. Ge, C. H. Yang, K. S. Hsu, G. L. Ming, and H. Song, "A critical period for enhanced synaptic plasticity in newly generated neurons of the adult brain," *Neuron*, vol. 54, no. 4, pp. 559–566, 2007.

[22] J. P. Brown, S. Couillard-Després, C. M. Cooper-Kuhn, J. Winkler, L. Aigner, and H. G. Kuhn, "Transient expression of doublecortin during adult neurogenesis," *Journal of Comparative Neurology*, vol. 467, no. 1, pp. 1–10, 2003.

[23] O. Von Bohlen Und Halbach, "Immunohistological markers for staging neurogenesis in adult hippocampus," *Cell and Tissue Research*, vol. 329, no. 3, pp. 409–420, 2007.

[24] W. Deng, M. D. Saxe, I. S. Gallina, and F. H. Gage, "Adult-born hippocampal dentate granule cells undergoing maturation modulate learning and memory in the brain," *Journal of Neuroscience*, vol. 29, no. 43, pp. 13532–13542, 2009.

[25] K. M. Myers and W. A. Carlezon, "Extinction of drug- and withdrawal-paired cues in animal models: relevance to the treatment of addiction," *Neuroscience and Biobehavioral Reviews*, vol. 35, no. 2, pp. 285–302, 2010.

[26] M. E. Bouton and D. Swartzentruber, "Sources of relapse after extinction in Pavlovian and instrumental learning," *Clinical Psychology Review*, vol. 11, no. 2, pp. 123–140, 1991.

[27] K. M. Kantak and B. A. Nic Dhonnchadha, "Pharmacological enhancement of drug cue extinction learning: translational challenges," *Annals of the New York Academy of Sciences*, vol. 1216, no. 1, pp. 122–137, 2011.

[28] M. E. Bouton, "A learning theory perspective on lapse, relapse, and the maintenance of behavior change," *Health Psychology*, vol. 19, no. 1, pp. 57–63, 2000.

[29] T. G. Bush, T. C. Savidge, T. C. Freeman et al., "Fulminant jejuno-ileitis following ablation of enteric gila in adult transgenic mice," *Cell*, vol. 93, no. 2, pp. 189–201, 1998.

[30] M. V. Sofroniew, T. G. Bush, N. Blumauer, Lawrence Kruger, L. Mucke, and M. H. Johnson, "Genetically-targeted and conditionally-regulated ablation of astroglial cells in the central, enteric and peripheral nervous systems in adult transgenic mice," *Brain Research*, vol. 835, no. 1, pp. 91–95, 1999.

[31] A. D. R. Garcia, N. B. Doan, T. Imura, T. G. Bush, and M. V. Sofroniew, "GFAP-expressing progenitors are the principal source of constitutive neurogenesis in adult mouse forebrain," *Nature Neuroscience*, vol. 7, no. 11, pp. 1233–1241, 2004.

[32] G. Paxinos and C. Watson, *The Rat Brain in Stereotaxic Coordinates*, Academic Press, San Diego, Calif, USA, 2007.

[33] J. M. Wojtowicz and N. Kee, "BrdU assay for neurogenesis in rodents," *Nature Protocols*, vol. 1, no. 3, pp. 1399–1405, 2006.

[34] A. J. Roberts, I. Y. Polis, and L. H. Gold, "Intravenous self-administration of heroin, cocaine, and the combination in Balb/c mice," *European Journal of Pharmacology*, vol. 326, no. 2-3, pp. 119–125, 1997.

[35] M. Rapanelli, L. R. Frick, and B. S. Zanutto, "Learning an operant conditioning task differentially induces gliogenesis in the medial prefrontal cortex and neurogenesis in the hippocampus," *PLoS ONE*, vol. 6, no. 2, Article ID e14713, 2011.

[36] M. A. Noonan, S. E. Bulin, D. C. Fuller, and A. J. Eisch, "Reduction of adult hippocampal neurogenesis confers vulnerability in an animal model of cocaine addiction," *Journal of Neuroscience*, vol. 30, no. 1, pp. 304–315, 2010.

[37] M. S. Rao and A. K. Shetty, "Efficacy of doublecortin as a marker to analyse the absolute number and dendritic growth of newly generated neurons in the adult dentate gyrus," *European Journal of Neuroscience*, vol. 19, no. 2, pp. 234–246, 2004.

[38] J. S. Snyder, J. S. Choe, M. A. Clifford et al., "Adult-born hippocampal neurons are more numerous, faster maturing, and more involved in behavior in rats than in mice," *Journal of Neuroscience*, vol. 29, no. 46, pp. 14484–14495, 2009.

[39] R. A. Fuchs, K. A. Evans, C. C. Ledford et al., "The role of the dorsomedial prefrontal cortex, basolateral amygdala, and dorsal hippocampus in contextual reinstatement of cocaine seeking in rats," *Neuropsychopharmacology*, vol. 30, no. 2, pp. 296–309, 2005.

[40] R. A. Fuchs, J. L. Eaddy, Z. I. Su, and G. H. Bell, "Interactions of the basolateral amygdala with the dorsal hippocampus and dorsomedial prefrontal cortex regulate drug context-induced reinstatement of cocaine-seeking in rats," *European Journal of Neuroscience*, vol. 26, no. 2, pp. 487–498, 2007.

[41] D. R. Ramirez, G. H. Bell, H. C. Lasseter, X. Xie, S. A. Traina, and R. A. Fuchs, "Dorsal hippocampal regulation of memory reconsolidation processes that facilitate drug context-induced cocaine-seeking behavior in rats," *European Journal of Neuroscience*, vol. 30, no. 5, pp. 901–912, 2009.

[42] M. A. Noonan, K. H. Choi, D. W. Self, and A. J. Eisch, "Withdrawal from cocaine self-administration normalizes deficits in proliferation and enhances maturity of adult-generated hippocampal neurons," *Journal of Neuroscience*, vol. 28, no. 10, pp. 2516–2526, 2008.

[43] A. Sahay and R. Hen, "Adult hippocampal neurogenesis in depression," *Nature Neuroscience*, vol. 10, no. 9, pp. 1110–1115, 2007.

[44] A. Sankararaman, I. Masiulis, D. R. Richardson, J. M. Andersen, J. Mørland, and A. J. Eisch, "Methadone does not alter key parameters of adult hippocampal neurogenesis in the heroin-naïve rat," *Neuroscience Letters*, vol. 516, no. 1, pp. 99–104, 2012.

[45] L. Kahn, G. Alonso, E. Normand, and O. J. Manzoni, "Repeated morphine treatment alters polysialylated neural cell adhesion molecule, glutamate decarboxylase-67 expression and cell proliferation in the adult rat hippocampus," *European Journal of Neuroscience*, vol. 21, no. 2, pp. 493–500, 2005.

[46] A. A. Arguello, S. J. Fischer, J. R. Schonborn, R. W. Markus, R. A. Brekken, and A. J. Eisch, "Effect of chronic morphine on the dentate gyrus neurogenic microenvironment," *Neuroscience*, vol. 159, no. 3, pp. 1003–1010, 2009.

[47] A. A. Arguello, G. C. Harburg, J. R. Schonborn, C. D. Mandyam, M. Yamaguchi, and A. J. Eisch, "Time course of morphine's effects on adult hippocampal subgranular zone reveals preferential inhibition of cells in S phase of the cell cycle and a subpopulation of immature neurons," *Neuroscience*, vol. 157, no. 1, pp. 70–79, 2008.

Hindawi Publishing Corporation
Neural Plasticity
Volume 2012, Article ID 832757, 12 pages
doi:10.1155/2012/832757

Review Article

Effects of Antipsychotics on Dentate Gyrus Stem Cell Proliferation and Survival in Animal Models: A Critical Update

Gerburg Keilhoff,[1] Paolo Fusar-Poli,[2] and Axel Becker[3]

[1] *Institute of Biochemistry and Cell Biology, University of Magdeburg, Leipziger Straße 44, 39120 Magdeburg, Germany*
[2] *Institute of Psychiatry, King's College London, 5th Floor, Main Building, P.O. Box 63, De Crespigny Park, London SE5 8AF, UK*
[3] *Institute of Pharmacology and Toxicology, University of Magdeburg, Leipziger Straße 44, 39120 Magdeburg, Germany*

Correspondence should be addressed to Gerburg Keilhoff, gerburg.keilhoff@med.ovgu.de

Received 12 June 2012; Revised 19 September 2012; Accepted 20 September 2012

Academic Editor: Chitra D. Mandyam

Schizophrenia is a complex psychiatric disorder. Although a number of different hypotheses have been developed to explain its aetiopathogenesis, we are far from understanding it. There is clinical and experimental evidence indicating that neurodevelopmental factors play a major role. Disturbances in neurodevelopment might result in alterations of neuroanatomy and neurochemistry, leading to the typical symptoms observed in schizophrenia. The present paper will critically address the neurodevelopmental models underlying schizophrenia by discussing the effects of typical and atypical antipsychotics in animal models. We will specifically discuss the vitamin D deficiency model, the poly I:C model, the ketamine model, and the postnatal ventral hippocampal lesion model, all of which reflect core neurodevelopmental issues underlying schizophrenia onset.

1. Introduction

Schizophrenia is a complex psychiatric disorder which is characterized by a defined set of symptoms usually grouped into positive symptoms, negative symptoms, cognitive impairment, psychosocial impairments, and poor quality of life. It is commonly described as a developmental disorder, with onset in the early adulthood or adolescence and involving several genetic and environmental factors. The causes of schizophrenia are unknown. However, several hypotheses have been tested in the recent research. One of the most accepted theories is the "*two hit hypothesis*." Such a hypothesis proposes that an early disturbance is necessary but not sufficient to cause an increased vulnerability to schizophrenia. Thus, an early neurodevelopmental insult is requested to interact with either normal or abnormal postpubertal brain maturation to fully produce late neurodevelopmental brain structural and functional changes [1–4].

Evidence indicating a neurodevelopmental origin of schizophrenia is grounded on extensive research performed over the past two decades. In particular, a novel approach has allowed clinicians to specifically investigate the early phases of psychosis and to clarify the mechanisms underlying the onset of the illness. This approach has been variably termed as ultrahigh risk, at risk mental state, or clinical high risk [5]. This putatively prodromal psychotic phase is associated with an enhanced risk of developing the illness as compared to the general population (1%), ranging from 18% at six months up to 36% after three years [6]. The majority (73%) of the individuals developing a psychotic illness will transit towards a schizophrenia spectrum disorder [7]. The clinical high risk state for psychosis is also characterized by significant cognitive impairments [8] and deficits in social functioning and quality of life [5]. These alterations are associated with underlying neurodevelopmental abnormalities in the structure [9–11], function [12–14], connectivity [15], and neurochemistry [16–18] of the brain, resembling those observed in the established phase of the illness [19]. Interest in this area has exponentially grown to the extent that a new diagnostic category is being discussed in the forthcoming DSM-5 [20]. These findings taken together indicated that schizophrenia is characterized by dynamic neurobiological changes since its earliest phases. In theory, the early phases of schizophrenia can thus be particularly amenable to

treatments that can impact the underlying neurobiology, including antipsychotics. The present paper will critically address this point, focusing on the role played by the effect of antipsychotics on the neurogenesis during the onset of schizophrenia. These issues will be discussed in the light of the recent advances in animal models.

2. Methodological Approach

In the following sections, we will provide an update on the effects of antipsychotics on cell proliferation in animal models used in schizophrenia research. This critical paper is limited to models most traditionally employed in the laboratories, whereby all models reflect neurodevelopmental aspects. In Table 1, a selection of most relevant publications in the last 13 years is given. A survey of effects of neuroleptics on hippocampal neurogenesis is provided in Table 2.

3. Modelling Schizophrenia with Animal Paradigms

The development of animal models is a crucial issue in biological psychiatry for the study of alterations in neurochemistry, neuroanatomy, and behaviours resembling those observed in schizophrenia. Similarly, it can be useful for the discovery and development of effective treatments such as antipsychotic molecules. However, heterogeneity of the clinical symptoms of schizophrenia and the incomplete knowledge about the cause and progression of the illness make the development of valid animal models particularly difficult. Moreover, there is scepticism as to what extent the behaviour of animals can actually reflect highly complex disorders such as schizophrenia. Since each animal model is developed to target only each specific domain of schizophrenia, different complementary approaches are usually needed. Therefore (i) developmental, (ii) genetic, and (iii) pharmacological models have been used in experimental schizophrenia research [31–38].

(i) The developmental hypothesis and the respective animal models proceed from the assumption that malformations at very early stages of neurogenesis result in structural abnormalities of the adult brain [39–41]. Under this scenario, the pathogenesis of schizophrenia onset is attributed to abnormal neuronal development and/or reorganization of neuronal circuits in the frontal cortex or in limbic structures. Hippocampal volume reductions [42, 43], hippocampal shape deformation, or abnormalities in the hippocampal cell density [44] have been consistently reported. Many of these alterations, however, are essentially based on genetic deficits impacting the brain development (see below).

(ii) Genetic animal models are developed by translation of human genetic mutations into animals (for review: [33, 45–47]). They include whole-body mutant mouse strains ([48] (Reelin); [49] (Neuregulin-1)), mutant mouse strains in which distinct genes have been knocked down in a tissue- or cell-type-specific manner ([50] (NRG-1/ErbB); [51] (DISC)), and transgenic mice that overexpress schizophrenia-relevant genes ([52] (dopamine D2 receptor); [53] (Neuregulin-1)). More advanced and complex models, however, are under development. These models combine several environmental and/or genetic factors to better account for the complex aetiology of schizophrenia [54]. For example, combined genetic disruption of the NMDA receptor subunit 1 [55–58], the dopamine D2 and D4 receptors, the dopamine transporter [59–61], and, the mutants in the dopamine-degrading enzyme catechol-O-methyl-transferase [62, 63] may provide a unique tool to study imbalance in the functional regulation of neurotransmitters implicated in schizophrenia.

(iii) Finally, the development of schizophrenia-relevant animal models can also target the pharmacodynamics of common antipsychotic drugs, to identify their molecular substrates, and to optimise their pharmacokinetics, to develop new drugs, or to test potential antipsychotics.

4. Concept of Neurogenesis in Schizophrenia

Neuronal stem cells (NSCs) belong to the class of adult stem cells. They are multipotent and able to generate (only) the specific cell lineages of the nervous system: neurons, astrocytes, and oligodendrocytes [68]. NSCs will be generated throughout the whole life, but with declining intensity. NSCs are primarily located in the subgranular zone (SGZ) of the hippocampal dentate gyrus and in the subventricular zone (SVZ) of the lateral ventricles. But there is also evidence that NSCs are present in multiple areas of the adult brain [69]. Under the influence of their local microenvironment, that is, their niche, NSCs take different developmental pathways/roads of life. NSCs in the SVZ become neuroblasts, migrate towards the rostral migratory stream into the olfactory bulb, and develop into interneurons. NSCs in the SGZ, on the other hand, develop into local dentate granule cells [70].

Even if most evidences were acquired from different animal models, there is converging consensus that adult neurogenesis seems to be essential for different processes, such as learning and memory [71, 72], mood regulation [73, 74], physiological (maintenance) neuroregeneration, neurorestoration after mechanical brain injuries [75, 76], stroke [76, 77], multiple sclerosis [78, 79], and Parkinson's disease [80]. Thus it is not surprising that disturbances of adult neurogenesis are investigated in a wide range of pathological processes including neurodegenerative diseases, brain tumours, seizures, and mental illnesses such as schizophrenia, major depression, dementia, and alcoholism [70].

Because of these reasons, over the past years there has been a growing interest into neurogenesis-relevant research on postmortem human tissues of schizophrenic patients. Arnold and Watt [21] found abnormal neuronal densities in the olfactory epithelium of schizophrenics. Rioux and Arnold [81] demonstrated a deregulated expression of retinoid receptors in schizophrenia and that retinoid signalling plays a central role in neurogenesis. Reif and coworkers [22] were able to demonstrate that the first

Table 1: Critical selection of most relevant papers published in the last 13 years.

Author	Year	Finding	Reason for selection
Bayer et al. [1]	1999	Genetic or environmental first hit affects brain development, a second hit later in life initiated the outbreak of schizophrenia	Comprehensive hypothesis concerning schizophrenia etiology
Arnold and Watt [21]	2001	Number of immature cells is increased in the olfactory epithelium of schizophrenics	First report on altered cell density in schizophrenics
Reif et al. [22]	2006	Cell proliferation is diminished in the dentate gyrus of schizophrenics	First report on altered cell proliferation in the human brain
Kippin et al. [23] Wakade et al. [24] Wang et al. [25]	2005 2002 2004	Typical and atypical neuroleptics enhance neurogenesis in the subventricular zone	Effects of neuroleptics on neurogenesis in the subventricular zone
Kodama et al. [26] Wakade et al. [24] Halim et al. [27]	2004 2002 2004	Atypical but not typical neuroleptics interfere with hippocampal neurogenesis	Effects of neuroleptics on neurogenesis in the hippocampus
Piontkewitz et al. [28]	2012	Risperidone partially restored impaired neurogenesis in poly I:C offspring	Effect of an atypical neuroleptic on neurogenesis in an model of maternal infection
Keilhoff et al. [29]	2010	Subchronic treatment with haloperidol ameliorated decreased neurogenesis and normalised behaviour in vitamin D-deficient rats	Effect of neuroleptics on neurogenesis in the vitamin D model
Keilhoff et al. [30]	2010	Risperidone and haloperidol promoted survival in stem cells in the hippocampus of rats subchronically treated with ketamine	Effects of typical and atypical neuroleptics on neurogenesis in the ketamine model

Table 2: Survey of the effects of neuroleptics on hippocampal neurogenesis.

Model	Effect on neuroleptics on neurogenesis in the hippocampus	Literature
Vitamin D deficiency	Haloperidol ↑	[29]
Maternal infection	Risperidone ↑	[28]
	Clozapine Ø↗	[64]
Ketamine	Haloperidol ↑*	[30]
	Risperidone ↑*	[30]
Phencyclidine	Risperidone Ø	[65]
	Clozapine ↑	[66]
Postnatal lesion of ventral hippocampus	Haloperidol Ø↗	[67], indirect

Ø: no effect, ↑: increase, ↗: increase after application of low doses, *: cell survival.

step of adult neurogenesis, that is, cell proliferation, is diminished in the dentate gyrus of patients suffering from schizophrenia. Atz and coworkers [82] found a genotypic association of NCAM1 polymorphisms with schizophrenia. NCAM expression is a characteristic feature of the postnatal neurogenic niche [83].

Beside differentiation into neurons, adult neuronal stem cells may also undergo gliogenesis. There is an increasing body of evidence for glial pathology in schizophrenia [84]. Oligodendrocyte and myelin dysfunction are perturbed in schizophrenia since its earliest phases. Through changes in synaptic formation and/or function, they can induce cognitive dysfunction, one of the core symptoms of schizophrenia [85, 86]. The activation of astrocytes has been discussed as an important pathogenic factor for the development of schizophrenia [87], and also microglia has been shown to remodel the CNS during development as well as after injuries [88–92].

5. Neurogenic Potential of Antipsychotics

Basically, antipsychotic drugs are divided into typical (first-generation antipsychotics) and atypical (second-generation antipsychotics). Within the commonly used typical antipsychotics in early psychosis are butyrophenone (e.g., haloperidol) and phenothiazine (e.g., chlorpromazine) derivates and within the atypical are clozapine, olanzapine, quetiapine, risperidone (reviewed in [93]).

Antipsychotics interfere with neuronal remodelling. Thereby dopaminergic effects seem to be involved [23, 94]. It was suggested that stimulation of dopamine D2 receptors

inhibits the proliferation of neuronal stem cells and that tonic endogenous dopamine inhibits their proliferation [88]. Moreover, it was reported that blockade of D2 receptors activates transcription factors which regulate the expression of genes of neuronal growth factors [95].

Olanzapine, which has less affinity for the dopaminergic receptors [96], is also able to enhance SVZ [24, 25] and hippocampal neurogenesis [26]. The latter, on the other hand, is not influenced by haloperidol [24, 27] clearly indicating that typical and atypical antipsychotics differentially regulate neurogenesis. In support to this notion, it was also reported that typical and atypical antipsychotics differentially induce neuronal plasticity and synaptic remodelling. Atypical but not typical antipsychotics are effective not only in the striatum but also in the prefrontal cortex and hippocampus [94]. With respect to the schizophrenia-relevant morphometric changes and cellular abnormalities in the grey matter, the reported effects of antipsychotics in the prefrontal cortex are particularly interesting (reviewed in [97]). Interestingly, long-term antipsychotic treatment induced glial but not neuronal cell proliferation in monkeys and that with no difference between typical and atypical antipsychotics [98]. All these results, however, do not challenge the scientific consensus that the adult cortex under physiological conditions belongs to the so-called nonneurogenic tissue.

6. Mechanisms Underlying the Effects of Antipsychotics on Neurogenesis

There are different possible mechanisms by which antipsychotics realize their influence on cell proliferation/neurogenesis. Earlier studies indicate that a couple of factors, for example, trophic and transcription factors, can interact in a fine-tuned network. Clozapine, for example, selectively increased FGF-2 (fibroblast growth factor-2, belongs to trophic factors) in the striatum [99]. In the hippocampus, FGF-2 is induced by quetiapine, but only when the NMDA receptor system is downregulated [100]. Other studies addressed the schizophrenia-relevant role of BDNF (brain-derived neurotrophic factor). However, the findings on BDNF status in naïve patients as well as in patients treated with antipsychotics are highly discrepant (reviewed by [101–105]). In animals, BDNF in the hippocampus was decreased by haloperidol and high-dosed risperidone [106–108], while olanzapine therapy enhanced BDNF [109]. It was also shown that haloperidol reduced NGF (neuronal growth factor) while olanzapine raised NGF levels and risperidone was ineffective on NGF [110, 111]. VEGF (vascular endothelial growth factor, angiogenic neurotrophin) seemed also to be involved in the action of antipsychotics. Haloperidol and olanzapine increased its hippocampal levels [112].

Antipsychotics can additionally influence cell proliferation/neurogenesis via targeting transcription factors implicated in mitotic activity regulation. Thus, it was shown that haloperidol, risperidone, and clozapine affected phosphorylation of extracellular signal-regulated kinases (ERKs) and cyclic adenosine 3′,5′-monophosphate (cAMP) response element (CRE) binding protein (CREB), each with different profiles. In fact, haloperidol and risperidone promoted phosphorylation [113, 114], while clozapine reduced ERK1/2 and CREB phosphorylation [113]. Furthermore, haloperidol treatment of mice increased phosphorylation of Akt1. With respect to the Akt/GSK-3 system, clozapine had similar effects as haloperidol increasing the Akt1 phosphorylation [115].

It is important to note that a direct antipsychotic drug-gene interaction should be taken into close consideration, even if a direct intervention on genes (belonging to the glutamate/NMDA receptor family) has been shown only for haloperidol [116, 117]. Since modulation of progenitor cell proliferation as well as neurogenesis resulting in NMDA receptor modulation has been described [118], these findings set one possible agenda by which a direct antipsychotic drug-gene interaction can become neurogenic.

Assuming that the pathology of the schizophrenia [119, 120] and the cell proliferation/neurogenesis [70, 121] are subjected to epigenetic control mechanisms, future research is needed to address the exact neurogenic mechanisms of antipsychotics adjusting by epigenetic factors.

A comprehensive summary of questions concerning neurogenic actions of antipsychotic drugs is given by Newton and Duman [122].

7. From the Bench: Interplay of Schizophrenia, Neuroleptics, and Neurogenesis

Several pathophysiological models have been proposed to explain schizophrenia and may appear to reflect distinct aspects of this disease. None of the pharmacological, genetic, and neurodevelopmental models have been evaluated in detail for translational relevance or to satisfy requirements of the different levels of validity (face, construct, and predictive validity; for review see [123]). Pharmacological models focused on alterations in the dopaminergic, glutamatergic, serotonergic, and GABAergic neurotransmitter systems [54, 124, 125]. They are based on alterations in these neurotransmitter systems, mimicking the *in vivo* conditions which are clinically relevant for schizophrenia. These alterations may be manipulated by drug challenges.

8. First Lesson: The Aspect of Maternal Vitamin D Deficiency

Given the apparent polygenic nature of schizophrenia and the limited translational significance of the available pharmacological models, neurodevelopmental models may offer a better chance of success [126]. Different animal models in schizophrenia research were developed to shed light on the developmental aspects of the disease.

In the course of ontogenesis there are two critical phases when the organism is susceptible to disturbances which can contribute to schizophrenia, that is, the embryonic/postdelivery phase (first hit) and during puberty (second hit). The vitamin D deficiency model appears to be useful to study the impact of brain disturbances during embryonic/fetal development [127, 128]. Interestingly, some

of the vitamin D deficiency effects could be related to later gestational periods thus possibly expanding the hazardous time window for the neurobiological development of schizophrenia [129]. Moreover, it was shown that the neurosteroid can impact brain development by affecting migration and survival of developing neurones in the brain, by influencing brain levels of neurotrophins and their receptors [130], by altering brain apoptotic activity [131], and by exerting immunoregulatory and neuroprotective effects (for review see [127]).

The vitamin D deficiency model is one of the most commonly explored and used also in our lab [29, 132]. Generally, animals from a normal diet control group which were left untreated revealed a basal level of 5-bromo-2′-deoxyuridine (BrdU) immunolabelling in the hippocampal subgranular zone which was in line with previous reports [133]. Cell typing 5 days after BrdU application offered a large pool of BrdU-labelled cells costained with DCX, a marker for immature neurons (~55%). A colabelling with NeuN, a marker for mature neurons, was only rarely found. The second largest group of BrdU-positive cells (~10%) were round or oval, medium-sized, and immunopositive for nestin, an intermediate filament that is expressed in neuronal stem or progenitor cells, identifying these cells as granule cell precursors. The remaining BrdU-positive cells expressed the common astroglia marker GFAP (~4%), the NG2 proteoglycan, a marker for oligodendroglial precursor cells and/or synantocytes (~4%), or were free of any co-labelling (~7%). There were no obvious differences in the distribution of BrdU-immunoreactive cells at different longitudinal levels of the dentate gyrus.

When cell typing was done 3 weeks after the last BrdU application, about 75% of the BrdU-labelled cells could be identified as granule cells. They had a small round soma, were immunopositive for NeuN, and some of them were shifted from the subgranular cell layer towards the middle part of the granule cell layer. Now, DCX co-labelling was found to be poor. Saline treatment did not alter this BrdU-labelling/costaining pattern.

Prenatal vitamin D deficiency reduced cell proliferation in the subgranular zone. The loss was proportionally distributed between the different cell types. Due to the very little counts of BrdU-positive cells marked with all nonneuronal markers, only the loss of DCX-positive cells was numerically evident.

In control animals, the typical (first-generation) neuroleptic haloperidol significantly increased the total number of BrdU-labelled cells. In vitamin D-deficient mice, the deficiency-induced reduction of cell proliferation was completely normalized by haloperidol resulting in a mitotic activity adequate to the untreated control level. In both cases, haloperidol treatment revealed tendentially more DCX-expressing cells. Moreover, a cytoskeletal hypertrophy of radial glia-like GFAP-positive astrocytes, possibly serving as climbing frame for the migrating neuronal newcomers, was found.

There is general consensus that neuroleptic drugs improve the psychopathology of schizophrenia. Treatment with typical neuroleptics is considered to result in minimal improvement or in worsening of cognitive processes [134], but there are also reports showing that typical neuroleptics provide modest gains in multiple cognitive domains [135]. As hippocampal neurogenesis plays an important role in learning and memory processes [74], we speculate that the previously demonstrated normalization of a vitamin D deficiency-induced habituation deficit in the hole board by haloperidol [136] could result from an at least partially restored mitotic activity. This idea is supported by findings that vitamin D depletion depressed promitotic genes [131, 137]. It is also plausible that there is some kind of exhaustion of the mitotic cell potency due to the overshooting activity in young animals, as it was demonstrated for the vitamin D deficiency model [131] and the NOS knock-out model [138]. Using learning paradigms dependent on hippocampal integrity in subsequent experiments, effects of APDs on both learning behaviour and neurogenesis should be studied in detail.

9. Second Lesson: The Aspect of Maternal Infection

Epidemiological studies have shown that maternal infection and inflammation in definite periods of pregnancy are significantly associated with an increased risk of schizophrenia in the offsprings. Infection with influenza virus [139] or application of polyriboinosinic-polyribocytidylic acid (poly I:C), an inflammatory agent which mimics inflammation by stimulation cytokine release through Toll-like receptor TLR3 activation is accepted models in schizophrenia research (for review see [140]). Prenatal immune stimulation reduces hippocampal neurogenesis [141–144]. The beneficial effects of atypical neuroleptic drugs (APDs), on the other hand, have been attributed to their capacity to increase neurogenesis [22, 145–147]. Together with the group of Weiner and Piontkewitz [28], some of the present authors studied the effects of adolescent poly I:C and risperidone treatment by analyzing a battery of cellular markers referring to cell proliferation and differentiation of hippocampal cell populations. The offspring of poly I:C-treated dams were characterized by an impaired neurogenesis including a decrease of calretinin-positive neurons, disturbed microvascularization and granular cell density in the dentate area, and a reduction of parvalbumin-expressing interneurons, whose deficit is a well-replicated neuropathological finding in schizophrenia [148]. Risperidone normalized the disturbed cell proliferation and/or survival, the number of calretinin and parvalbumin-expressing cells, and counteracted the disturbance in angiogenesis.

Together with previous reports on deficient hippocampal neurogenesis in offspring of poly I:C-exposed mice [64, 144] and LPS-exposed rats [141], our findings confirm the hypothesis that impaired neurogenesis is an important aetiopathological factor for hippocampal abnormalities and related cognitive dysfunctions in animal models and in patients with schizophrenia [22, 30, 74, 123, 141, 145–147, 149, 150]. Studies concerning an influence of antipsychotics in the poly I:C model are rare. Thus, Meyer et al. [64]

demonstrated that chronic clozapine treatment had no effect on poly I:C-hampered neurogenesis.

With respect to neurogenesis, our findings about a risperidone-mediated normalization of the byprenatal poly I:C disturbed angiogenesis are of special interest. In general, angiogenesis and neurogenesis are closely linked with each other [151]. Thus, VEGF modulates neurogenesis directly and also subsequently releasing neurotrophic factors such as BDNF [145, 152, 153]. Moreover, in a previous study we showed that administration of risperidone was able to increase VEGF expression [30] and angiogenesis [28] in the hippocampus of rats. Given that alterations of brain capillaries have been observed in schizophrenia [154], the demonstrated angiogenic effect of risperidone might be a partial mechanism by which antipsychotics realized their action.

Deficit of parvalbumin-expressing interneurons is an accepted feature in schizophrenia [148, 155], also demonstrable in animal models [156–160]. Our findings that risperidone counteracts this induced by prenatal poly I:C deficiency may also have important implications for understanding its antipsychotic mechanism.

To fully complete the present section, it is important to note that, in line with previously reported findings [161, 162], we found no effects of prenatal poly I:C treatment and/or risperidone intervention on astrocytes, oligodendrocytes, and microglial cells.

10. Third Lesson: The Aspect of Imbalances in Central Glutamatergic Neurotransmission

Glutamatergic alterations have been consistently showed in psychosis, since its earliest stages [54]. Repeated administration of noncompetitive NMDA receptor antagonists like ketamine, dizocilpine, and phencyclidine (PCP) to neonatal and pubertal rats leads to a number of molecular, neurochemical, and behavioural alterations that resemble those observed in schizophrenia [156, 163, 164]. Administration of the NMDA receptor antagonist dizocilpine and PCP in late fetal and early postnatal period of life in the rat will increase neuronal death by apoptosis [165]. On the contrary, administration of these substances to rats at an adult age will increase neuronal damage by necrosis with subsequent gliosis [166] which results in enduring alteration in the neuronal circuitry. Maeda and coworkers [66] showed that PCP-induced decreased adult neurogenesis was counteracted by coadministered glycine and D-serine confirming the involvement of NMDA receptors in disruption of neurogenesis. Moreover, they were able to demonstrate a reconstruction of neurogenesis by clozapine, but not haloperidol.

However, we found that acute application of ketamine in sub-anaesthetic doses had no effect on cell proliferation. Animals, decapitated 3 weeks after ketamine application, however, showed a significant increased number of BrdU-labelled nuclei in the subgranular zone compared to saline-treated and untreated animals, whereby the cell-type assignment did not differ between the groups. There was no difference between the left and the right hippocampus, but significantly more BrdU-labelled cells were found in the lateral than in the medial blade of the dentate gyrus. In our first respective paper [167], this was interpreted as stimulating effect of ketamine on neurogenesis. Later on [30], however, we speculated that the withdrawal rather than the application of ketamine was essential and that beside an increase of cell proliferation there was a better survival of proliferated cells. These effects were accompanied by an enhanced mRNA level of BDNF.

Haloperidol and the atypical antipsychotic risperidone increased the total number of BrdU-labelled cells surviving for three weeks within the granule cell layer in untreated animals. Hereby, VEGF (vascular endothelial growth factor, signalling protein involved in angiogenesis and cell proliferation in general), MMP2 (matrix metallopeptidase 2 (Gelatinase A)), a proteolytic enzyme involved in cell proliferation, adhesion, and migration), CREB, and p38 MAP kinase seemed to be involved at mRNA as well as protein levels. The ketamine withdrawal-induced changes in proliferation/survival, however, were not additionally affected by the neuroleptics [30].

Malberg and Monteggia [168] showed that chronic administration of haloperidol increased the level of BDNF in the frontal cortex and amygdala, a possible mechanism for the neuroproliferative potency of haloperidol. Together with its direct effect on MMP2 and the subsequent effect on VEGF (possibly by processing the VEGF binding proteins HARP (heparin affine regulatory peptide)) and CTGF (connective tissue growth factor), the cell proliferative/protective potency of haloperidol is plausible.

Nevertheless, the demonstrated haloperidol effect on cell proliferation is in agreement with some, but not all, previous reports [24, 25, 27, 169]. The differences can reflect methodological heterogeneity across different experimental settings (dosage, application regime, and used rat strain).

Interestingly, in the PCP model risperidone was unable to reverse the PCP-induced decreases in parvalbumin expression in the prefrontal cortex [65]. This indicates that the antipsychotic effects of risperidone differ (prenatal poly I:C insult (see above) versus chronic administration of PCP to adult animals).

11. Fourth Lesson: The Aspect of Mechanical Lesions

Lesion models such as the neonatal ventral hippocampal lesion result in schizophrenia-related alterations in behaviour, neurochemistry, and neuropathology when performed on postnatal day (PD) 7, but not on PD 14 or PD 21 [32]. Interestingly, lesions performed in adolescent rats result in less pronounced and qualitatively different schizophrenia-related alterations [170–173]. However, only few data suggesting a link between brain lesion and neurogenesis are available. Lipska et al. [174] and Ashe et al. [175] studied the expression of BDNF mRNA in rats with neonatal lesions of the ventral hippocampus and found consistently a suppressed BDNF level in the dentate gyrus. From that and from the BrdU incorporation studies, they concluded

that "a transient disconnection in the CA1 and CA2 area of the hippocampus may have long-lasting consequences for neurogenesis in the dentate gyrus" [176]. Negrete-Díaz et al. [67] showed that nitric oxide (NO) levels in the prefrontal cortex, the occipital cortex, and the cerebellum are higher in the damaged animals and that haloperidol, in part, attenuates these altered NO levels. NO itself is known to be anti-proliferative and it should be allowed to suppose a connection between the enhanced NO level and the reduced BrdU incorporation in animals with ventral hippocampus lesions. It is not clear, however, how haloperidol-induced reduction in NO may lead to a restored cell proliferation. This might be, at least partially, a mechanism by which haloperidol decreased stereotypy in ventral hippocampus damaged rats [177].

12. Conclusions/Outlook

Clinical and experimental researches indicate that neurogenesis is disturbed in schizophrenia, since its earliest phases. Moreover, antipsychotics specifically interact with these alterations, affecting the neurogenesis. By increasing the neurogenesis it may be possible to provide beneficial gains for processes related with learning and memory formation. The regulation of neurogenesis may be a promising novel target for the treatment and the prevention of schizophrenia.

References

[1] T. A. Bayer, P. Falkai, and W. Maier, "Genetic and non-genetic vulnerability factors in schizophrenia: the basis of the "two hit hypothesis"," *Journal of Psychiatric Research*, vol. 33, no. 6, pp. 543–548, 1999.

[2] T. M. Maynard, L. Sikich, J. A. Lieberman, and A. S. LaMantia, "Neural development, cell-cell signaling, and the "two-hit" hypothesis of schizophrenia," *Schizophrenia Bulletin*, vol. 27, no. 3, pp. 457–476, 2001.

[3] C. Pantelis, M. Yücel, S. J. Wood, P. D. McGorry, and D. Velakoulis, "Early and late neurodevelopmental disturbances in schizophrenia and their functional consequences," *Australian and New Zealand Journal of Psychiatry*, vol. 37, no. 4, pp. 399–406, 2003.

[4] J. L. Rapoport, A. M. Addington, S. Frangou, and M. R. C. Psych, "The neurodevelopmental model of schizophrenia: update 2005," *Molecular Psychiatry*, vol. 10, no. 5, pp. 434–449, 2005.

[5] P. Fusar-Poli, S. Borgwardt, A. Bechdolf et al., "The psychosis high risk state: a comprehensive state of the art review," *Archives of General Psychiatry*. In press.

[6] P. Fusar-Poli, I. Bonoldi, and A. R. Yung, "Predicting psychosis: meta-analysis of transition outcomes in individuals at high clinical risk," *Archives of General Psychiatry*, vol. 69, no. 3, pp. 220–229, 2012.

[7] P. Fusar-Poli, A. Bechdolf, M. Taylor et al., "At risk for schizophrenic or affective psychosis? A meta-analysis of ICD/DSM diagnostic outcomes in individuals at high clinical risk," *Schizophrenia Bulletin*. In press.

[8] P. Fusar-Poli, G. Deste, R. Smieskova et al., "Cognitive functioning in prodromal psychosis: a meta-analysis," *Archives of General Psychiatry*, vol. 69, no. 6, pp. 562–571, 2012.

[9] P. Fusar-Poli, S. Borgwardt, A. Crescini et al., "Neuroanatomy of vulnerability to psychosis: a voxel-based meta-analysis," *Neuroscience and Biobehavioral Reviews*, vol. 35, no. 5, pp. 1175–1185, 2011.

[10] P. Fusar-Poli, J. Radua, P. McGuire, and S. Borgwardt, "Neuroanatomical maps of psychosis onset: voxel-wise meta-analysis of antipsychotic-naive VBM studies," *Schizophrenia Bulletin*. In press.

[11] R. Smieskova, P. Fusar-Poli, P. Allen et al., "Neuroimaging predictors of transition to psychosis—a systematic review and meta-analysis," *Neuroscience and Biobehavioral Reviews*, vol. 34, no. 8, pp. 1207–1222, 2010.

[12] P. Fusar-Poli, O. D. Howes, P. Allen et al., "Abnormal frontostriatal interactions in people with prodromal signs of psychosis: a multimodal imaging study," *Archives of General Psychiatry*, vol. 67, no. 7, pp. 683–691, 2010.

[13] P. Fusar-Poli, O. D. Howes, P. Allen et al., "Abnormal prefrontal activation directly related to pre-synaptic striatal dopamine dysfunction in people at clinical high risk for psychosis," *Molecular Psychiatry*, vol. 16, no. 1, pp. 67–75, 2011.

[14] P. Fusar-Poli, J. Perez, M. Broome et al., "Neurofunctional correlates of vulnerability to psychosis: a systematic review and meta-analysis," *Neuroscience and Biobehavioral Reviews*, vol. 31, no. 4, pp. 465–484, 2007.

[15] N. A. Crossley, A. Mechelli, P. Fusar-Poli et al., "Superior temporal lobe dysfunction and frontotemporal dysconnectivity in subjects at risk of psychosis and in first-episode psychosis," *Human Brain Mapping*, vol. 30, no. 12, pp. 4129–4137, 2009.

[16] P. Fusar-Poli and A. Meyer-Lindenberg, "Striatal presynaptic dopamine in schizophrenia—part II: meta-analysis of [18F]/[11C] DOPA PET studies," *Schizophrenia Bulletin*. In press.

[17] P. Fusar-Poli, J. Stone, M. Broome, I. Valli, A. Mechelli, and M. McLean, "Thalamic glutamate levels as a predictor of cortical response during executive functioning in subjects at high risk for psychosis," *Archives of General Psychiatry*, vol. 68, no. 9, pp. 881–890, 2011.

[18] O. D. Howes, A. J. Montgomery, M. C. Asselin et al., "Elevated striatal dopamine function linked to prodromal signs of schizophenia," *Archives of General Psychiatry*, vol. 66, no. 1, pp. 13–20, 2009.

[19] P. McGuire, O. D. Howes, J. Stone, and P. Fusar-Poli, "Functional neuroimaging in schizophrenia: diagnosis and drug discovery," *Trends in Pharmacological Sciences*, vol. 29, no. 2, pp. 91–98, 2008.

[20] P. Fusar-Poli and A. R. Yung, "Should attenuated psychosis syndrome be included in the DSM5? The debate," *The Lancet*, vol. 379, no. 9816, pp. 591–592, 2012.

[21] I. Arnold and F. M. Watt, "c-Myc activation in transgenic mouse epidermis results in mobilization of stem cells and differentiation of their progeny," *Current Biology*, vol. 11, no. 8, pp. 558–568, 2001.

[22] A. Reif, S. Fritzen, M. Finger et al., "Neural stem cell proliferation is decreased in schizophrenia, but not in depression," *Molecular Psychiatry*, vol. 11, no. 5, pp. 514–522, 2006.

[23] T. E. Kippin, S. Kapur, and D. Van Der Kooy, "Dopamine specifically inhibits forebrain neural stem cell proliferation, suggesting a novel effect of antipsychotic drugs," *Journal of Neuroscience*, vol. 25, no. 24, pp. 5815–5823, 2005.

[24] C. G. Wakade, S. P. Mahadik, J. L. Waller, and F. C. Chiu, "Atypical neuroleptics stimulate neurogenesis in adult rat

brain," *Journal of Neuroscience Research*, vol. 69, no. 1, pp. 72–79, 2002.

[25] H. D. Wang, F. D. Dunnavant, T. Jarman, and A. Y. Deutch, "Effects of antipsychotic drugs on neurogenesis in the forebrain of the adult rat," *Neuropsychopharmacology*, vol. 29, no. 7, pp. 1230–1238, 2004.

[26] M. Kodama, T. Fujioka, and R. S. Duman, "Chronic olanzapine or fluoxetine administration increases cell proliferation in hippocampus and prefrontal cortex of adult rat," *Biological Psychiatry*, vol. 56, no. 8, pp. 570–580, 2004.

[27] N. D. Halim, C. S. Weickert, B. W. McClintock, D. R. Weinberger, and B. K. Lipska, "Effects of chronic haloperidol and clozapine treatment on neurogenesis in the adult rat hippocampus," *Neuropsychopharmacology*, vol. 29, no. 6, pp. 1063–1069, 2004.

[28] Y. Piontkewitz, H. G. Bernstein, H. Dobrowolny, B. Bogerts, I. Weiner, and G. Keilhoff, "Effects of risperidone treatment in adolescence on hippocampal neurogenesis, parvalbumin expression, and vascularization following prenatal immune activation in rats," *Brain, Behavior, and Immunity*, vol. 26, no. 2, pp. 353–363, 2012.

[29] G. Keilhoff, G. Grecksch, and A. Becker, "Haloperidol normalized prenatal vitamin D depletion-induced reduction of hippocampal cell proliferation in adult rats," *Neuroscience Letters*, vol. 476, no. 2, pp. 94–98, 2010.

[30] G. Keilhoff, G. Grecksch, H. G. Bernstein, T. Roskoden, and A. Becker, "Risperidone and haloperidol promote survival of stem cells in the rat hippocampus," *European Archives of Psychiatry and Clinical Neuroscience*, vol. 260, no. 2, pp. 151–162, 2010.

[31] V. Castagné, P. C. Moser, and R. D. Porsolt, "Preclinical behavioral models for predicting antipsychotic activity," *Advances in Pharmacology*, vol. 57, pp. 381–418, 2009.

[32] C. A. Jones, D. J. Watson, and K. C. Fone, "Animal models of schizophrenia," *British Journal of Pharmacology*, vol. 164, no. 4, pp. 1162–1194, 2011.

[33] L. Lu, T. Mamiya, T. Koseki, A. Mouri, and T. Nabeshima, "Genetic animal models of schizophrenia related with the hypothesis of abnormal neurodevelopment," *Biological & Pharmaceutical Bulletin*, vol. 34, no. 9, pp. 1358–1363, 2011.

[34] H. Moore, "The role of rodent models in the discovery of new treatments for schizophrenia: updating our strategy," *Schizophrenia Bulletin*, vol. 36, no. 6, pp. 1066–1072, 2010.

[35] J. C. Neill, S. Barnes, S. Cook et al., "Animal models of cognitive dysfunction and negative symptoms of schizophrenia: focus on NMDA receptor antagonism," *Pharmacology and Therapeutics*, vol. 128, no. 3, pp. 419–432, 2010.

[36] S. B. Powell, "Models of neurodevelopmental abnormalities in schizophrenia," *Current Topics in Behavioral Neurosciences*, vol. 4, pp. 435–481, 2010.

[37] S. Tordjman, D. Drapier, O. Bonnot et al., "Animal models relevant to schizophrenia and autism: validity and limitations," *Behavior Genetics*, vol. 37, no. 1, pp. 61–78, 2007.

[38] J. W. Young, X. Zhou, and M. A. Geyer, "Animal models of schizophrenia," *Current Topics in Behavioral Neurosciences*, no. 4, pp. 391–433, 2010.

[39] P. C. Ashe, M. D. Berry, and A. A. Boulton, "Schizophrenia, a neurodegenerative disorder with neurodevelopmental antecedents," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 25, no. 4, pp. 691–707, 2001.

[40] M. Cannon and R. M. Murray, "Neonatal origins of schizophrenia," *Archives of Disease in Childhood*, vol. 78, no. 1, pp. 1–3, 1998.

[41] D. R. Weinberger, "From neuropathology to neurodevelopment," *Lancet*, vol. 346, no. 8974, pp. 552–557, 1995.

[42] P. Falkai and B. Bogerts, "Cell loss in the hippocampus of schizophrenics," *European Archives of Psychiatry and Neurological Sciences*, vol. 236, no. 3, pp. 154–161, 1986.

[43] S. Heckers and C. Konradi, "Hippocampal neurons in schizophrenia," *Journal of Neural Transmission*, vol. 109, no. 5-6, pp. 891–905, 2002.

[44] S. Heckers, "The hippocampus in schizophrenia," *American Journal of Psychiatry*, vol. 161, no. 11, pp. 2138–2139, 2004.

[45] N. C. Allen, S. Bagade, M. B. McQueen et al., "Systematic meta-analyses and field synopsis of genetic association studies in schizophrenia: the SzGene database," *Nature Genetics*, vol. 40, no. 7, pp. 827–834, 2008.

[46] L. Desbonnet, J. L. Waddington, and C. M. P. O'Tuathaigh, "Mutant models for genes associated with schizophrenia," *Biochemical Society Transactions*, vol. 37, no. 1, pp. 308–312, 2009.

[47] P. J. Harrison, D. Pritchett, K. Stumpenhorst, J. F. Betts, W. Nissen, and J. Schweimer, "Genetic mouse models relevant to schizophrenia: taking stock and looking forward," *Neuropharmacology*, vol. 62, no. 3, pp. 1164–1167, 2012.

[48] A. Guidotti, C. Pesold, and E. Costa, "New neurochemical markers for psychosis: a working hypothesis of their operation," *Neurochemical Research*, vol. 25, no. 9-10, pp. 1207–1218, 2000.

[49] H. Stefansson, E. Sigurdsson, V. Steinthorsdottir et al., "Neuregulin 1 and susceptibility to schizophrenia," *American Journal of Human Genetics*, vol. 71, no. 4, pp. 877–892, 2002.

[50] C. S. Barros, B. Calabrese, P. Chamero et al., "Impaired maturation of dendritic spines without disorganization of cortical cell layers in mice lacking NRG1/ErbB signaling in the central nervous system," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 106, no. 11, pp. 4507–4512, 2009.

[51] M. Niwa, A. Kamiya, R. Murai et al., "Knockdown of DISC1 by in utero gene transfer disturbs postnatal dopaminergic maturation in the frontal cortex and leads to adult behavioral deficits," *Neuron*, vol. 65, no. 4, pp. 480–489, 2010.

[52] C. Kellendonk, E. H. Simpson, H. J. Polan et al., "Transient and selective overexpression of dopamine D2 receptors in the striatum causes persistent abnormalities in prefrontal cortex functioning," *Neuron*, vol. 49, no. 4, pp. 603–615, 2006.

[53] I. H. Deakin, W. Nissen, A. J. Law, T. Lane, R. Kanso, and M. H. Schwab, "Transgenic overexpression of the type I isoform of neuregulin 1 affects working memory and hippocampal oscillations but not long-term potentiation," *Cerebral Cortex*, vol. 22, no. 7, pp. 1520–1529, 2012.

[54] A. Egerton and J. M. Stone, "The glutamate hypothesis of schizophrenia: neuroimaging and drug development," *Current Pharmaceutical Biotechnology*, vol. 13, no. 8, pp. 1500–1512, 2012.

[55] G. E. Duncan, S. S. Moy, A. Perez et al., "Deficits in sensorimotor gating and tests of social behavior in a genetic model of reduced NMDA receptor function," *Behavioural Brain Research*, vol. 153, no. 2, pp. 507–519, 2004.

[56] R. L. Fradley, G. F. O'Meara, R. J. Newman, A. Andrieux, D. Job, and D. S. Reynolds, "STOP knockout and NMDA NR1 hypomorphic mice exhibit deficits in sensorimotor gating," *Behavioural Brain Research*, vol. 163, no. 2, pp. 257–264, 2005.

[57] T. B. Halene, R. S. Ehrlichman, Y. Liang et al., "Assessment of NMDA receptor NR1 subunit hypofunction in mice as a

model for schizophrenia," *Genes, Brain and Behavior*, vol. 8, no. 7, pp. 661–675, 2009.

[58] A. J. Ramsey, "NR1 knockdown mice as a representative model of the glutamate hypothesis of schizophrenia," *Progress in Brain Research*, vol. 179, pp. 51–58, 2009.

[59] R. R. Gainetdinov, "Dopamine transporter mutant mice in experimental neuropharmacology," *Naunyn-Schmiedeberg's Archives of Pharmacology*, vol. 377, no. 4–6, pp. 301–313, 2008.

[60] C. M. O'Tuathaigh, L. Desbonnet, P. M. Moran, B. P. Kirby, and J. L. Waddington, "Molecular genetic models related to schizophrenia and psychotic illness: heuristics and challenges," *Current Topics in Behavioral Neurosciences*, vol. 7, pp. 87–119, 2011.

[61] C. M. P. O'Tuathaigh, B. P. Kirby, P. M. Moran, and J. L. Waddington, "Mutant mouse models: genotype-phenotype relationships to negative symptoms in schizophrenia," *Schizophrenia Bulletin*, vol. 36, no. 2, pp. 271–288, 2010.

[62] D. Babovic, C. M. O'Tuathaigh, A. M. O'Connor et al., "Phenotypic characterization of cognition and social behavior in mice with heterozygous versus homozygous deletion of catechol-O-methyltransferase," *Neuroscience*, vol. 155, no. 4, pp. 1021–1029, 2008.

[63] J. A. Gogos, M. Morgan, V. Luine et al., "Catechol-O-methyltransferase-deficient mice exhibit sexually dimorphic changes in catecholamine levels and behavior," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 95, no. 17, pp. 9991–9996, 1998.

[64] U. Meyer, I. Knuesel, M. Nyffeler, and J. Feldon, "Chronic clozapine treatment improves prenatal infection-induced working memory deficits without influencing adult hippocampal neurogenesis," *Psychopharmacology*, vol. 208, no. 4, pp. 531–543, 2010.

[65] C. E. McKibben, T. A. Jenkins, H. N. Adams, M. K. Harte, and G. P. Reynolds, "Effect of pretreatment with risperidone on phencyclidine-induced disruptions in object recognition memory and prefrontal cortex parvalbumin immunoreactivity in the rat," *Behavioural Brain Research*, vol. 208, no. 1, pp. 132–136, 2010.

[66] K. Maeda, H. Sugino, T. Hirose et al., "Clozapine prevents a decrease in neurogenesis in mice repeatedly treated with phencyclidine," *Journal of Pharmacological Sciences*, vol. 103, no. 3, pp. 299–308, 2007.

[67] J. Negrete-Díaz, E. Baltazar-Gaytán, M. Bringas et al., "Neonatal ventral hippocampus lesion induces increase in no levels which is attenuated by subchronic haloperidol treatment," *Synapse*, vol. 64, no. 12, pp. 941–947, 2010.

[68] F. H. Gage, "Mammalian neural stem cells," *Science*, vol. 287, no. 5457, pp. 1433–1438, 2000.

[69] A. Alvarez-Buylla and J. M. García-Verdugo, "Neurogenesis in adult subventricular zone," *Journal of Neuroscience*, vol. 22, no. 3, pp. 629–634, 2002.

[70] D. K. Ma, M. C. Marchetto, J. U. Guo, G. L. Ming, F. H. Gage, and H. Song, "Epigenetic choreographers of neurogenesis in the adult mammalian brain," *Nature Neuroscience*, vol. 13, no. 11, pp. 1338–1344, 2010.

[71] B. Artegiani and F. Calegari, "Age-related cognitive decline: can neural stem cells help us?" *Aging*, vol. 4, no. 3, pp. 176–186, 2012.

[72] A. Marin-Burgin and A. F. Schinder, "Requirement of adult-born neurons for hippocampus-dependent learning," *Behavioural Brain Research*, vol. 227, no. 2, pp. 391–399, 2012.

[73] J. S. Snyder, A. Soumier, M. Brewer, J. Pickel, and H. A. Cameron, "Adult hippocampal neurogenesis buffers stress responses and depressive behaviour," *Nature*, vol. 476, no. 7361, pp. 458–461, 2011.

[74] A. J. Eisch, H. A. Cameron, J. M. Encinas, L. A. Meltzer, G. L. Ming, and L. S. Overstreet-Wadiche, "Adult neurogenesis, mental health, and mental illness: hope or hype?" *Journal of Neuroscience*, vol. 28, no. 46, pp. 11785–11791, 2008.

[75] A. Foret, R. Quertainmont, O. Botman et al., "Stem cells in the adult rat spinal cord: plasticity after injury and treadmill training exercise," *Journal of Neurochemistry*, vol. 112, no. 3, pp. 762–772, 2010.

[76] Y. Xiong, A. Mahmood, and M. Chopp, "Angiogenesis, neurogenesis and brain recovery of function following injury," *Current Opinion in Investigational Drugs*, vol. 11, no. 3, pp. 298–308, 2010.

[77] A. Erlandsson, C. H. Lin, F. Yu, and C. M. Morshead, "Immunosuppression promotes endogenous neural stem and progenitor cell migration and tissue regeneration after ischemic injury," *Experimental Neurology*, vol. 230, no. 1, pp. 48–57, 2011.

[78] C. Zhao, M. Zawadzka, A. J. A. Roulois, C. C. Bruce, and R. J. M. Franklin, "Promoting remyelination in multiple sclerosis by endogenous adult neural stem/precursor cells: defining cellular targets," *Journal of the Neurological Sciences*, vol. 265, no. 1-2, pp. 12–16, 2008.

[79] J. P. Hugnot and R. Franzen, "The spinal cord ependymal region: a stem cell niche in the caudal central nervous system," *Frontiers in Bioscience*, vol. 16, no. 3, pp. 1044–1059, 2011.

[80] M. Geraerts, O. Krylyshkina, Z. Debyser, and V. Baekelandt, "Concise review: therapeutic strategies for Parkinson disease based on the modulation of adult neurogenesis," *Stem Cells*, vol. 25, no. 2, pp. 263–270, 2007.

[81] L. Rioux and S. E. Arnold, "The expression of retinoic acid receptor alpha is increased in the granule cells of the dentate gyrus in schizophrenia," *Psychiatry Research*, vol. 133, no. 1, pp. 13–21, 2005.

[82] M. E. Atz, B. Rollins, and M. P. Vawter, "NCAM1 association study of bipolar disorder and schizophrenia: polymorphisms and alternatively spliced isoforms lead to similarities and differences," *Psychiatric Genetics*, vol. 17, no. 2, pp. 55–67, 2007.

[83] E. Gascon, L. Vutskits, and J. Z. Kiss, "The role of PSA-NCAM in adult neurogenesis," *Advances in Experimental Medicine and Biology*, vol. 663, pp. 127–136, 2010.

[84] H. G. Bernstein, J. Steiner, and B. Bogerts, "Glial cells in schizophrenia: pathophysiological significance and possible consequences for therapy," *Expert Review of Neurotherapeutics*, vol. 9, no. 7, pp. 1059–1071, 2009.

[85] R. D. Fields, "White matter in learning, cognition and psychiatric disorders," *Trends in Neurosciences*, vol. 31, no. 7, pp. 361–370, 2008.

[86] N. Takahashi, T. Sakurai, K. L. Davis, and J. D. Buxbaum, "Linking oligodendrocyte and myelin dysfunction to neurocircuitry abnormalities in schizophrenia," *Progress in Neurobiology*, vol. 93, no. 1, pp. 13–22, 2010.

[87] M. Rothermundt, J. N. Ahn, and S. Jorgens, "S100B in schizophrenia: an update," *General Physiology and Biophysics*, vol. 28, pp. F76–F81, 2009.

[88] S. D. Bilbo and J. M. Schwarz, "Early-life programming of later-life brain and behavior: a critical role for the immune system," *Frontiers in Behavioral Neuroscience*, vol. 3, article 14, 2009.

[89] G. J. Harry and A. D. Kraft, "Microglia in the developing brain: a potential target with lifetime effects," *Neurotoxicology*, vol. 33, no. 2, pp. 191–206, 2012.

[90] H. Neumann, M. R. Kotter, and R. J. M. Franklin, "Debris clearance by microglia: an essential link between degeneration and regeneration," *Brain*, vol. 132, no. 2, pp. 288–295, 2009.

[91] M. Olah, S. Amor, N. Brouwer, J. Vinet, B. Eggen, and K. Biber, "Identification of a microglia phenotype supportive of remyelination," *Glia*, vol. 60, no. 2, pp. 306–321, 2012.

[92] M. E. Tremblay, B. Stevens, A. Sierra, H. Wake, and A. Bessis, "Nimmerjahn A. The role of microglia in the healthy brain," *Journal of Neuroscience*, vol. 31, no. 45, pp. 16064–16069, 2011.

[93] N. A. Crossley, M. Constante, P. McGuire, and P. Power, "Efficacy of atypical v. typical antipsychotics in the treatment of early psychosis: meta-analysis," *British Journal of Psychiatry*, vol. 196, no. 6, pp. 434–439, 2010.

[94] J. Horacek, V. Bubenikova-Valesova, M. Kopecek et al., "Mechanism of action of atypical antipsychotic drugs and the neurobiology of schizophrenia," *CNS Drugs*, vol. 20, no. 5, pp. 389–409, 2006.

[95] F. Angelucci, A. A. Mathé, and L. Aloe, "Neurotrophic factors and CNS disorders: findings in rodent models of depression and schizophrenia," *Progress in Brain Research*, vol. 146, pp. 151–165, 2004.

[96] E. Prommer, "Olanzapine: palliative medicine update," *American Journal of Hospice Palliative Care*. In press.

[97] R. G. Steen, C. Mull, R. McClure, R. M. Hamer, and J. A. Lieberman, "Brain volume in first-episode schizophrenia: systematic review and meta-analysis of magnetic resonance imaging studies," *British Journal of Psychiatry*, vol. 188, pp. 510–518, 2006.

[98] L. D. Selemon, M. S. Lidow, and P. S. Goldman-Rakic, "Increased volume and glial density in primate prefrontal cortex associated with chronic antipsychotic drug exposure," *Biological Psychiatry*, vol. 46, no. 2, pp. 161–172, 1999.

[99] M. A. Riva, R. Molteni, F. Tascedda, A. Massironi, and G. Racagni, "Selective modulation of fibroblast growth factor-2 expression in the rat brain by the atypical antipsychotic clozapine," *Neuropharmacology*, vol. 38, no. 7, pp. 1075–1082, 1999.

[100] F. Fumagalli, R. Molteni, F. Bedogni et al., "Quetiapine regulates FGF-2 and BDNF expression in the hippocampus of animals treated with MK-801," *NeuroReport*, vol. 15, no. 13, pp. 2109–2112, 2004.

[101] F. Angelucci, S. Brenè, and A. A. Mathé, "BDNF in schizophrenia, depression and corresponding animal models," *Molecular Psychiatry*, vol. 10, no. 4, pp. 345–352, 2005.

[102] P. F. Buckley, A. Pillai, D. Evans, E. Stirewalt, and S. Mahadik, "Brain derived neurotropic factor in first-episode psychosis," *Schizophrenia Research*, vol. 91, no. 1–3, pp. 1–5, 2007.

[103] P. F. Buckley, A. Pillai, and K. R. Howell, "Brain-derived neurotrophic factor: findings in schizophrenia," *Current Opinion in Psychiatry*, vol. 24, no. 2, pp. 122–127, 2011.

[104] M. J. Green, S. L. Matheson, A. Shepherd, C. S. Weickert, and V. J. Carr, "Brain-derived neurotrophic factor levels in schizophrenia: a systematic review with meta-analysis," *Molecular Psychiatry*, vol. 16, no. 9, pp. 960–972, 2011.

[105] O. Guillin, C. Demily, and F. Thibaut, "Brain-derived neurotrophic factor in schizophrenia and its relation with dopamine," *International Review of Neurobiology*, vol. 78, pp. 377–395, 2007.

[106] J. Chlan-Fourney, P. Ashe, K. Nylen, A. V. Juorio, and X. M. Li, "Differential regulation of hippocampal BDNF mRNA by typical and atypical antipsychotic administration," *Brain Research*, vol. 954, no. 1, pp. 11–20, 2002.

[107] N. M. Dawson, E. H. Hamid, M. F. Egan, and G. E. Meredith, "Changes in the pattern of brain-derived neurotrophic factor immunoreactivity in the rat brain after acute and subchronic haloperidol treatment," *Synapse*, vol. 39, no. 1, pp. 70–81, 2001.

[108] A. M. Lindén, J. Väisänen, M. Lakso, H. Nawa, G. Wong, and E. Castrén, "Expression of neurotrophins BDNF and NT-3, and their receptors in rat brain after administration of antipsychotic and psychotrophic agents," *Journal of Molecular Neuroscience*, vol. 14, no. 1-2, pp. 27–37, 2000.

[109] V. Parikh, M. M. Khan, and S. P. Mahadik, "Olanzapine counteracts reduction of brain-derived neurotrophic factor and TrkB receptors in rat hippocampus produced by haloperidol," *Neuroscience Letters*, vol. 356, no. 2, pp. 135–139, 2004.

[110] V. Parikh, A. V. Terry, M. M. Khan, and S. P. Mahadik, "Modulation of nerve growth factor and choline acetyltransferase expression in rat hippocampus after chronic exposure to haloperidol, risperidone, and olanzapine," *Psychopharmacology*, vol. 172, no. 4, pp. 365–374, 2004.

[111] A. V. Terry Jr., D. A. Gearhart, S. Warner et al., "Protracted effects of chronic oral haloperidol and risperidone on nerve growth factor, cholinergic neurons, and spatial reference learning in rats," *Neuroscience*, vol. 150, no. 2, pp. 413–424, 2007.

[112] A. Pillai and S. P. Mahadik, "Differential effects of haloperidol and olanzapine on levels of vascular endothelial growth factor and angiogenesis in rat hippocampus," *Schizophrenia Research*, vol. 87, no. 1–3, pp. 48–59, 2006.

[113] L. Pozzi, K. Håkansson, A. Usiello et al., "Opposite regulation by typical and atypical anti-psychotics of ERK1/2, CREB and Elk-1 phosphorylation in mouse dorsal striatum," *Journal of Neurochemistry*, vol. 86, no. 2, pp. 451–459, 2003.

[114] B. H. Yang, H. Son, S. Hyeon Kim, J. H. Nam, J. H. Choi, and J. S. Lee, "Phosphorylation of ERK and CREB in cultured hippocampal neurons after haloperidol and risperidone administration," *Psychiatry and Clinical Neurosciences*, vol. 58, no. 3, pp. 262–267, 2004.

[115] Z. Freyberg, S. J. Ferrando, and J. A. Javitch, "Roles of the Akt/GSK-3 and Wnt signaling pathways in schizophrenia and antipsychotic drug action," *American Journal of Psychiatry*, vol. 167, no. 4, pp. 388–396, 2010.

[116] J. J. Crowley, Y. Kim, J. P. Szatkiewicz, A. L. Pratt, C. R. Quackenbush, and D. E. Adkins, "Genome-wide association mapping of loci for antipsychotic-induced extrapyramidal symptoms in mice," *Mammalian Genome*. vol. 23, no. 5-6, pp. 322–335, 2012.

[117] D. K. Putnam, J. Sun, and Z. Zhao, "Exploring schizophrenia drug-gene interactions through molecular network and pathway modeling," in *Proceedings of the AMIA Annual Symposium*, pp. 1127–1133, 2011.

[118] J. Nacher and B. S. McEwen, "The role of N-methyl-D-asparate receptors in neurogenesis," *Hippocampus*, vol. 16, no. 3, pp. 267–270, 2006.

[119] T. Archer, R. J. Beninger, T. Palomo, and R. M. Kostrzewa, "Epigenetics and biomarkers in the staging of neuropsychiatric disorders," *Neurotoxicity Research*, vol. 18, no. 3-4, pp. 347–366, 2010.

[120] N. P. Maric and D. M. Svrakic, "Why schizophrenia genetics needs epigenetics: a review," *Psychiatria Danubina*, vol. 24, no. 1, pp. 2–18, 2012.

[121] J. Sun, J. Sun, G. L. Ming, and H. Song, "Epigenetic regulation of neurogenesis in the adult mammalian brain," *European Journal of Neuroscience*, vol. 33, no. 6, pp. 1087–1093, 2011.

[122] S. S. Newton and R. S. Duman, "Neurogenic actions of atypical antipsychotic drugs and therapeutic implications," *CNS Drugs*, vol. 21, no. 9, pp. 715–725, 2007.

[123] U. Meyer and J. Feldon, "Epidemiology-driven neurodevelopmental animal models of schizophrenia," *Progress in Neurobiology*, vol. 90, no. 3, pp. 285–326, 2010.

[124] M. S. Trivedi and T. Jarbe, "A brief review on recent developments in animal models of schizophrenia," *Indian Journal of Pharmacology*, vol. 43, no. 4, pp. 375–380, 2011.

[125] M. Yanagi, S. Southcott, J. Lister, and C. A. Tamminga, "Animal models of schizophrenia emphasizing construct validity," *Progress in Molecular Biology and Translational Science*, vol. 105, pp. 411–444, 2012.

[126] C. Wilson and A. V. Terry Jr., "Neurodevelopmental animal models of schizophrenia: role in novel drug discovery and development," *Clinical Schizophrenia and Related Psychoses*, vol. 4, no. 2, pp. 124–137, 2010.

[127] J. P. Kesby, D. W. Eyles, T. H. J. Burne, and J. J. McGrath, "The effects of vitamin D on brain development and adult brain function," *Molecular and Cellular Endocrinology*, vol. 347, no. 1-2, pp. 121–127, 2011.

[128] J. J. McGrath, T. H. Burne, F. Féron, A. MacKay-Sim, and D. W. Eyles, "Developmental vitamin D deficiency and risk of schizophrenia: a 10-year update," *Schizophrenia Bulletin*, vol. 36, no. 6, pp. 1073–1078, 2010.

[129] J. O'Loan, D. W. Eyles, J. Kesby, P. Ko, J. J. McGrath, and T. H. J. Burne, "Vitamin D deficiency during various stages of pregnancy in the rat; its impact on development and behaviour in adult offspring," *Psychoneuroendocrinology*, vol. 32, no. 3, pp. 227–234, 2007.

[130] D. Eyles, J. Brown, A. Mackay-Sim, J. McGrath, and F. Feron, "Vitamin D3 and brain development," *Neuroscience*, vol. 118, no. 3, pp. 641–653, 2003.

[131] P. Ko, R. Burkert, J. McGrath, and D. Eyles, "Maternal vitamin D3 deprivation and the regulation of apoptosis and cell cycle during rat brain development," *Developmental Brain Research*, vol. 153, no. 1, pp. 61–68, 2004.

[132] A. Becker, D. W. Eyles, J. J. McGrath, and G. Grecksch, "Transient prenatal vitamin D deficiency is associated with subtle alterations in learning and memory functions in adult rats," *Behavioural Brain Research*, vol. 161, no. 2, pp. 306–312, 2005.

[133] G. Kempermann and F. H. Gage, "Neurogenesis in the adult hippocampus," in *Proceedings of the Novartis Foundation Symposium*, vol. 231, pp. 220–235, 2000.

[134] H. Y. Meltzer, S. Park, and R. Kessler, "Cognition, schizophrenia, and the atypical antipsychotic drugs," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 96, no. 24, pp. 13591–13593, 1999.

[135] A. L. Mishara and T. E. Goldberg, "A meta-analysis and critical review of the effects of conventional neuroleptic treatment on cognition in schizophrenia: opening a closed book," *Biological Psychiatry*, vol. 55, no. 10, pp. 1013–1022, 2004.

[136] A. Becker and G. Grecksch, "Pharmacological treatment to augment hole board habituation in prenatal Vitamin D-deficient rats," *Behavioural Brain Research*, vol. 166, no. 1, pp. 177–183, 2006.

[137] F. Féron, T. H. J. Burne, J. Brown et al., "Developmental Vitamin D3 deficiency alters the adult rat brain," *Brain Research Bulletin*, vol. 65, no. 2, pp. 141–148, 2005.

[138] G. Keilhoff, "nNOS deficiency-induced cell proliferation depletes the neurogenic reserve," *Neuroscience Letters*, vol. 505, no. 3, pp. 248–253, 2011.

[139] S. H. Fatemi, E. S. Emamian, D. Kist et al., "Defective corticogenesis and reduction in Reelin immunoreactivity in cortex and hippocampus of prenatally infected neonatal mice," *Molecular Psychiatry*, vol. 4, no. 2, pp. 145–154, 1999.

[140] D. S. Macedo, D. P. Araujo, L. R. Sampaio, S. M. Vasconcelos, P. M. Sales, and F. C. Sousa, "Animal models of prenatal immune challenge and their contribution to the study of schizophrenia: a systematic review," *Brazilian Journal of Medical and Biological Research*, vol. 45, no. 3, pp. 179–186, 2012.

[141] K. Cui, H. Ashdown, G. N. Luheshi, and P. Boksa, "Effects of prenatal immune activation on hippocampal neurogenesis in the rat," *Schizophrenia Research*, vol. 113, no. 2-3, pp. 288–297, 2009.

[142] F. Lanté, J. Meunier, J. Guiramand et al., "Late N-acetylcysteine treatment prevents the deficits induced in the offspring of dams exposed to an immune stress during gestation," *Hippocampus*, vol. 18, no. 6, pp. 602–609, 2008.

[143] U. Meyer, M. Nyffeler, B. K. Yee, I. Knuesel, and J. Feldon, "Adult brain and behavioral pathological markers of prenatal immune challenge during early/middle and late fetal development in mice," *Brain, Behavior, and Immunity*, vol. 22, no. 4, pp. 469–486, 2008.

[144] S. A. Wolf, A. Melnik, and G. Kempermann, "Physical exercise increases adult neurogenesis and telomerase activity, and improves behavioral deficits in a mouse model of schizophrenia," *Brain, Behavior, and Immunity*, vol. 25, no. 5, pp. 971–980, 2011.

[145] G. Kempermann, J. Krebs, and K. Fabel, "The contribution of failing adult hippocampal neurogenesis to psychiatric disorders," *Current Opinion in Psychiatry*, vol. 21, no. 3, pp. 290–295, 2008.

[146] A. Reif, A. Schmitt, S. Fritzen, and K. P. Lesch, "Neurogenesis and schizophrenia: dividing neurons in a divided mind?" *European Archives of Psychiatry and Clinical Neuroscience*, vol. 257, no. 5, pp. 290–299, 2007.

[147] C. T. Toro and J. F. W. Deakin, "Adult neurogenesis and schizophrenia: a window on abnormal early brain development?" *Schizophrenia Research*, vol. 90, no. 1–3, pp. 1–14, 2007.

[148] Z. J. Zhang and G. P. Reynolds, "A selective decrease in the relative density of parvalbumin-immunoreactive neurons in the hippocampus in schizophrenia," *Schizophrenia Research*, vol. 55, no. 1-2, pp. 1–10, 2002.

[149] Y. Piontkewitz, M. Arad, and I. Weiner, "Risperidone administered during asymptomatic period of adolescence prevents the emergence of brain structural pathology and behavioral abnormalities in an animal model of schizophrenia," *Schizophrenia Bulletin*, vol. 37, no. 6, pp. 1257–1269, 2011.

[150] Y. Piontkewitz, Y. Assaf, and I. Weiner, "Clozapine administration in adolescence prevents postpubertal emergence of brain structural pathology in an animal model of schizophrenia," *Biological Psychiatry*, vol. 66, no. 11, pp. 1038–1046, 2009.

[151] N. M. Fournier and R. S. Duman, "Role of vascular endothelial growth factor in adult hippocampal neurogenesis: implications for the pathophysiology and treatment of depression," *Behavioural Brain Research*, vol. 227, no. 2, p. 440449, 2012.

[152] A. Louissaint Jr., S. Rao, C. Leventhal, and S. A. Goldman, "Coordinated interaction of neurogenesis and angiogenesis in the adult songbird brain," *Neuron*, vol. 34, no. 6, pp. 945–960, 2002.

[153] A. Schänzer, F. P. Wachs, D. Wilhelm et al., "Direct stimulation of adult neural stem cells in vitro and neurogenesis in vivo by vascular endothelial growth factor," *Brain Pathology*, vol. 14, no. 3, pp. 237–248, 2004.

[154] N. A. Uranova, I. S. Zimina, O. V. Vikhreva, N. O. Krukov, V. I. Rachmanova, and D. D. Orlovskaya, "Ultrastructural damage of capillaries in the neocortex in schizophrenia," *The World Journal of Biological Psychiatry*, vol. 11, no. 3, pp. 567–578, 2010.

[155] 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.

[156] Z. Abdul-Monim, J. C. Neill, and G. P. Reynolds, "Subchronic psychotomimetic phencyclidine induces deficits in reversal learning and alterations in parvalbumin-immunoreactive expression in the rat," *Journal of Psychopharmacology*, vol. 21, no. 2, pp. 198–205, 2007.

[157] S. Berretta, N. Lange, S. Bhattacharyya, R. Sebro, J. Garces, and F. M. Benes, "Long-term effects of amygdala GABA receptor blockade on specific subpopulations of hippocampal interneurons," *Hippocampus*, vol. 14, no. 7, pp. 876–894, 2004.

[158] M. K. Harte, S. B. Powell, N. R. Swerdlow, M. A. Geyer, and G. P. Reynolds, "Deficits in parvalbumin and calbindin immunoreactive cells in the hippocampus of isolation reared rats," *Journal of Neural Transmission*, vol. 114, no. 7, pp. 893–898, 2007.

[159] D. J. Lodge, M. M. Behrens, and A. A. Grace, "A loss of parvalbumin-containing interneurons is associated with diminished oscillatory activity in an animal model of schizophrenia," *Journal of Neuroscience*, vol. 29, no. 8, pp. 2344–2354, 2009.

[160] S. Penschuck, P. Flagstad, M. Didriksen, M. Leist, and A. T. Michael-Titus, "Decrease in parvalbumin-expressing neurons in the hippocampus and increased phencyclidine-induced locomotor activity in the rat methylazoxymethanol (MAM) model of schizophrenia," *European Journal of Neuroscience*, vol. 23, no. 1, pp. 279–284, 2006.

[161] M. Makinodan, K. Tatsumi, T. Manabe et al., "Maternal immune activation in mice delays myelination and axonal development in the hippocampus of the offspring," *Journal of Neuroscience Research*, vol. 86, no. 10, pp. 2190–2200, 2008.

[162] M. Nyffeler, U. Meyer, B. K. Yee, J. Feldon, and I. Knuesel, "Maternal immune activation during pregnancy increases limbic GABAA receptor immunoreactivity in the adult offspring: implications for schizophrenia," *Neuroscience*, vol. 143, no. 1, pp. 51–62, 2006.

[163] A. Becker, G. Grecksch, H. Schwegler, and T. Roskoden, "Expression of mRNA of neurotrophic factors and their receptors are significantly altered after subchronic ketamine treatment," *Medicinal Chemistry*, vol. 4, no. 3, pp. 256–263, 2008.

[164] A. Becker, B. Peters, H. Schroeder, T. Mann, G. Huether, and G. Grecksch, "Ketamine-induced changes in rat behaviour: a possible animal model of schizophrenia," *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, vol. 27, no. 4, pp. 687–700, 2003.

[165] C. Ikonomidou, F. Bosch, M. Miksa et al., "Blockade of NMDA receptors and apoptotic neurodegeneration in the developing brain," *Science*, vol. 283, no. 5398, pp. 70–74, 1999.

[166] J. W. Olney, J. Labruyere, G. Wang, D. F. Wozniak, M. T. Price, and M. A. Sesma, "NMDA antagonist neurotoxicity: mechanism and prevention," *Science*, vol. 254, no. 5037, pp. 1515–1518, 1991.

[167] G. Keilhoff, H. G. Bernstein, A. Becker, G. Grecksch, and G. Wolf, "Increased neurogenesis in a rat ketamine model of schizophrenia," *Biological Psychiatry*, vol. 56, no. 5, pp. 317–322, 2004.

[168] J. E. Malberg and L. M. Monteggia, "VGF, a new player in antidepressant action?" *Science Signaling*, vol. 1, no. 18, article pe19, 2008.

[169] J. E. Malberg, A. J. Eisch, E. J. Nestler, and R. S. Duman, "Chronic antidepressant treatment increases neurogenesis in adult rat hippocampus," *Journal of Neuroscience*, vol. 20, no. 24, pp. 9104–9110, 2000.

[170] A. Becker, G. Grecksch, H. G. Bernstein, V. Höllt, and B. Bogerts, "Social behaviour in rats lesioned with ibotenic acid in the hippocampus: quantitative and qualitative analysis," *Psychopharmacology*, vol. 144, no. 4, pp. 333–338, 1999.

[171] E. W. P. M. Daenen, J. A. Van der Heyden, C. G. Kruse, G. Wolterink, and J. M. Van Ree, "Adaptation and habituation to an open field and responses to various stressful events in animals with neonatal lesions in the amygdala or ventral hippocampus," *Brain Research*, vol. 918, no. 1-2, pp. 153–165, 2001.

[172] K. Y. Tseng, R. A. Chambers, and B. K. Lipska, "The neonatal ventral hippocampal lesion as a heuristic neurodevelopmental model of schizophrenia," *Behavioural Brain Research*, vol. 204, no. 2, pp. 295–305, 2009.

[173] G. K. Wood, B. K. Lipska, and D. R. Weinberger, "Behavioral changes in rats with early ventral hippocampal damage vary with age at damage," *Developmental Brain Research*, vol. 101, no. 1-2, pp. 17–25, 1997.

[174] B. K. Lipska, Z. Z. Khaing, C. S. Weickert, and D. R. Weinberger, "BDNF mRNA expression in rat hippocampus and prefrontal cortex: effects of neonatal ventral hippocampal damage and antipsychotic drugs," *European Journal of Neuroscience*, vol. 14, no. 1, pp. 135–144, 2001.

[175] P. C. Ashe, J. Chlan-Fourney, A. V. Juorio, and X. M. Li, "Brain-derived neurotrophic factor (BDNF) mRNA in rats with neonatal ibotenic acid lesions of the ventral hippocampus," *Brain Research*, vol. 956, no. 1, pp. 126–135, 2002.

[176] B. K. Lipska, "Using animal models to test a neurodevelopmental hypothesis of schizophrenia," *Journal of Psychiatry and Neuroscience*, vol. 29, no. 4, pp. 282–286, 2004.

[177] H. A. Al-Amin, D. R. Weinberger, and B. K. Lipska, "Exaggerated MK-801-induced motor hyperactivity in rats with the neonatal lesion of the ventral hippocampus," *Behavioural Pharmacology*, vol. 11, no. 3-4, pp. 269–278, 2000.