Spectroscopy 17 (2003) 231–239
IOS Press

Increased frontal lobe phosphocreatine levels observed in heavy cocaine users after treatment for cocaine dependence – An ^{1}H MRS T_2 relaxometry study

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Abstract. We have recently reported that relative concentrations of phosphocreatine (PCr) and creatine (Cr) may be estimated from brain 1H MR spectra based upon T_2 relaxation times. Emission tomography studies have consistently associated cocaine dependence and abstinence with decreased cerebral metabolism. We hypothesized that increased frontal lobe PCr levels would accompany treatment for cocaine dependence. Twenty-four cocaine dependent (CD) subjects were studied before and after 8 weeks of cocaine dependence treatment. Nine comparison subjects were studied at the same time points. At baseline, left frontal lobe ratios of PCr/tCr were 0.406 ± 0.081 in CD subjects and 0.411 ± 0.016 in comparison subjects. After treatment, these ratios increased 14.3% (0.464 vs. 0.406; $p = 0.006$) in CD subjects, remaining unchanged in comparison subjects (2.9%, 0.399 vs. 0.411; $p = 0.480$). At baseline, PCr levels of non-responders were 17.8% lower (0.375 vs. 0.442; $p = 0.042$) than those of responders, defined as 25% decrease in urine cocaine metabolites. After treatment, CD subjects had increased PCr levels: 18.4% (0.444 vs. 0.375; $p = 0.035$) for non-responders and 10.4% (0.488 vs. 0.442; $p = 0.092$) for responders. These results are consistent with decreased cerebral metabolism during treatment, measured as increased PCr. This is the first report using ^{1}H MRS T_2 relaxometry to measure a change in human brain energetics.

1. Introduction

Phosphocreatine (PCr) and creatine (Cr) are important molecules in cerebral bioenergetics. The proton (^{1}H) magnetic resonance spectroscopy (MRS) creatine resonance at 3.0 ppm arises from both PCr and Cr. The relative concentrations of PCr and Cr may vary depending on cerebral metabolic status [6,15–17, 19,24,27]. For example, during photic stimulation, phosphorus – (31P) MRS has been used to document decreases in occipital cortex PCr levels [12–14,23,26].

The effects of chronic cocaine abuse on human brain metabolites are not well understood. Volkow [30] and Wang [31] reported increases in cerebral metabolism during active cocaine use and craving for cocaine. Animal studies have also reported a decrease in PCr, consistent with increased metabolism, following injection of cocaine in rat brain [2]. No change in ^{1}H MRS Cr levels has been reported following

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acute cocaine administration in human subjects [5]. A number of investigators have reported increases in ^{1}H MRS brain creatine resonance intensity [4,22] in cocaine users, which could explain the reported lower ratio of N-acetyl aspartate (NAA) to creatine [3,18] in some cocaine users. These observations suggest that cocaine's effect on cerebral metabolism and on PCr level is time dependent [28] and additional studies are needed to further understanding of cocaine's effects on neurochemical markers.

Proton MRS is potentially a much more sensitive method for measuring changes in brain creatine stores *in vivo* than ^{31}P MRS. However, signals originating from PCr and Cr cannot be separated directly because they have almost identical resonance frequencies in ^{1}H MRS. We have recently demonstrated, however, that PCr and Cr have markedly different T_2 relaxation times *in vivo* and, on this basis, their relative concentrations may be estimated from ^{1}H MR spectra [14].

In this study, we collected 2D MRS data sets from cohorts of cocaine dependent ($N = 24$) and non-cocaine using subjects ($N = 9$). The cocaine dependent subjects were assessed immediately prior to and again following their participation in an eight-week, three arm, NIDA-sponsored treatment trial examining the efficacy of venlafaxine, pramipexole, or placebo in reducing cocaine use. The comparison subjects were assessed on two separate occasions separated by an eight-week interval. We hypothesized that the cocaine dependent subjects would have lower levels of PCr at study entry and that these levels would increase with treatment and reduction of cocaine use.

2. Methods

2.1. Clinical

All subjects provided written informed consent for a study that was approved by the Institutional Review Boards of Boston Medical Center, McLean Hospital, and the Boston VA Healthcare System. Eligible subjects were between 18 and 60 years of age. All cocaine dependent (CD) subjects were enrolled in a double blind, NIDA-sponsored, treatment study: three arm, placebo controlled assessment of venlafaxine, or pramipexole as treatment of cocaine dependence (CREST II). All CD subjects met the DSM-IV diagnosis of cocaine dependence and reported using cocaine on at least six occasions within the 28-day period prior to screening. Self-report of current cocaine use was substantiated with three urine specimens that were positive for the cocaine metabolite benzoylecsomine (BE) (ng/ml) over a two week period prior to study entry. Women with childbearing capacity were required to use an acceptable method of birth control. Potential subjects were excluded if they had a current dependence on any psychoactive drug other than cocaine, alcohol, or nicotine. In addition, those with neurological or psychiatric disorders requiring immediate treatment or that would make medication compliance difficult were excluded. Other reasons for exclusion included serious medical illness, current drug treatment, asthma, amenorrhea, due to pregnancy (by urine test), and abnormal lab results during screening. CD subjects with a current history of alcohol dependence did not require medical detoxification.

Medical history, routine blood work, and physical examination, including vital signs and weight, were obtained from all CD subjects as part of the CREST II trial. The following structured instruments were used to evaluate the CD subjects: (1) Structured Clinical Interview for DSM-IV Axis I Disorders, Research Version, Non-patient Edition (SCID-I/NP) obtained during the screening interview, (2) Addiction Severity Index (ASI) obtained prior to randomization and after eight weeks of treatment, (3) A quantitative urine toxicology screen for BE obtained three times a week during screening and throughout the study.

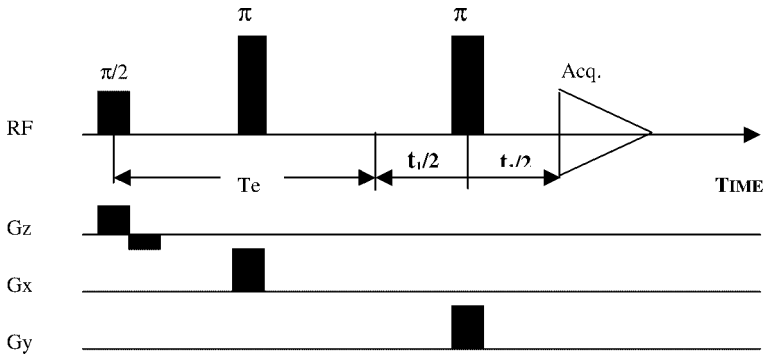

Fig. 1. Localized PRESS MRS pulse sequence with variable TE, the echo time, TE $=$ Te $+ t_1$, where $t_1 = t_{10} + n\Delta$te, $n = 0, 1, 2, \ldots, 63$; Δte $= 10$ msec and t_{10} is the minimum t_1.

MRI examinations were completed during active cocaine use prior to treatment and after 8-weeks of treatment. All CD subjects were randomized into one of three treatment cells: placebo, venlafaxine, or pramipexole. All treatment arms included manual-driven psychotherapy.

Comparison subjects were recruited by newspaper advertisement. A recruitment objective was to match the CD subjects on the basis of age, gender, and years of education. Comparison subjects had no Axis 1 diagnoses. Other exclusion criteria were similar to those employed for the CD subjects. The comparison group also completed two MRI examinations separated by the same time period as the CD subjects.

Response to treatment was assessed in two ways: by self-report (SR) and by urine toxicology report (UR). The response to treatment based on self-report of cocaine use was determined by the number of days cocaine was used in the last 30 days prior to treatment, as reported on the ASI and compared to the ASI Follow-Up Questionnaire after 8 weeks of treatment. A 25 percent drop in the number of days of reported cocaine use in a 30-day period was considered to be SR positive. All quantitative urine BE levels obtained the week prior to treatment were averaged, as were all quantitative urine BE levels after eight weeks of treatment. A twenty-five percent drop in mean urine BE levels from the first week to the last week was considered to be UR positive. These measures were chosen because they were temporally related to the scan acquisition.

2.2. Technical

A modified PRESS MRS sequence (Fig. 1) was implemented on a GE 1.5 Tesla SIGNA MR scanner at McLean Hospital to collect study data. Localized spectra were acquired from the left anterior frontal lobe with a voxel size of 18.75 cm^3 ($2.5 \times 2.5 \times 3$ cm^3; Fig. 2). Other scan parameters included: TR $= 2.32$ sec and 64 values of TE ranging from 48 msec to 678 msec. Each recorded FID had eight averages. Spectral bandwidth $= 2000$ Hz with 2048 data points.

After fully automated spectral processing, signal amplitudes for the 3.0 PPM creatine resonance were extracted from each spectrum at each of the 64 TE values (Fig. 3).

The Cre amplitude–TE curve obtained from each visit was fit to a mono-exponential model, as well as a bi-exponential model with PCr/tCr $= 0.40$ [7,9–11,25] initially for the T_2 values. Random permutations of the residues from the mono-exponential model were also fit to the bi-exponential model, in order to obtain non-parametric significance values for the goodness of fit [14]. Any results yielding significance

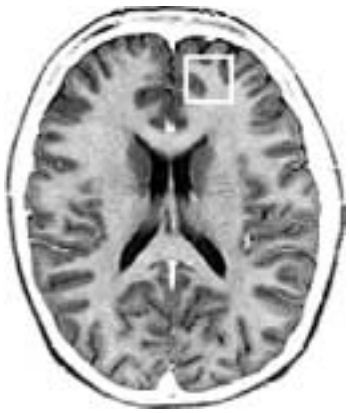

Fig. 2. Voxel location where MRS data were acquired in left anterior frontal lobe.

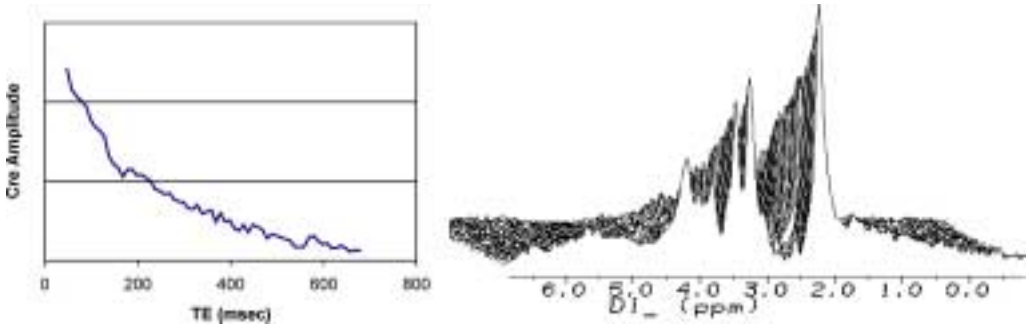

Fig. 3. Creatine amplitude–TE curve and its stack plot of ^{1}H MRS acquired from a cocaine dependent subject.

values greater than 0.02 were eliminated (Fig. 4). Only subjects with both visits meeting this significance criterion were included.

From the models, T_2 values were obtained for PCr from all the control subjects, and the median was calculated; the same procedure was performed for Cr. These values were in turn used to estimate the relative concentrations of PCr and Cr for both groups. Overall, the PCr/tCr ratios obtained were similar to the 0.40 value employed in the initial bi-exponential fits, lending confidence to this starting value. We employed this procedure because a full four-parameter fit, with the PCr/tCr ratio and both decay times variable, proved untenable given the available signal-to-noise ratio in the data.

3. Results

Comparison subjects and cocaine-dependent subjects were matched for gender, age and years of education. As expected, these two groups did not differ significantly for gender, age, or years of education (Table 1). In addition, cocaine-dependent subjects did not differ from comparison subjects in years of alcohol use (Table 1).

T_2 relaxation decay curves for the Cr resonance at 3.0 ppm both for cocaine-dependent and comparison subjects clearly demonstrated a bi-exponential decay characteristic for most subjects ($T_{2\mathrm{PCr}} = 116 \pm 17$ msec, $T_{2\mathrm{Cr}} = 338 \pm 19$ msec) in comparison with the results using a mono exponential fit.

Table 1
Baseline sample characteristics of comparison subjects and cocaine-dependent subjects

Characteristic	Comparison subjects $N = 9$		Cocaine dependent subjects				All cocaine subjects versus comparison subjects		Cocaine nonresponders versus responders	
			Nonresponders ($N = 13$)		Responders ($N = 11$)					
	N	%	N	%	N	%	χ^2 ($df = 2$)	p-value	χ^2 ($df = 2$)	p-value
Females	5	56	4	30.8	3	27.3	1.97	0.161	0.035	0.851
	Mean	SD	Mean	SD	Mean	SD	F-value	p-value	F-value	p-value
Age (years)	42.44	4.22	40.85	6.47	47.82	6.16	0.394	0.535	7.230	0.013*
Years education	13.17	0.83	12.48	2.26	12.67	1.30	0.868	0.359	0.064	0.803
Weight	160.22	26.00	158.98	33.09	182.82	22.33	0.702	0.409	4.130	0.054
Drug spending			488.46	336.75	590.91	417.96			0.443	0.513
30 day cocaine use			20.08	9.41	17.91	9.17			0.324	0.575
Urine cocaine			10868	11499	51315	62477			5.279	0.032*
Years cocaine use			16.77	8.90	15.36	8.29			0.158	0.695
30 day alcohol use			15.08	10.05	8.00	9.03			3.236	0.086
Years alcohol use	19.22	10.05	19.15	9.17	20.60	12.26	0.019	0.891	0.105	0.749
30 day marijuana use			6.92	8.60	5.36	6.44			0.245	0.626
Years marijuana use			18.54	11.36	10.09	10.31			3.581	0.072
30 day heroin use			0.62	2.22	0.91	3.02			0.075	0.786
Years heroin use			1.69	4.11	3.18	8.50			0.315	0.581
Cigarettes per day			8.89	7.93	8.84	5.62			0.000	0.988

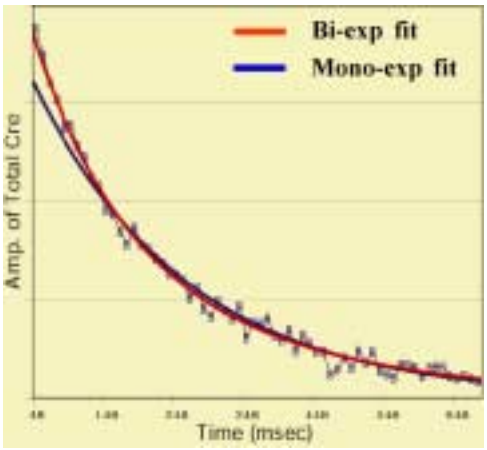

Fig. 4. Amplitude–TE curve of total creatine signal *in vivo* was fitted better with bi-exponential model than mono exponential model.

Table 2

Mean relative PCr/tCr concentrations of cocaine dependent and control subjects measured at their visit 1 and visit 2 and changes between two visits

Subjects (n)	Visit 1		Visit 2		% Change	t-value	p-value
	Mean	SD	Mean	SD			
Controls (9)	0.411	0.016	0.399	0.036	−2.9	−0.74	0.480
Cocaine dependent (24)	0.406	0.081	0.464	0.093	14.3	3.06	0.006
Responders (11)	0.442	0.098	0.488	0.103	10.4	1.864	0.092
Non-responders (13)	0.375[†]	0.048	0.444	0.081	18.4	2.374	0.035

[†] $p = 0.042$ for the comparison between Responders and Non-responders at visit 1.

Estimated values for T_{2PCr} and T_{2Cr} were not significantly different between cocaine-dependent subjects and comparison subjects nor between their two visits.

At baseline, mean (SD) ratios of phosphocreatine to total creatine (PCr/tCr) were 0.406 ($\pm$0.081) and 0.411 ($\pm$0.016) in the cocaine-dependent and comparison subjects, respectively ($F = 0.037$, $df = 31$, $p = 0.849$). After treatment, PCr/tCr ratios measured at visit 2 were 0.464 ($\pm$0.093) for cocaine-dependent subjects while this ratio for comparison subjects remained approximately the same, 0.399 ($\pm$0.036) ($F = 4.155$, df $= 31$, $p = 0.050$) (Table 2). The mean changes in PCr/tCr after treatment were +14.3% ($p = 0.006$) in cocaine-dependent subjects and −2.9% ($p = 0.480$) for the comparison subjects.

Furthermore, when cocaine-dependent subjects were divided into responder/non-responder groups based on urine drug measures, the PCr levels of non-responders at both baseline and after treatment were lower than corresponding values of responders. At baseline responders and non-responders differed significantly in age (47.82 and 40.85 years, respectively; $p = 0.013$) and urine cocaine measures (51315 and 10868 urine benzoylecsomine, respectively; $p = 0.032$). A trend also existed for responders to have reported fewer days of alcohol use in the previous 30 days than non-responders (8.00 and 15.08 days, respectively; $p = 0.086$). Other baseline characteristics of comparison subjects and cocaine dependant responders and non-responders are summarized in Table 1.

Table 3
Increases in brain PCr/tCr values after treatment in cocaine dependent patients treated with pramipexole, venlafaxine or placebo compared with their baseline PCr/tCr levels

Treatment medication (N)	Mean-increase	t-value	p-value
Pramipexole (8)	+0.076	1.692	0.135
Venlafaxine (9)	+0.062	2.411	0.042*
Placebo (7)	+0.033	1.195	0.277

PCr levels of both non-responders and responders increased after treatment (Table 2). Increased levels of PCr were accompanied by significant decreases in self-reported cocaine use for both non-responders and responders. At both timepoints, non-responders reported greater use of alcohol than responders, reaching significance after treatment ($F = 8.634$, $p = 0.008$). Non-responders were also significantly more likely to have current or past alcohol dependence or abuse than responders ($\chi^2 = 4.033$, $p = 0.045$). In addition, cocaine dependent subjects reported decreased alcohol consumption following treatment ($t = 2.187$, $p = 0.039$).

By dividing the patients into three treatment groups, there was a clear trend for increased PCr levels, consistent with a decrease in cerebral metabolism, associated with active medications used in treatment (Table 3). This increase in PCr levels for cocaine dependent subjects after pharmacotherapy suggests that treatment for cocaine dependence is associated with a decrease in cerebral metabolic rate, which may partly be due to a reduction in cocaine use.

4. Conclusion

Across our study sample, treatment for cocaine dependence appears to increase frontal lobe phosphocreatine levels by approximately 14% and non-responding cocaine users had lower baseline PCr levels than those of responding cocaine users. These findings are consistent with reports that cerebral metabolism decreases over time during abstinence [22,27,28]. In addition, this finding of increased PCr in left anterior frontal lobe is also consistent with decreased glucose metabolism in various frontal regions of past cocaine abusers [29]. As responders and non-responders reported decreased cocaine use after treatment, the increase in PCr seen in both groups could be partially attributed to decreased cocaine use.

The lower baseline levels of PCr in non-responders may partially be explained by higher alcohol consumption and a greater likelihood of suffering from past or current alcohol abuse or dependence. This is consistent with the report that chronic alcohol abuse in humans has been shown to decrease PCr as measured by ^{31}P MRS [21]. Alcohol consumption has also been shown to decrease PCr levels in rat brain, *in vivo* and *in vitro* [1,8]. As alcohol use is associated with decreased PCr, a decrease in alcohol consumption would be expected to result in increased PCr. Therefore, changes in self-reported alcohol use may have contributed to increased PCr in cocaine dependant subjects after treatment.

A preliminary assessment indicates that this increase was somewhat greater in subjects who were treated with either venlafaxine or pramipexole (Table 3). As responders and non-responders had similar percentage increases in PCr levels after treatment, and both groups observed decreased use of both alcohol and cocaine, differences in PCr may be attributable to these changes in substance abuse patterns. However, relative lower PCr concentrations of non-responders in comparison with responders were observed both at baseline and after treatment (Table 2).

As cocaine-dependent responders were significantly older than non-responders, it is possible that this difference in age may have contributed to the difference in PCr between the two groups. However, Longo et al. have shown that the intensity of the PCr resonance, as assessed by ^{31}P MRS, changed about 20% of the course of 60 years of normal aging [20]. Therefore, it is unlikely that this age difference of 6.98 years would have contributed significantly to the differences in PCr between responders and non-responders.

To the best of our knowledge, this is the first report involving the use of ^{1}H MRS T_2 relaxometry to measure a change in human brain energetics. The increased magnetic resonance sensitivity of the ^{1}H nucleus, relative to the ^{31}P nucleus, allows the assessment of relatively small brain regions. However, the temporal resolution of the method is limited by the need to fit a sufficient number of TE values for reliable bi-exponential fitting.

Acknowledgements

This study was supported, in part, by grants from the National Institute of Drug Abuse (DA09448 and DA14178 to PFR, DA50038 to DAC); Career Development Award CCS.

References

[1] B.M. Altura, B.T. Altura and R.K. Gupta, Alcohol intoxication results in rapid loss in free magnesium in brain and disturbances in brain bioenergetics: relation to cerebrovasospasm, alcohol-induced strokes, and barbiturate anesthesia-induced deaths, *Magnes. Trace Elem.* **10** (1991), 122–135.
[2] B.M. Altura and R.K. Gupta, Cocaine induces intracellular free Mg deficits, ischemia and stroke as observed by in-vivo 31P-NMR of the brain, *Biochim. Biophys. Acta* **1111** (1992), 271–274.
[3] L. Chang, T. Ernst, T. Strickland and C.M. Mehringer, Gender effects on persistent cerebral metabolite changes in the frontal lobes of abstinent cocaine users, *Am. J. Psychiatry* **156** (1999), 716–722.
[4] L. Chang, C.M. Mehringer, T. Ernst, R. Melchor, H. Myers, D. Horney and P. Satz, Neurochemical alterations in asymptomatic abstinent cocaine users: a proton magnetic resonance spectroscopy study, *Biol. Psychiatry* **42** (1997), 1105–1114.
[5] J.D. Christensen, M.J. Kaufman, B. Frederick, S.L. Rose, C.M. Moore, S.E. Lukas, J.H. Mendelson, B.M. Cohen and P.F. Renshaw, Proton magnetic resonance spectroscopy of human basal ganglia: response to cocaine administration, *Biol. Psychiatry* **48** (2000), 685–692.
[6] J.D. Christensen, M.J. Kaufman, J.M. Levin, J.H. Mendelson, B.L. Holman, B.M. Cohen and P.F. Renshaw, Abnormal cerebral metabolism in polydrug abusers during early withdrawal: a 31P MR spectroscopy study, *Magn. Reson. Med.* **35** (1996), 658–663.
[7] P. Christiansen, P. Toft, H.B. Larsson, M. Stubgaard and O. Henriksen, The concentration of N-acetyl aspartate, creatine + phosphocreatine, and choline in different parts of the brain in adulthood and senium, *Magn. Reson. Imaging* **11** (1993), 799–806.
[8] L.L. Fonseca, P.M. Alves, M.J. Carrondo and H. Santos, Effect of ethanol on the metabolism of primary astrocytes studied by (13)C- and (31)P-NMR spectroscopy, *J. Neurosci. Res.* **66** (2001), 803–811.
[9] V. Govindaraju, K. Young and A.A. Maudsley, Proton NMR chemical shifts and coupling constants for brain metabolites, *NMR Biomed.* **13** (2000), 129–153.
[10] O. Henriksen, In vivo quantitation of metabolite concentrations in the brain by means of proton MRS, *NMR Biomed.* **8** (1995), 139–148.
[11] T.Q. Hoang, S. Bluml, D.J. Dubowitz, R. Moats, O. Kopyov, D. Jacques and B.D. Ross, Quantitative proton-decoupled 31P MRS and 1H MRS in the evaluation of Huntington's and Parkinson's diseases, *Neurology* **50** (1998), 1033–1040.
[12] T. Kato, J. Murashita, T. Shioiri, H. Hamakawa and T. Inubushi, Effect of photic stimulation on energy metabolism in the human brain measured by 31P-MR spectroscopy, *J. Neuropsychiatry Clin. Neurosci.* **8** (1996), 417–422.
[13] T. Kato, J. Murashita, T. Shioiri, M. Terada, T. Inubushi and N. Kato, hotic stimulation-induced alteration of brain energy metabolism measured by 31P-MR spectroscopy in patients with MELAS, *J. Neurol. Sci.* **155** (1998), 182–185.
[14] Y. Ke, B.M. Cohen, S. Lowen, F. Hirashima, L. Nassar and P.F. Renshaw, Biexponential transverse relaxation (T(2)) of the proton MRS creatine resonance in human brain, *Magn. Reson. Med.* **47** (2002), 232–238.
[15] T. Kekelidze, I. Khait, A. Togliatti, J.M. Benzecry, B. Wieringa and D. Holtzman, Altered brain phosphocreatine and ATP regulation when mitochondrial creatine kinase is absent, *J. Neurosci. Res.* **66** (2001), 866–872.

[16] S.J. Kish, K.S. Kalasinsky, Y. Furukawa, M. Guttman, L. Ang, L. Li, V. Adams, G. Reiber, R.A. Anthony, W. Anderson, J. Smialek and L. DiStefano, Brain choline acetyltransferase activity in chronic, human users of cocaine, methamphetamine, and heroin, *Mol. Psychiatry* **4** (1999), 26–32.
[17] S. Klemm, R. Rzanny, S. Riehemann, H.P. Volz, B. Schmidt, U.J. Gerhard, C. Filz, A. Schonberg, H.J. Mentzel, W.A. Kaiser and B. Blanz, Cerebral phosphate metabolism in first-degree relatives of patients with schizophrenia, *Am. J. Psychiatry* **158** (2001), 958–960.
[18] S.J. Li, Y. Wang, J. Pankiewicz and E.A. Stein, Neurochemical adaptation to cocaine abuse: reduction of N-acetyl aspartate in thalamus of human cocaine abusers, *Biol. Psychiatry* **45** (1999), 1481–1487.
[19] X. Liu, D.B. Vaupel, S. Grant and E.D. London, Effect of cocaine-related environmental stimuli on the spontaneous electroencephalogram in polydrug abusers, *Neuropsychopharmacology* **19** (1998), 10–17.
[20] R. Longo, C. Ricci, L. Dalla Palma, R. Vidimari, A. Giorgini, J.A. den Hollander and C.M. Segebarth, Quantitative 31P MRS of the normal adult human brain. Assessment of interindividual differences and ageing effects, *NMR Biomed.* **6** (1993), 53–57.
[21] D.J. Meyerhoff, S. MacKay, D. Sappey-Marinier, R. Deicken, G. Calabrese, W.P. Dillon, M.W. Weiner and G. Fein, Effects of chronic alcohol abuse and HIV infection on brain phosphorus metabolites, *Alcohol Clin. Exp. Res.* **19** (1995), 685–692.
[22] J. O'Neill, V.A. Cardenas and D.J. Meyerhoff, Separate and interactive effects of cocaine and alcohol dependence on brain structures and metabolites: quantitative MRI and proton MR spectroscopic imaging, *Addict. Biol.* **6** (2001), 347–361.
[23] M. Rango, A. Castelli and G. Scarlato, Energetics of 3.5 s neural activation in humans: a 31P MR spectroscopy study, *Magn. Reson. Med.* **38** (1997), 878–883.
[24] S. Riehemann, H.P. Volz, S. Smesny, G. Hubner, B. Wenda, G. Rossger and H. Sauer, Phosphorus 31 magnetic resonance spectroscopy in schizophrenia research. Pathophysiology of cerebral metabolism of high-energy phosphate and membrane phospholipids, *Nervenarzt* **71** (2000), 354–363.
[25] D.L. Rothman, O.A. Petroff, K.L. Behar and R.H. Mattson, Localized 1H NMR measurements of gamma-aminobutyric acid in human brain in vivo, *Proc. Natl. Acad. Sci. USA* **90** (1993), 5662–5666.
[26] D. Sappey-Marinier, G. Calabrese, G. Fein, J.W. Hugg, C. Biggins and M.W. Weiner, Effect of photic stimulation on human visual cortex lactate and phosphates using 1H and 31P magnetic resonance spectroscopy, *J. Cereb. Blood Flow Metab.* **12** (1992), 584–592.
[27] L.M. Smith, L. Chang, M.L. Yonekura, K. Gilbride, J. Kuo, R.E. Poland, I. Walot and T. Ernst, Brain proton magnetic resonance spectroscopy and imaging in children exposed to cocaine in utero, *Pediatrics* **107** (2001), 227–231.
[28] N.D. Volkow, J.S. Fowler, A.P. Wolf, R. Hitzemann, S. Dewey, B. Bendriem, R. Alpert and A. Hoff, Changes in brain glucose metabolism in cocaine dependence and withdrawal, *Am. J. Psychiatry* **148** (1991), 621–626.
[29] N.D. Volkow, R. Hitzemann, G.J. Wang, J.S. Fowler, A.P. Wolf, S.L. Dewey and L. Handlesman, Long-term frontal brain metabolic changes in cocaine abusers, *Synapse* **11** (1992), 184–190.
[30] N.D. Volkow, G.J. Wang, J.S. Fowler, R. Hitzemann, B. Angrist, S.J. Gatley, J. Logan, Y.S. Ding and N. Pappas, Association of methylphenidate-induced craving with changes in right striato-orbitofrontal metabolism in cocaine abusers: implications in addiction, *Am. J. Psychiatry* **156** (1999), 19–26.
[31] G.J. Wang, N.D. Volkow, J.S. Fowler, P. Cervany, R.J. Hitzemann, N.R. Pappas, C.T. Wong and C. Felder, Regional brain metabolic activation during craving elicited by recall of previous drug experiences, *Life Sci.* **64** (1999), 775–784.