



ORIGINAL ARTICLE

Medicine Science 2020;9(2):450-3

Brain metabolite rates in amygdala and hippocampus in vaginismus patients

Mehmet Fatih Erbay¹, Esra Porgali Zayman²

¹Inonu University, Faculty of Medicine, Department of Radiology, Malatya, Turkey

²Inonu University, Faculty of Medicine, Department of Psychiatry, Malatya, Turkey

Received 16 January 2020; Accepted 16 March 2020

Available online 18.05.2020 with doi: [10.5455/medscience.2020.09.9222](https://doi.org/10.5455/medscience.2020.09.9222)

Abstract

Vaginismus has complex etiology. Regarding its clinical manifestation, it is a condition associated with anxiety and fear. The fact that hippocampus and amygdala are critical regions for the modulation of anxiety suggests that they might be playing a role in the neurobiology of vaginismus. In the present study, therefore, we aimed to investigate the concentrations of cerebral metabolites in both the amygdala and hippocampus in vaginismus patients and healthy controls via magnetic resonance spectroscopy method. A total of 39 vaginismus patients and 26 sexually active healthy controls were enrolled into the study. All the patients and healthy controls underwent magnetic resonance spectroscopy procedure. Comparison of the groups revealed no difference in terms of concentrations of cerebral metabolites in the amygdala, whereas median Cholin (Cho) concentration in the hippocampus was significantly higher in the patient vs. the control group. High concentration of Cho in the hippocampus of vaginismus patients might be indicating the changes in myelination or signal transduction in the relevant region. Considering the role of the hippocampus in fear conditioning and fear memory, such changes in the patients are considered as the neurobiological reflections of fear conditioning. Repeated and further studies will further contribute to the clarification of this subject.

Keywords: Vaginismus, hippocampus, amygdala, magnetic resonance spectroscopy

Introduction

Vaginismus is a condition that makes sexual intercourse and vaginal examination almost impossible due to the spasm of the muscles in the outer third of the vagina [1-3]. It is usually classified under the name of sexual pain disorder. In the Diagnostic and Statistical Manual of Mental Disorders, fifth edition (DSM 5), vaginismus is defined as failed vaginal penetration and substantial fear and anxiety while waiting for the action or during vaginal penetration [4].

Vaginismus has complex etiology. Multiple biopsychosocial processes including bidirectional connections between pelvic-genital regions and higher mental functions are thought to play a role in vaginismus [3,5].

Regarding its clinic, it is seen that vaginismus is an anxiety-related condition. The anxiety is like that observed during phobic

situations, and somatic components of anxiety accompany the other symptoms of vaginismus.

Phobic situations, anxiety and related clinical conditions are associated with hypothalamic-pituitary-adrenal axis (HPA). Hippocampus and amygdala are the critical limbic and paralimbic cerebral regions in emotional regulation [6]. On the other hand, there is a significant relationship between hippocampal region and anxiety [7,8]. Hippocampus and amygdala's being critical regions in the modulation of anxiety suggests that they might be playing a role in the neurobiology of vaginismus as well.

H-magnetic resonance spectroscopy (HMRS) is a noninvasive method for in-vivo detection of endogenous tissue metabolites. When applied at 3 Tesla to the human brain, it allows estimation of neurobiological structure of the brain at that moment based on the concentrations of 17 different neurochemical substances [9].

Accordingly, we aimed to investigate the concentrations of N-acetylaspartate (NAA), creatinine (Cr), Glutamine (Gln), Glutathione (GSH), Cholin (Cho) Myo-inositol (mIns), Glutamate (Glu) and Lactate (Lac) in the amygdala and hippocampus of vaginismus patients and healthy controls.

*Corresponding Author: Mehmet Fatih Erbay, Inonu University, Department of Radiology, Malatya, Turkey, E-mail: drfatiherbay@hotmail.com

Materials and Methods

Participants

Based on the power analysis with $\alpha=0.005$ and $1-\beta$ (power):0.80, it was calculated that at least 23 subjects are required in each group for the mean change in cerebral metabolites on MR spectroscopy to be 6.5 Units. The patients are selected among subjects that were diagnosed with vaginismus based on DSM 5 diagnostic criteria after being examined by a psychiatrist via SCID and that applied for sexual therapy program performed by an experienced psychiatrist. A total of 43 patients and 26 sexually active healthy controls, who had no history of medication in the last 3 months at the least, who were nonsmokers, who using the right hand and who gave written informed consent to participate in the study, were enrolled. MRS imaging of patients was done before treatment. Four patients were excluded from the study (3 discontinued MRS scanning and 1 had incorrect data concerning MRS results). Finally, a total of 39 patients were evaluated. All patients and control subjects underwent a detailed physical and neurological examination, as well as clinical assessment to exclude any neurological or comorbid conditions. Subjects who have clinically significant major depressive disorder, bipolar disorder and/or psychotic disorder, who have history of clinically significant personality disorder during clinical interview, who have history of substance abuse or addiction, convulsion or another neurological disorder or closed head trauma together with loss of conscious, and who have mental retardation were excluded from the study.

The study protocol was approved by the local Ethics Committee of University (protocol code: 2016-06). The study was carried out in accordance with the principles of "Helsinki Agreement on Human Rights -2013 version" and "Good Clinical Practice". The study was financially supported by Inonu University Scientific Research Projects within the scope of the project no. 2018/994.

MRS neuroimaging procedures

All patients and controls underwent MRS imaging at the University School of Medicine, Radiology Department on a 3T MR device (MagnetomSkyra-Siemens, Erlangen, Germany) using of a 20-channel phase array head coil. To determine voxel localisation and exclude parenchymal lesions, T1-weighted sagittal three-dimensional magnetisation-prepared rapid-acquisition gradient echo and T2-weighted fluid-attenuated inversion recovery axial-sagittal images were obtained, respectively (TR: 2,300 ms; TE: 2.98 ms; slice thickness: 1 mm; FOV 256 mm; matrix size: 240×256). After ruling out pathological lesions on the T2 sequence, thin slice images (1 mm) at three orthogonal planes (sagittal, axial and coronal) were obtained by multiplanar reconstruction. We placed a single $15 \times 20 \times 15$ mm volume of interest in left hippocampus and amygdala avoiding cerebrospinal fluid contamination. Additional "manual shimming" was made to enhance local magnetic homogeneity in the voxel. Subsequently, single voxel spectroscopy-short echo spectroscopic imaging was performed using a point-resolved spectroscopy sequence (TR: 2000, TE: 30). After the imaging procedure, the spectroscopic data were transferred to a work station, and peak metabolite ratios were calculated automatically using software (syngo.via Client 5.1, Siemens). The integral values of the metabolites were proportionated to that of Cr, which was used as the reference

metabolite (Figure 1, 2) [10].

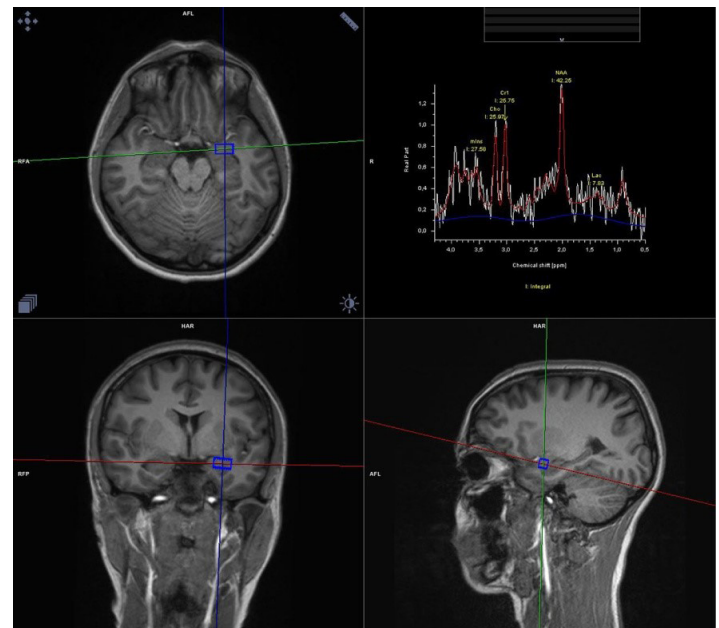


Figure 1. ROI placement and proton magnetic resonance spectra derived from amygdala

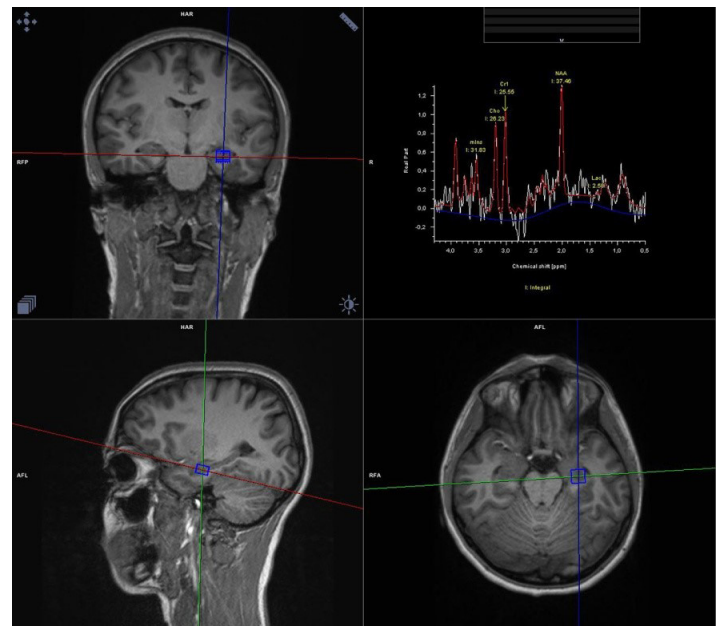


Figure 2. ROI placement and proton magnetic resonance spectra derived from hippocampus

Statistical Analysis

Statistical analyses were performed using the Statistical Program for Social Sciences for Windows, version 17.0, software (SPSS Inc., Chicago, IL, USA). Normality of the data distribution was assessed using the Shapiro-Wilk test. The data were summarized with median, minimum and maximum values and Mann Whitney U test was used for comparison because data not following a normal distribution. Categorical variables were indicated by number and percentage. Pearson exact test was used in the comparisons. A p-value < 0.05 was considered significant.

Results

A total of 39 vaginismus patients and 26 sexually active women without vaginismus (as the control group) were included into the study. The median age was 27 (19-45) years in the study group and 28 (22-41) years in the control group with no statistically significant difference determined between the groups ($p=0.125$). There was significant difference between the groups in terms of education status ($P=0.008$). Of the vaginismus group, 20.5% were

intermediate school graduates, 43.6% were high school graduates, and 35.9% were university graduates. Of the control group, 30.8% were high school graduates and 69.2% were university graduates.

There was no difference between the groups in terms of the concentration of metabolites in the amygdala, whereas median cholin (Cho) concentration in the hippocampus was significantly higher in the vaginismus group ($p=0.013$). The groups were similar in terms of other parameters (table 1).

Table 1. Comparison of the concentrations of the metabolites in the amygdala and hippocampus regions between the groups

		Patient Group (n=39)			Control Group (n=26)			P value
		Median	Minimum	Maximum	Median	Minimum	Maximum	
AGE		27.00	19.00	45.00	28.00	22.00	41.00	0.125
	NAA/CR	1.64	0.0001	2.62	1.75	0.8800	3.25	0.233
	CHO/CR	1.16	0.3000	2.98	0.96	0.7700	2.10	0.748
	MLNS/CR	1.30	0.0001	10.17	1.30	0.4900	2.58	0.574
AMYGDALA	LAC/CR	0.20	0.0001	1.35	0.29	0.0001	1.32	0.148
	GLU/CR	1.99	0.0001	4.98	1.84	0.0001	3.03	0.399
	GLN/CR	0.50	0.0001	3.22	0.00	0.0001	2.05	0.054
	GSH/CR	1.09	0.0001	2.64	0.87	0.0001	2.98	0.315
	NAA/CR	1.46	0.7500	2.06	1.33	0.6600	2.36	0.213
HIPPOCAMPUS	CHO/CR	0.97	0.3500	2.24	0.76	0.4400	1.44	0.013
	MLNS/CR	1.30	0.0001	2.74	1.41	1.0300	3.42	0.322
	LAC/CR	0.14	0.0001	1.26	0.13	0.0001	0.89	0.324
	GLU/CR	1.86	0.0001	4.34	1.72	0.0001	2.86	0.272
	GLN/CR	0.23	0.0001	4.91	0.09	0.0001	2.60	0.978
LDL-C, mg/dl	GSH/CR	0.86	0.0001	3.08	1.09	0.0001	1.55	0.639

n= number of samples

Discussion

As far as we know, this is the first study investigating the concentrations of metabolites in the amygdala and hippocampus of vaginismus patients. Our results indicated that the concentration of CHO is higher in the hippocampus of women suffering from vaginismus.

Cho, hippocampal concentration of which is high in vaginismus patients, is the precursor for the synthesis of phospholipids in human brain and has a series of important functions like cholesterol transport [11]. In addition, choline is necessary also for the synthesis and secretion of acetylcholine, which is critical neurotransmitter mediating memory storage [12,13]. Cho has significant influence on the developing brain, and any Cho disorder is potentially devastating. Increased Cho concentration in adults has been associated with numerous neurodegenerative conditions including multiple sclerosis and adrenoleukodystrophy [14]. It has been suggested that higher Cho concentrations indicate increased cell membrane transportation [15]. Several MRS studies on anxiety patients report elevated Cho in certain regions of the brain. According to the authors, this might be associated with the changes in myelination or signal transduction [16]. This is consistent with the results of the present study. High concentrations of Cho in the hippocampus indicate impaired

metabolism in this region of the patients with vaginismus, which is an anxiety-related condition.

Fear, which takes place in the symptomatology of many psychiatric disorders, is the basis also for vaginismus. It indicates risk and directs the adaptable behavior. Reviews and meta-analysis of neuroimaging studies concerning classical fear conditioning emphasize the connection of amygdala with insula and cingulate cortex. It is activated in case of delayed outcome predicted by hippocampus [17]. Conditioning experiments show that response of amygdala to repeated conditional threats decreases in time [18]. As the consequence; hippocampus is deemed responsible for fear-related learning and contextual processing [19]. Literature reports indicate hippocampal functional and volume abnormalities and neuronal volume loss associated with fear memory in post-traumatic stress disorder (PTSD) [20,21]. Similarly, metabolic alterations in hippocampus of vaginismus patients might be the neurobiological reflections of behavioral reactions due to fear conditioning occurring against sexual penetration.

An important feature of our results is the fact that they have been obtained from a patient sample with "pure" vaginismus without past or current major psychiatric comorbidity and they were compared with age-matched controls. However, the present study has some limitations. First, although major psychiatric situations

have been stated as the study exclusion criteria, the degree of subclinical depression or anxiety symptoms might have been effective on the outcomes. Second, sample size of the study group is small. Third, we evaluated only the hippocampus and amygdala regions associated with anxiety and fear but not the other cerebral regions that could be associated with anxiety and fear. Fourth, cross-sectional design of the study limits the interpretation of the results. Therefore, results of a longitudinal study where the patients are screened for several times can be interpreted more clearly.

Conclusion

In conclusion, the results of the present study reveal metabolic abnormality in the hippocampal choline concentration in vaginismus patients. These changes can be considered to play a role in the etiopathogenesis of vaginismus. Anxiety is a large group and it is not possible to generalize that increase of Cho is specific; may be categorized in subtypes. Repeated studies are needed to investigate whether these results are coincidental. However, the facts that this study is the first and may be pioneer for further studies make our results important.

Conflict of interests

The authors declare that they have no conflict of interest and any financial disclosures.

Financial Disclosure

All authors declare no financial support.

Ethical approval

The study protocol was approved by the local Ethics Committee of University (protocol code: 2016-06).

References

- Gyuris J. Sexological problems in neurological disorders: neurosexology. *Ideggyogy Sz.* 2009;62:292-7.
- Mernone L, Fiacco S, Ehlert U. Psychobiological Factors of Sexual Functioning in Aging Women - Findings From the Women 40+ Healthy Aging Study. *Front Psychol.* 2019;10:546.
- Kadir ZS, Sidi H, Kumar J, et al. The Neurobiology and Psychiatric Perspective of Vaginismus: Linking the Pharmacological and Psycho-Social Interventions. *Curr Drug Targets.* 2018;19:916-26.
- American Psychiatric Association. *Diagnostic and Statistical Manual of Mental Disorders*, 5th ed. Washington D.C., 2013. APA.
- Boyer SC, Goldfinger C, Thibault-Gagnon S, et al. Management of female sexual pain disorders. *Adv Psychosom Med.* 2011;31:83-104.
- Strakowski SM, Adler CM, DelBello MP. Volumetric MRI studies of mood disorders: do they distinguish unipolar and bipolar disorder? *Bipolar Disord.* 2002;4:80-8.
- Mizoguchi K, Ishige A, Aburada M, et al. Chronic stress attenuates glucocorticoid negative feedback: involvement of the prefrontal cortex and hippocampus. *Neuroscience.* 2003;119:887-97.
- Widman AJ, Cohen JL, McCoy CR, et al. Rats bred for high anxiety exhibit distinct fear-related coping behavior, hippocampal physiology, and synaptic plasticity-related gene expression. *Hippocampus.* 2019;29:939-56.
- Tkac I, Öz G, Adriany G, et al. In vivo 1H NMR spectroscopy of the human brain at high magnetic fields: Metabolite quantification at 4T vs. 7T. *Mag Reson Med.* 2009;62:868-79.
- Angelic E, Bonmartin A, Boudraa A, et al. Regional differences and metabolic changes in normal aging of the human brain: Proton MR spectroscopic imaging study. *AJNR Am J Neuroradiol.* 2001;22:119-27.
- Zeisel SH and da Costa KA. Choline: an essential nutrient for public health. *Nutr Rev.* 2009;67:615-23.
- Zeisel SH. The fetal origins of memory: the role of dietary choline in optimal brain development. *J Pediatr.* 2006;149:131-6.
- Bekdash RA. Choline, the brain and neurodegeneration: insights from epigenetics. *Front Biosci.* 2018;23:1113-43.
- Murata T, Kimura H, Kado H, et al. Neuronal damage in the interval form of CO poisoning determined by serial diffusion weighted magnetic resonance imaging plus 1H-magnetic resonance spectroscopy. *J Neurol Neurosurg Psychiatry.* 2001;71:250-3.
- Ende G, Braus DF, Walter S, et al. The hippocampus in patients treated with electroconvulsive therapy: a proton magnetic resonance spectroscopic imaging study. *Arch Gen Psychiatry.* 2000;57:937-43.
- Trzesniak C, Araujo D, Crippa JAS. Magnetic resonance spectroscopy in anxiety disorders. *Acta Neuropsychiatrica.* 2008;20:56-71.
- Sehlmeyer C, Schöning S, Zwieterlood P, et al. Human fear conditioning and extinction in neuroimaging: a systematic review. *Plos One.* 2009;4:e5865.
- Phelps EA, O'Connor KJ, Gatenby JC, Gore JC, et al. Activation of the left amygdala to a cognitive representation of fear. *Nat Neurosci.* 2001;4:437-41.
- Maren S, Phan KL, Liberzon I. The contextual brain: implications for fear conditioning, extinction and psychopathology. *Nat Rev Neurosci* 2013;14:417-28.
- Brown S, Freeman T, Kimbrell T, et al. In vivo proton magnetic resonance spectroscopy of the medial temporal lobes of former prisoners of war with and without post-traumatic stress disorder. *J Neuropsychiatry Clin Neurosci.* 2003;15:367-70.
- Joshi SA, Duval ER, Kubat B, et al. A review of hippocampal activation in post-traumatic stress disorder. *Psychophysiology.* 2019;4:e13357.