ORIGINAL ARTICLE

Annals of Agricultural and Environmental Medicine
www.aaem.pl

Brain-derived neurotrophic factor and matrix metalloproteinase-9 activity during rehabilitation therapy of schizophrenic patients – environmental pilot study

Renata Markiewicz[1,A-D,F], **Agnieszka Anna Markiewicz-Gospodarek**[1,B-D,F], **Beata Dobrowolska**[1,C-D,F] ✉, **Iwona Bojar**[2,C-D,F], **Bartosz Łoza**[3,A,C-F]

[1] *Medical University, Lublin, Poland*
[2] *Department of Women's Health, Institute of Rural Heath, Lublin, Poland*
[3] *Department of Psychiatry, Faculty of Medicine, Medical University, Warsaw, Poland*
A – Research concept and design, B – Collection and/or assembly of data, C – Data analysis and interpretation, D – Writing the article, E – Critical revision of the article, F – Final approval of the article

Markiewicz R, Markiewicz – Gospodarek AA, Dobrowolska B, Bojar I, Łoza B. Brain-Derived Neurotrophic Factor and Matrix Metalloproteinase-9 activity during rehabilitation therapy of schizophrenic patients – environmental pilot study. Ann Agric Environ Med. doi: 10.26444/aaem/163327

Abstract

Introduction and Objective. The aim of the study was to evaluate the peripheral level of brain-derived neurotrophic factor (BDNF) and matrix metalloproteinase-9 (MMP-9) during rehabilitation therapy, combined with neurofeedback in schizophrenic patients, and to investigate whether these biomarkers are related to psychopathological symptoms, changes in auditory evoked potentials (AEPs), and quantitative EEG (QEEGs) mapping.

Materials and method. The study involved two groups of patients diagnosed with paranoid schizophrenia in partial remission who participated in a 3-month structured rehabilitation programme combined with neurofeedback (REH group) and a standard support group (CON group). The following parameters were assessed: BDNF and MMP-9 serum levels, AEPs, QEEGs, and psychopathological symptoms (PANSS).

Results. A clinical improvement within the 3-month rehabilitation therapy course was correlated with the increase in BDNF and MMP-9 serum level. Despite the increase in BDNF and MMP-9 during the 3-month rehabilitation therapy, it was not possible to demonstrate any strong and significant correlation between the 2 examined neuropeptides. During the 3-month rehabilitation therapy, the theta waveform share reduction in QEEG, P50 latency reduction and amplitude increase correlated with PANSS Total and MMP-9 results.

Conclusions. All clinical (PANSS Positive, Negative, General, Total) and biochemical results (BDNF, MMP-9) of the REH group changed significantly over the 3-month period. Positive symptoms improved only in the CON group.

Key words

schizophrenia, rehabilitation, brain-derived neurotrophic factor, matrix metalloproteinase-9, neurofeedback, clinical trial

Abbreviations

AEPs – auditory evoked potentials; **BDNF** – brain-derived neurotrophic factor; **CNS** – central nervous system; **DC** – direct current; **EEG** – electroencephalogram; **GSR** – galvanic skin response; **ICD** – international classification of diseases; **MMP-9** – matrix metalloproteinase-9; **NF** – neurofeedback; **NPS** – neuropeptide S; **PANSS** – positive and negative syndrome scale; **SCL** – skin conductance level; **SCRs** – skin conductance responses; **TIMP** – tissue inhibitor of metalloproteinase-9; **QEEG** – quantitative electroencephalogram

INTRODUCTION

Various abnormalities of the neuropeptide system have been found in patients with a diagnosed with schizophrenia [1–5]. Currently, there is no evidence of either a primary or secondary relationship between neuropeptides and the symptomatology of schizophrenia, which is closely related to its etiopathogenesis. Neuropeptide hypotheses are part of a larger group of the etiopathogenetic concepts of schizophrenia related to various disruptions in neuronal signalling in the central nervous system (CNS), involving neurotransmitters, neuropeptides, hormones, cytokines, or other systems. There are differences in the function of both neuropeptides and neurotransmitters, which include different activity, response, and site of action. Neuropeptides show a slower action that develops gradually, causing an apparent change in the modulation of the regulatory mechanism of gene expression and metabolic pathways.

A meta-analysis reported that the level of peripheral brain-derived neurotrophic factor (BDNF) might be changed in schizophrenia, however, with considerable heterogeneity in the serum results and the lack of BDNF level studies depending on the treatment phase [6]. BDNF, a key regulator of synaptic plasticity, is initially synthesized as a precursor protein preproBDNF, and finally proBDNF is transformed into mature BDNF by extracellular protease, such as matrix metalloproteinase-9 (MMP-9) [2]. Also, MMP-9 is converted from its inactive form (pro-MMP-9) into active MMP-9 under

✉ Address for correspondence: Beata Dobrowolska, Medical University, Lublin, Poland
E-mail: beata.dobrowolska@umlub.pl

Received: 26.01.2023; accepted: 15.04.2023; first published: 02.05.2023

the influence of the tissue inhibitor of metalloproteinase (TIMP) [2, 7–14]. Abnormal MMP-9 activity may influence BDNF and hypothetically lead to the deficit syndrome which is the holy grail of the neurodevelopmental theory of schizophrenia [13, 15–20]. Some specific BDNF and MMP-9 genotypes show significantly higher Positive and Negative Syndrome Scale (PANSS) scores and could be understood as the biomarkers of schizophrenia patients [5].

Schizophrenia is a chronic, relapsing mental illness with a gradual course. It is characterized by the development of cognitive, social, educational and occupational deficits [3,13]. In addition, there are disturbances in thinking, perception, communication, as well as shallowed and maladjusted affect (emotional expression). Approximately half of the patients remain in a residual or actively psychotic state, despite pharmacological treatment [21]. Patients with a diagnosis of schizophrenia should be offered other support programmes, such as cognitive training or more complex rehabilitation therapies.

The effectiveness the above methods is related to the mechanism of stimulation which results in the restricting of neuronal connections [22–24]. Reorganization of neuronal circuits (anatomical) is dependent on biochemical changes and changes in action potential in dendritic spikes (spourting). These changes result in an increased release of neurotransmitters, thus enabling the formation of new connections based on long-term potentiation [22–26].

Optimal treatment of schizophrenia in different settings (urban, rural) remains a strategic challenge and should include various and specific psychosocial interventions in addition to the optimal use of medications, with the aim of improving the well-being of patients from the above-mentioned settings [27]. Intensive rehabilitation, especially using the neurofeedback (NF) technique on the levels of neuropeptides such BDNF or neuropeptide S (NPS) and its relationship to the patient's clinical condition, has already been demonstrated in previous human studies [4, 23, 28].

The aim of the present study was to demonstrate the potential associations of peripheral levels of BDNF and MMP-9 with clinical parameters during structured rehabilitation therapy in patients with a diagnosis of schizophrenia.

MATERIALS AND METHOD

Study design and participants. The study was a randomized, controlled 3-month trial reported with the use of CONsolidated Standards of Reporting Trials (CONSORT) guidelines [29]. The trial was registered in the ISRCTN registry (Trial ID: ISRCTN78612833). Forty-two male patients with paranoid schizophrenia (according to ICD-10-DCR participated in the study [30]). The sample size (N) was calculated for the test power in the range not lower than 0.8 [31]. The decision to include only men in the study was made based on the available publications that verify differences in the level of BDNF [32–34], MMP-9 [35, 36] and TIMP [36], depending on gender. Analysis of these markers in a group of women requires taking into account the phases of the menstrual cycle [37, 38].

The men included in the study were divided into two groups: 1) consisted of patients who underwent intensive rehabilitation (N=17, REH); 2) patients who received standard rehabilitation, mainly social support (N=25, CON). All patients included in the study continued pharmacological treatment (atypical anti-psychotics – equivalents to a daily dose of olanzapine in mg: REH vs CON: M 18.74 SD 5.16 vs M 19.62 SD 6.06) [39]. None of the patients took anti-cholinergics.

The exclusion criteria were lack of patients' consent, female gender, clinical diagnosis other than of paranoid schizophrenia [ICD], age below 18 and over 50, left-handedness (writing), current neurological diseases, mental disability, or alcohol and/or psychoactive sub-stance addiction, unstable condition, i.e., active psychotic episodes for more than 18 months.

The REH and CON groups did not differ significantly in terms of basic variables (Tab. 1, Fig. 1, Fig. 2). Detailed clinical and demographic characteristics are presented in Table 1.

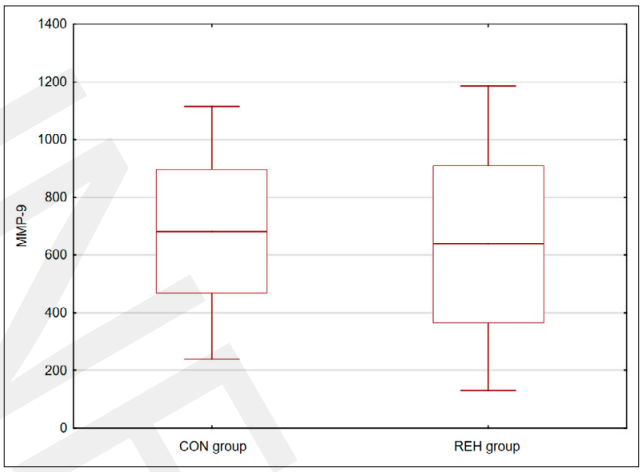

Figure 1. MMP-9 initial results: REH group s. CON group means and standard deviations

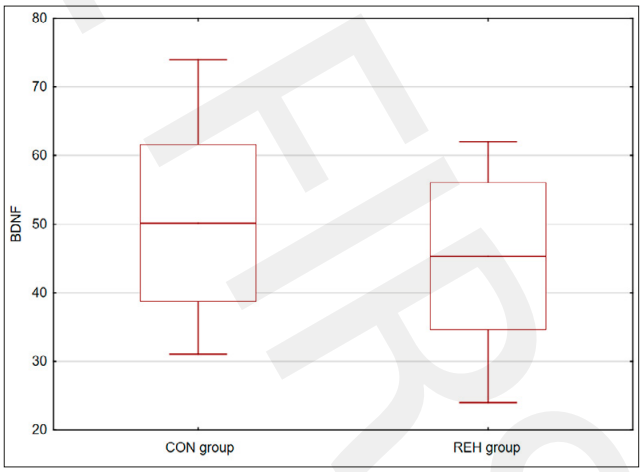

Figure 2. BDNF initial results: REH group vs. CON group means and standard deviations

Measurement indicators. In order to assess clinical, biochemical and electrophysiological parameters as a result of rehabilitation interventions, the following were included in the analysis:

1) scale of Positive and Negative Teams (PANSS) [40];
2) level of markers – BDNF and MMP-9 in the blood serum of patients tested on an empty stomach at 7:00.

Table 1. Initial (T1) parameters and pairwise comparisons (t test/Mann-Whitney test) for REH and CON groups

Variable	REH		CON		REH vs CON	
	M	SD	M	SD	t^t/U^U	p
BDNF (ng/ml)	45.35	10.73	50.16	11.38	1.37^t	0.177
MMP-9 (ng/ml)	638.23	271.78	681.91	213.56	0.58^t	0.564
PANSS-Positive	9.24	0.97	9.20	1.63	193.50^U	0.636
PANSS-Negative	14.06	2.49	15.08	3.43	1.05^t	0.299
PANSS-General	24.94	2.02	26.32	2.55	1.87^t	0.069
PANSS-Total	48.23	4.19	50.60	5.71	1.46^t	0.152
Age (years)	38.00	5.62	35.68	8.28	-1.01^t	0.320
Age of first hospitalization (years)	26.77	4.93	24.60	5.45	-1.28^T	0.205
Education (years)	12.47	2.89	14.00	1.80	150.00^U	0.112
Antipsychotics in milligrams (equivalents of olanzapine)	18.74	5.16	19.62	6.06	206.50^U	0.888

REH – rehabilitation group; CON – control group; BDNF – brain-derived neurotrophic factor; MMP-9 - matrix metalloproteinase-9; PANSS – Positive, Negative, General and Total: results of Positive and Negative Syndrome Scale; M – mean; SD – standard deviation; t – Student's t-test; U – Mann-Whitney U-test; p – p-value significance at $p < 0.05$.

Serum BDNF levels were analyzed using the ELISA test (Human BDNF ELISA Kit (R&D Systems, Minneapolis, MN, USA), and MMP-9 was tested using the Human MMP-9 ELISA Kit (Biorbyt, Cambridge, UK). Additionally, 30 electrophysiological measurements were made:

a) auditory evoked potentials (AEP) were analyzed based on the Cognitrace system, in accordance with the international system 10–20); all patients stayed in a separate room during the examination, with the lights turned off. The examination was carried out in a sitting position, with eyes closed, in headphones through which acoustic stimuli were emitted according to the oddball paradigm scheme (tones with a frequency of 1,000 Hz – 2,000 Hz, values of 70 dB for 100 ms in a random sequence). One test lasted 3 minutes and 20 seconds and contained 80% of frequent stimuli and 20% of rare (target) stimuli, to which the subject responded by pressing a button;
b) quantitative electroencephalography (QEEG) [41] was performed using the EEG Digi-Track apparatus to map and meta-analyze frequency bands (delta, theta, alpha, beta SMR, beta1/beta2, gamma), and attention factors (theta/beta) concentration (theta/SMR), tension (SMR/beta2), sensory and motor activity (alpha/SMR), executive functions (alpha/beta), thinking and acting (beta/alpha) in the area of F-z and C-z.

All examinations were performed twice in each group, at the beginning (T1) and at the end of the 3-month period (T2).

Rehabilitation training. The aim of the rehabilitation programme was to strengthen independence and modify everyday activities, improve social relations, and increase self-esteem. The classes took place for 8 hours a day (except weekends) and included group activities: assertiveness training, role-playing techniques, psychotherapy, psychoeducation, cognitive training, art therapy, physiotherapy, sports, social events, entertainment and culinary activities, and relaxation training. The scheme of the programme was based on selected principles of cognitive remediation developed by Wykes [42]. The original scheme used in the work focused mainly on learning to acquire skills and improving metacognition and solving social difficulties. Its assumption was to improve the patient's functioning in society [42] through the use of modules: social training, motivation/planning skills, cognitive training, computer-assisted training (perception, attention, reasoning) and a creativity module.

Computer-assisted cognitive training was based on neurofeedback training as an additional form of rehabilitation, in accordance with the methods of Markiewicz et al. [43, 44].

The main assumption of the trainings was to gradate the difficulty of the tasks, taking into account the individual abilities of each patient. The neuro-feedback training used the method of galvanic skin response (GSR) with the use of the Digi-Track device (EEG-DigiTrack Biofeedback-EEG + SpO2 + HR, Warsaw, Poland) [45] and 3 modules: relaxation (CENTRUM), concentration (BALANCE) and improving executive functions (insects).

The aim of the exercises on the CENTRUM module was to achieve relaxation by controlling/modulating breathing and heart rate. The greater the relaxation, the faster the patient reaches the next level of difficulty. Training on the BALANCE module was aimed at improving concentration by placing and balancing the ball in the middle of the tilted board. The aim of training on the INSECT module was to achieve a state of internal balance between cognitive and executive functions. The task of the subjects was to recognize moving and hidden insects on the monitor screen and clicking on them with the mouse. The slow movement of the insects reflected the achievement of internal balance during training. The time of each exercise was determined by a computer programme, for the CENTRUM and BALANCE modules it was 5 minutes, for the INSECTS module 10 minutes.

Rehabilitation therapy for the CON group consisted of primary care, counseling, medication management, unstructured meetings and interviews, meal offerings, administrative assistance, and social worker support.

Statistical analysis. The values of the analyzed variables are presented as means (M) and standard deviations (SD). Sociological and demographic data are presented in numerical and percentage form. The results were compared using the Student's t test for related samples, the non-parametric Mann-Whitney U test and the Pearson product correlation coefficient r. The Shapiro-Wilk test was used to check the normal distribution. Differences were considered statistically significant at $p < 0.05$. Analyses were performed using the Statistica 13.3 programme.

Ethical issues. The study protocol was approved by the local Bioethics Committee (Approval No. KE-0254/35/2016). All patients invited to take part in the study gave their written informed consent.

RESULTS

The baseline (T1) and 3-month (T2) clinical and biochemical results of rehabilitation therapy (REH group) versus standard therapy (CON group) programmes are presented in Table 2.

During the 3-month period, all clinical and biochemical results of the REH group changed significantly In contrast, in the CON group, only a statistical improvement in positive symptoms was observed (Tab. 2).

Table 2. T1 versus T2 clinical and biochemical results

Test	Subtest	Group	Baseline (T1)		3-month (T2)		t/U	p
			M	SD	M	SD		
PANSS	Positive	REH	9.24	0.97	7.47	0.80	26.00[U]	**0.000**
		CON	9.20	1.63	8.24	1.35	134.50[U]	**0.047**
	Negative	REH	14.06	2.49	11.53	2.32	3.07[t]	**0.005**
		CON	15.08	3.43	14.47	2.96	0.60[t]	0.554
	General	REH	24.94	2.02	22.18	3.43	61.50[u]	**0.005**
		CON	26.32	2.55	25.88	4.86	0.38[t]	0.705
	Total	REH	48.23	4.19	41.18	4.64	4.66[t]	**0.001**
		CON	50.60	5.71	48.59	7.97	0.96[t]	0.346
Neuro-peptides	BDNF	REH	45.35	10.73	56.24	10.62	-2.97[t]	**0.006**
		CON	50.16	11.38	55.29	10.27	-1.49[t]	0.144
	MMP-9	REH	638.23	271.78	757.26	369.21	-2.11[t]	**0.045**
		CON	681.91	213.56	807.26	460.70	205.00[U]	0.858

REH – rehabilitation group; CON – control group; BDNF – brain-derived neurotrophic factor; MMP-9 - matrix metalloproteinase-9; PANSS – Positive, Negative, General and Total: results of Positive and Negative Syndrome Scale; M – mean; SD – standard deviation; t – Student's t-test; U – Mann-Whitney U-test; p – p-value significance at $p < 0.05$.

The REH and CON results were then compared for the magnitude of changes of pre- and post-therapy results (T2–T1 differences). Changes that occurred over time from T1 to T2 differentiated significantly for the REH vs. CON 3-month PANSS Total, PANSS Negative and BDNF results (Tab. 3). Although this is only statistically true for the Negative subsyndrome, the tendency is clear for all 3 PANSS subsyndromes, resulting in the Total score being significantly different between REH and CON.

Table 3. Differences in the magnitude of change from pre- (T1) to post-therapy (T2) results in REH and CON groups

Test	Subtest	REH (T2-T1)		CON (T2-T1)		In-between comparisons	
		M	SD	M	SD	t / U	p
PANSS	Positive	-1.77	0.83	-1.04	1.46	144.50[U]	0.084
	Negative	-2.53	1.46	-1.04	2.32	126.00[U]	**0.028**
	General	-2.77	2.36	-0.32	4.86	148.00[U]	0.101
	Total	-7.06	2.95	-2.40	6.12	110.50[U]	**0.009**
Neuro-peptides	BDNF (ng/ml)	10.88	7.55	2.80	8.89	-3.07[t]	**0.004**
	MMP-9 (ng/ml)	119.04	272.46	82.68	314.79	194.00[U]	0.645

REH – rehabilitation group. CON – control group. BDNF – brain-derived neurotrophic factor. MMP-9 - matrix metalloproteinase-9; Positive. Negative. General and Total - results of Positive and Negative Syndrome Scale; M – mean; SD – standard deviation; t – Student's t-test; U – Mann-Whitney U-test; p – p-value significance at $p < 0.05$.

The biochemical (BDNF, MMP-9) and clinical (PANSS) results for REH group were correlated (Tab. 4).

Only those correlations that were negative were significant between PANSS and both neuropeptides during treatment (Tab. 4). The T2-T1 BDNF and MMP-9 results correlated with the Positive subsyndrome (mainly delusions and hallucinations) but most of all with the Total PANSS scores. In other words, a significant increase in BDNF and MMP-9 (T2-T1) was associated with comprehensive clinical improvement. There was also a strong negative correlation between T2-T1 BDNF (but not MMP-9) and negative symptoms. Finally, it should be stated that the interrelationships of BDNF and MMP-9 were not significant in any measurement (Tab. 4).

Table 4. The Pearson's r product-moment correlation coefficients for REH group (only): BDNF and MMP-9 results correlated with clinical variables. Significant ($p<0.05$) and strong correlations for absolute values of $r>0.5$ were bolded

Test	Subtest	BDNF			MMP-9		
		T1	T2	T2-T1	T1	T2	T2-T1
PANSS T1	Positive	-0.10	-0.32	**-0.63**	0.21	-0.44	**-0.86**
	Negative	0.50	-0.08	**-0.86**	0.58	0.46	-0.05
	General	0.24	-0.14	-0.55	0.44	0.07	-0.42
	Total	0.34	-0.14	**-0.69**	0.55	0.15	-0.45
PANSS T2	Positive	-0.48	**-0.73**	-0.23	0.18	-0.35	**-0.69**
	Negative	0.32	-0.01	-0.50	0.43	0.36	-0.01
	General	-0.10	-0.47	-0.48	-0.04	-0.33	-0.40
	Total	-0.04	-0.50	**-0.61**	0.18	-0.17	-0.45
BDNF	T1	1.00	**0.76**	-0.52	0.37	0.58	0.50
	T2		1.00	0.16	0.14	0.42	0.42
	T2-T1			1.00	-0.38	-0.48	-0.20
MMP-9	T1				1.00	**0.71**	-0.21
	T2					1.00	0.54
	T2-T1						1.00

BDNF – brain-derived neurotrophic factor; MMP-9 - matrix metalloproteinase-9; Positive. Negative. General and Total – results of Positive and Negative Syndrome Scale. Repeated values for BDNF x MMP-9 correlations were removed

In order to analyze the relationships between electrophysiological variables (QEEGs and AEPs latencies and amplitudes) and clinical variables (PANSS), as well as BDNF and MMP-9, correlations of differences between the results before and after the 3-month rehabilitation therapy were performed. Insignificant relationships were omitted (Tab. 5).

Table 5. The Pearson's r product-moment correlation coefficients for REH group (only). Differences between T2 and T1: electrophysiological results (QEEGs and AEPs) vs. BDNF. MMP-9 and Total PANSS scores. Only significant ($p<0.05$) and strong correlations for absolute values of $r>0.5$ were presented

Variables	Waveform/Potential		Electrodes	Total PANSS	MMP-9	BDNF
QEEGs	theta		Fz		-0.58	
			Cz		-0.53	
	theta/alpha		Fz	0.61		
			Cz	0.62		
AEPs	P50	latency	Fz	0.63		
			Cz	0.60		
		amplitude	Fz		0.72	
			Cz		0.64	
	N1	amplitude	Cz	0.55		
	P2	amplitude	Fz		0.56	
	N2	latency	Fz		0.60	
		amplitude	Cz	0.56		

AEP - Auditory Evoked Potential; QEEG - Quantitative Electroencephalography; Total PANSS – total results of Positive and Negative Syndrome Scale; MMP-9 - matrix metalloproteinase-9; BDNF – brain-derived neurotrophic factor

Among several dozen QEEG indices, only the scores of theta waves or theta to alpha ratio were significantly correlated with the severity of clinical symptoms (PANSS), as well as inversely correlated with the increase in MMP-9.

Among AEPs, consistent results were obtained only for P50, where the greater reduction in latency was proportional to the decrease in severity of clinical symptoms (PANSS), while the increase in amplitude was accompanied by increase in the MMP-9 level.

Some significant, but only single correlations for N1, P2 and N2 were also obtained. No changes in QEEGs or AEPs were associated with changes in BDNF level between T1 and T2.

DISCUSSION

There is growing evidence that rehabilitation, psycho-education, cognitive exercises, vocational and social skills trainings, can effectively enhance the psycho-social functioning of patients with schizophrenia spectrum disorders [46]. Rehabilitation programmes are not only the vehiculum for cognitive or psychosocial stimulation, but they also go beyond THE simple compensation of psychotic deficits and have the potential to improve more general symptoms and syndromes of schizophrenia [4, 47].

All PANSS subsyndromes and its total score, as well as the biochemical (BDNF, MMP-9) results of the REH group, improved over the 3-month rehabilitation therapy. In contrast, in the CON group only a statistical improvement in positive symptoms was observed. Changes that occurred over time from T1 to T2 were differentiated significantly for the REH vs. CON 3-month PANSS Total and BDNF results, but not for MMP-9. Several studies pooling BDNF and MMP-9 data, as well as clinical variables, can be found [2, 5, 13, 48, 49].

In the study by Niitsu et al. [13], no general differences in BDNF and MMP-9 levels were found between patients and healthy controls. Similarly, previous research by the authors of the current study, found no difference between patients and healthy controls in terms of BDNF, but the treatment changed this situation. Also, for the current study, a one-time measurement was not performed, as it was a 3-month follow-up with a control group of patients treated as usual, which led to changes in the level of neuropeptides (Tab. 2, Tab. 3). On the other hand, the study by Niitsu et al. [13] proved a selective relationship between BDNF and negative symptoms of schizophrenia, similar to that found in the current study (Tab. 2). Niitsu et al. [13] stated that serum MMP-9 level of male patients who smoked was higher than those of non-smoking patients; however, this was not observed in male controls and female patients. The current *ad hoc* analysis revealed no differences in the MMP-9 level in the REH group for smokers and non-smokers (for T1, T2 and T1-T2, respectively; Student's t: -0.44, -.016, 0.22). Despite similar findings, the studies were methodologically different.

In Yamamori et al. [2], MMP-9 was increased in patients with schizophrenia but no change was found for mature BDNF; however, mature BDNF and MMP-9 plasma levels were significantly correlated. Neither mature BDNF nor MMP-9 plasma levels were associated with clinical variables. The trial was methodologically unique which made it difficult to compare the results. Measurements were made only in patients with drug-resistant schizophrenia treated with clozapine. Meanwhile, in the current study, the patients remained clinically stable at T1, neither drug-resistant, nor treated with clozapine. Since only drug-resistant patients were recruited in the study by Yamamori et al. [2], it would be difficult to expect even moderate variability on the side of clinical symptoms, *ergo* identification of their association with neuropeptides. And again, the one-time measurement cannot be compared to a 3-month trial in which patients participated in active treatment.

Pan et al. [5] examined the relationship between the single-nucleotide polymorphisms of BDNF and MMP-9 and the clinical variability of schizophrenia phenotype (PANSS). Some BDNF and MMP-9 genotypes were identified as significantly correlated to PANSS Total (both neuropeptides), to PANSS Positive (BDNF) or to PANSS General (MMP-9). Ali et al. [35] also proved the association of BDNF and MMP-9 specific single nucleotide polymorphisms with the higher expression of schizophrenia (PANSS). In this context, the male gender was identified as an independent predictive factor for a higher PANSS score. This was one of the reasons for including only males in the current study.

Romash et al. [48] postulated a negative correlation between BDNF and MMP-9 in paranoid schizophrenia. As the duration of the disease increased, BDNF activity decreased and MMP-9 expression increased, an imbalance that could be used as a marker of the disease process. The current study, however, did not demonstrate this type of negative correlation (Tab. 4), but in the context of the overall progressive biochemical and clinical results of the REH group, the lack of correlation between BDNF and MMP-9 in any of the parameters (T1, T2, T1-T2) opens possibilities for hypotheses analogous to those in Romash et al. [48].

Arabska et al. [49] found a decrease in BDNF in patients with schizophrenia – regardless of metabolic abnormalities, and an increase in MMP-9 – secondary to metabolic abnormalities. Of course, there is a consensus that MMP-9 is induced by many pro-inflammatory cytokines, including a rise in metabolic disorders. However, the inflammatory pattern of psychotic exacerbations is also known, therefore, the primary clinical impact of MMP-9 cannot be dismissed [50]. The current study which used a longitudinal 3-month trial differed in methodology from the study by Arabska et al. [49]. However, that the assessment of MMP-9 activity requires precise adjusting for many parameters, is accepted by the authors of the current study. Two groups of the authors' electrophysiological results clearly fit into the contemporary etiopathogenetic research on schizophrenia.

The scores of theta frequency and theta to alpha ratio were significantly correlated with the severity of clinical symptoms (PANSS), as well as inversely correlated with the increase in MMP-9. Meanwhile, as demonstrated experimentally by Adams et al. [51], fronto-temporal dysconnectivity – one of the key etiopathological hypotheses in schizophrenia – is the basis for the dysfunctional theta phase coupling between hippocampus / medial temporal lobe and the medial prefrontal cortex. Impaired neural synchrony in the theta frequency range is present in patients from the onset of the disease, and even as early as in adolescents at familial risk for schizophrenia [52].

Among AEPs in the current study, consistent results were obtained only for P50 potential, where the greater reduction in P50 latency was proportional to the decrease in severity of clinical symptoms (PANSS), while the increase in P50 amplitude was accompanied by increase in MMP-9 level. The P50 gating dysfunctions are widely understood as endophenotypes for schizophrenia [53, 54]. The deficits in

inhibitory gating of response to paired sensory stimuli using the auditory P50 evoking response in early sensory processing mechanisms, demonstrate the general model of attentional performance dysfunction in schizophrenia patients [55].

The problem of 100-year research on schizophrenia, on the one hand, is the deficit of etiopathogenetic findings and, on the other hand, the urgent need to improve comprehensive treatment, including environmental forms of rehabilitation and not just drug treatment. This leads to inevitable limitations in research and publications.

The current study confirmed the serums BDNF and MMP-9 increase as a phenomenon accompanying the clinical improvement during rehabilitation therapy of patients with schizophrenia. This relationship, based on patients' clinical improvement, enables better treatment planning and prognosis.

Neurofeedback training uses visual perception processes, including saccadic eye movements [56, 57]. As is known, about 80% of information is transmitted through the visual channel, which may have a significant impact on the results obtained in the REH group studied by the authors. This is explained, among other things, by the microgenetic theory of symptom formation and recovery from illness. It is worth mentioning that neurofeedback has been used successfully in the treatment of cognitive disorders in various disease entities [58, 59], including schizophrenia [60].

However, the current study has some clear limitations: only small groups consisting solely of men were recruited, and only the subtype episodic schizophrenia, with stable or progressive development of negative symptoms and focus on rehabilitation effects, was studied. This means that the verification of all conclusions requires an extension of the study. Nevertheless, the results are pioneering, the possible association of BDNF and MMP-9 (neuropeptides) with clinical objectives in schizophrenia should be carefully considered as an opportunity to meet the diagnostic and therapeutic needs of patients with schizophrenia.

CONCLUSIONS

All clinical (PANSS Positive, Negative, General, Total) and biochemical results (BDNF, MMP-9) of the REH group changed significantly over the 3-month period. Positive symptoms improved only in the CON group.

The structured rehabilitation therapy combined with cognitive enhancement training significantly reduced schizophrenia symptoms, compared to standard supportive therapy among patients from the different study communities.

The overall results should be interpreted with caution due to limitations in the size and gender of the participants – males only.

REFERENCES

1. Rodriguez B, Nani JV, Almeida PGC, Brietzke E, Lee RS, Hayashi MAF. Neuropeptides and oligopeptidases in schizophrenia. Neurosci Biobeh Rev. 2020;108:679–693. doi: https://doi.org/10.1016/j.neubiorev.2019.11.024
2. Yamamori H, Hashimoto R, Ishima T, et al. Plasma levels of mature brain-derived neurotrophic factor (BDNF) and matrix metalloproteinase-9 (MMP9) in treatment-resistant schizophrenia treated with olanzapine. Neurosci Letter. 2013;556:37–41. doi: https://doi.org/10.1016/j.neulet.2013.09.059
3. Peng S, Li W, Lv L, Zhang Z, Zhan X. BDNF as a biomarker in diagnosis and evaluation of treatment for schizophrenia and depression. Discovery Med. 2018;26(143):127–136.
4. Markiewicz-Gospodarek A, Markiewicz R, Dobrowolska B, Rahnama M, Łoza B. Relationship of neuropeptide S (NPS) with neurocognitive, clinical, and electrophysiological parameters of patients during structured rehabilitation therapy for schizophrenia. J Clin Med. 2022;11(18):5266. doi: https://doi.org/10.3390/jcm11185266
5. Pan L, Cao Z, Chen L, Qian M, Yan Y. Association of BDNF and MMP-9 single-nucleotide polymorphisms with the clinical phenotype of schizophrenia. Front Psychiatry. 2022;13:941973. doi: https://doi.org/10.3389/fpsyt.2022.941973
6. Nieto RR, Carrasco A, Corral S, Castillo R, Gaspar PA, Bustamante ML, Silva H. BDNF as a biomarker of cognition schizophrenia/psychosis: An updated review. Front Psychiatry. 2021;12:662407. doi: https://doi.org/10.3389/fpsyt.2021.662407
7. Bothwell M. Recent advances in understanding context-dependent mechanisms controlling neurotrophin signalling and function. F1000 Faculty Rev-1658. 2019;8. doi: https://doi.org/10.12688/f1000research.19174.1
8. Deyama S, Bang E, Kato T, Li XY, Duman RS. Neurotrophic and antidepressant actions of brain-derived neurotrophic factor require vascular endothelial growth factor. Biological Psychiatry. 2019;86(2):143–152. doi: https://doi.org/10.1016/j.biopsych.2018.12.014
9. Pijet B, Stefaniuk M, Kaczmarek L. MMP-9 contributes to dendritic spine remodeling following traumatic brain injury. Neural Plasticity. 2019;2019:3259295. doi: https://doi.org/10.1155/2019/3259295
10. Figiel I, Kruk PK, Zaręba-Kozioł M, Rybak P, Bijata M, Wlodarczyk J, Dzwonek J. MMP-9 signaling pathways that engage rho GTPases in brain plasticity. Cells. 2021;10(1):166. doi: https://doi.org/10.3390/cells1001066
11. Szepesi Z, Hostel E, Ruszczycky B, et al. Synaptically released matrix metalloproteinase activity in control of structural plasticity and the cell surface distribution of GluA1-AMPA receptors. PLoS One. 2014;9(5):e98274. doi: https://doi.org/10.1371/journal.pone.0098274
12. Lepeta K, Kaczmarek L. Matrix metalloproteinase-9 as a novel player in synaptic plasticity and schizophrenia. Schizoph Bull. 2015;41(5):1003–1009. doi: https://doi.org/10.1093/schbul/sbv036
13. Niitsu T, Ishima T, Yoshida T, Hashimoto T, Matsuzawa D, et al. A positive correlation between serum levels of mature brain-derived neurotrophic factor and negative symptoms in schizophrenia. Psychiatry Res. 2014;215(2):268–273. doi: https://doi.org/10.1016/j.psychres.2013.12.009
14. Luo L, Li C, Deng Y, Wang Y, Meng P, Wang Q. High-intensity interval training on neuroplasticity, balance between brain-derived neurotrophic factor and precursor brain-derived neurotrophic factor in poststroke depression rats. J Stroke Cerebrovascular Dis. 2019;28(3):672–682. doi: https://doi.org/10.1016/j.strokecerebrovasidis.2018.11.009
15. Li W, Kou C, Yu Y, et al. Association of catechol-o-methytransferase gene polymorphisms with schizophrenia and negative symptoms in a Chinese population. Am J Med Genet B Neuropsychiatr Genet. 2012;0(4):370–375. doi: https://doi.org/10.1002/ajmg.b.32038
16. Kaczmarek L, Łapińska-Dzwonek J, Szymczak S. Matrix metalloproteinases in the adult brain physiology: A link between c-Fos, AP-1, and remodelling of neuronal connections? EMBO J. 2002;21(24):6643–6648. doi: https://doi.org/10.1093/emboj/cdf676
17. Xia QR, Zhang C, Liang J, Xu YY. The association of functional polymorphism of matrix metalloproteinase-9 gene (rs3918242) with schizophrenia: A meta-analysis. Int J Psychiatry Clin Practice. 2019;23(3):207–214. doi: https://doi.org/10.1080/13651501.2019.1581895
18. Lepeta K, Purzycka K, Pachulska-Wieczorek K, et al. A normal genetic variation modulates synaptic MMP-9 protein levels and the severity of schizophrenia symptoms. EMBO Molecular Med. 2017;9(8):1100–1116. doi: https://doi.org/10.15252/emmm.201707723
19. Michaelsen K, Zagrebelsky M, Berndt-Huch J, et al. Neurotrophin receptors TrkB.T1 and p 75NTR cooperate in modulating both functional and structural plasticity in mature hippocampal neurons. Eur J Neurosci. 2010;32(11):1854–1865. doi: https://doi.org/10.1111/j.1460–9568.2010.07460.x
20. Cyran A, Piotrowski P, Samochowiec J, Grążlewski T, Misiak B. Risk factors of deficyt and non-deficit schizophrenia: Results from a cross-sectional study. Revista de Psiquiatria y Salud Mental (English Edition). 2022;15(4):223–229. doi: https://doi.org/10.1016/j.rpsmen.2022.05.001
21. Faden J, Citrome L. Resistance is not futile: Treatment-refractory schizophrenia-overview, evaluation, and treatment. Exp Opinion Pharmacotherapy. 2019;20(1):11–24. doi: https://doi.org/10.10180/14656566.2018.1543409

22. Markiewicz R. The use of EEG Biofeedback/Neurofeedback in psychiatric rehabilitation. Psychiatria Pol. 2017;51(6):1095–1106. doi: https://doi.org/10.12740/PP/68919
23. Markiewicz R, Kozioł M, Olajossy M, Masiak J. Can the neurotrophic factor BDNF be an indicator of effective rehabilitation influences in schizophrenia? Psychiatria Pol. 2018;52(5):819–834. doi: https://doi.org/10.12740/PP/OnlineFirst/76040
24. Gandara V, Pineda JA, Shu IW, Singh F. A systematic review of the potential use of neurofeedback in patients with schizophrenia. Schizophrenia Bulletin. 2020;1(1):sgaa005. doi: https://doi.org/10.1093/schbul/sbaa044
25. Popova NK, Naumenko VS. Neuronal and behavioural plasticity: The role of serotonin and BDNF systems tandem. Expert Opinion on Therapeutic Targets. 2019;23(3):227–239. doi: https://doi.org/10.1080/14728222.2019.1572747
26. Mennerick S, Zorumski C. Neural activity, and survival in the developing nervous system. Mol Neurobiol. 2000;22(1–3):41–54. doi: https://doi.org/10.1385/MN:22:1-3:041
27. Lefebvre S, Pavlidou A, Walther S. What is the potential of neurostumulation in the treatment of motor symptoms in schizophrenia? Expert Rev Neurotherapeutics. 2020;20(7):697–706. doi: https://doi.org/10.1080/14737175.2020.1775586
28. Markiewicz-Gospodarek A, Markiewicz R, Dobrowolska B, Maciejewski R, Łoza B. Relationship of neuropeptide S with clinical and metabolic parameters of patients during rehabilitation therapy for schizophrenia. Brain Sci. 2022;12(6):768. doi: https://doi.org/10.3390/brainsci12060768
29. Schulz KF, Altman DG, Moher D. CONSORT Group. CONSORT 2010 statement: Updated guidelines for reporting parallel group randomized trials. Ann Intern Med. 2010;152(11):726–32. doi: https://doi.org/10.7326/0003-4819-152-11-201006010-00232
30. Valle R. Schizophrenia in ICD-11: Comparsion of ICD-10 and DSM-5. Revista de Psiquiatria y Salud Mental (English Edition). 2020;13(2):95–104. doi: https://doi.org/10.1016/j.rpsmen.2020.01.002
31. Cohen J. Statistical power analysis for the behavioral sciences. 2nd ed. Hillsdale, New Jersey: Lawrence Erlabaum Associates, Publishers; 1988. ISBN 0-8058-083-5.
32. Qu M, Wang J, Chen DC, Chen S, Xiu MH, Zhang XY. Sex-specific association between peripheral superoxide dismutase, BDNF, and cognitive impairment in drug-naïve first episode patients with schizophrenia. Free Radical Biol Med. 2020;160:887–893. doi: https://doi.org/10.1016/j.freeradbiomed.2020.09.014
33. Yang F, Wang K, Du X, Deng H, Wu HE, et al. Sex difference in the association of body mass index and BDNF levels in Chinese patients with chronic schizophrenia. Psychopharmacol. 2019;236(2):753–762. doi: https://doi.org/10.1007/s00213-018-5107-1
34. Yang Y, Zhang Y, Wang J, Ning X, et al. Sex differences in the association of HOMA-IR index and BDNF in Han Chinese patients with chronic schizophrenia. Front Psychiatry. 2021;12:656230. doi: https://doi.org/10.3389/fpsyt.2021.656230
35. Ali FT, Abd El-Azeem EM, Hamed MA, Ali MAM, Abd Al-Kader NM, Hassan EA. Redox dysregulation, immuno-inflammatory alterations, and genetic variants of BDNF and MMP-9 in schizophrenia: Pathophysiological and phenotypic implications. Schizophr Res. 2017;188:98–109. doi: https://doi.org/10.1016/j.schres.2017.01.016
36. Collazos J, Asensi V, Martin G, Montes AH, Suárez-Zarracina T, Valle-Garay E. The effect of gender and genetic polymorphisms on matrix metalloprotease (MMP) and tissue inhibitor (TIMP) plasma levels in different infectious and non-infectious conditions. Clin Exp Immunol. 2015;182(2):213–219. doi: https://doi.org/10.1111/cei.12686
37. Wei SM, Berman KF. Ovarian hormones, genes, and the brain: the case of estradiol and the brain-derived neurotrophic factor (BDNF) gene. Neuropsychopharmacology. 2019;44:223–224. doi: https://doi.org/10.1038/s41386-018-022305
38. Jaillon S, Berthenet K, Garlanda C. Sexual dimorphism in innate immunity. Clin Rev Allergy Immunol. 2019;56:308–321. doi: https://doi.org/10.1007/s12016-017-8648-x
39. Takeuchi H, MacKenzie NE, Samaroo D, Agid O, Remington G, Leucht S. Antipsychotic dose in acute schizophrenia: A meta-analysis. Schizophrenia Bulletin. 2020;46(6):1439–1458. doi: https://doi.org/10.1093/schbul/sbaa063
40. Lim K, Peh OH, Yang Z, Rekhi G, Rapisarda A, See YM, et al. Large-scale evaluation of the Positive and Negative Syndrome Scale (PANSS) symptom architecture in schizophrenia. Asian J Psychiatry. 2021;62:102732. doi: https://doi.org/10.1016/j.ajp.2021.102732
41. Thompson M, Thompson L. The Neurofeedback Book, 2nd edition: An Introduction to Basic Concepts in Applied Psychophysiology. 2015, ISBN 10: 069292101X ISBN 13: 9780692921012, Association for Applied Psychophysiology and Biofeedback.
42. Wykes T, Reeder C, Williams C, Corner J, Rice C, Everitt B. Are the effects of cognitive remediation therapy (CRT) durable? Results from an exploratory trial in schizophrenia. Schizophrenia Res. 2003;61:163–174. doi: https://doi.org/10.1016/S0920-9964(02)00239-6
43. Markiewicz R, Dobrowolska B. Cognitive and social rehabilitation in schizophrenia – from neurophysiology to neuromodulation. Pilot Study. Int J Environ Res Public Health. 2020;17(11):4034. doi: https://doi.org/10.3390/ijerph17114034
44. Markiewicz R, Dobrowolska B. Reinforcement of self-regulated brain activity in schizophrenia patients undergoing rehabilitation. BioMed Res Int. 2021;Article ID 8030485, 1–9. doi: https://doi.org/10.1155/2021/8030485
45. Braithwaite J, Watson D, Jones R, Rowe M. A guide for analysing electrodermal activity (EDA) & and skin conductance responses (SCRs) for psychological experiments. Technical Report, 2nd version: Selective Attention & Awareness Laboratory (SAAL). Behavioural Brain Sciences Centre, University of Birmingham, UK, 2015.
46. Lu EY, Cheng ASK, Tsang HWH, Chen J, Leung S, Yip A, et al. Psychoeducation, motivational interviewing, cognitive remediation training, and/or social skills training in combination for psychosocial functioning of patients with schizophrenia spectrum disorders: A systematic review and meta-analysis of randomized controlled trials. Front Psychiatry. 2022;13:899840. doi: https://doi.org/10.3389/fpsyt.2022.899840
47. Markiewicz R, Markiewicz-Gospodarek A, Dobrowolska B, Łoza B. Improving clinical, cognitive, and psychosocial dysfunctions in patients with schizophrenia: A neurofeedback randomized control trial. Neural Plast. 2021;2021:4488664. doi: https://doi.org/10.1155/2021/4488664
48. Romash IR, Vynnyk MI. Peculiarities of brain-derived neurotrophic factor and matrix metalloproteinase-9 expression dynamics in patients with paranoid schizophrenia depending on the duration of the disease. Wiad Lek. 2021;74:2728–2732. PMID: 35023483
49. Arabska J, Margulska A, Strzelecki D, Wysokiński A. Does metabolic status affect serum levels of BDNF and MMP-9 in patients with schizophrenia? Nord J Psychiatry. 2019;73(8):515–521. doi: https://doi.org/10.1080/08039488.2019.1658126
50. Vandooren J, Van den Steen PE, Opdenakker G. Biochemistry and molecular biology of gelatinase B or matrix metalloproteinase-9 (MMP-9): The next decade. Critical Reviews in Biochemistry and Molecular Biology. 2013;48(3):222–272. doi: https://doi.org/10.3109/10409238.2013.770819
51. Adams RA, Bush D, Zheng F, Meyer SS, Kaplan R, et al. Impaired theta phase coupling underlies frontotemporal dysconnectivity in schizophrenia. Brain. 2020;143(4):1261–1277. doi: https://doi.org/10.1093/brain/awaa035
52. Donkers FC, Schwikert SR, Evans AM, Cleary KM, Perkins DO, Belger A. Impaired neural synchrony in the theta frequency range in adolescents at familial risk for schizophrenia. Front Psychiatry. 2011;2:51. doi: https://doi.org/10.3389/fpsyt.2011.00051
53. Lee HS, Kim JS. Implication of electrophysiological biomarkers in psychosis: Focusing on diagnosis and treatment response. J Pers Med. 2022;12(1):31. doi: https://doi.org/10.3390/jpm12010031
54. Atagun MI, Drukker M, Hall MH, Altun IK, Tatli SZ, Guloksuz S, van Os J, van Amelsvoort T. Meta-analysis of auditory P50 sensory gating in schizophrenia and bipolar disorder. Psych Res Neuroimaging. 2020;300:111078. doi: https://doi.org/10.1016/j.pscychresns.2020.11.111078
55. Javitt DC, Freedman R. Sensory processing dysfunction in the personal experience and neuronal machinery of schizophrenia. Am J Psychiatry. 2015;172(1):17–31. doi: https://doi.org/10.1176/appi.ajp.2014.13121691
56. Pachalska M, Góral-Pólrola J, Mueller A, Kropotov JD. Neuropsychology and the neurophysiology of perceptual microgenesis. Acta Neuropsychol. 2017;15(4):365–389. doi: 10.5604/01.3001.0010.7243
57. Kropotov JD. Functional neuromarkers for neuropsychology. Acta Neuropsychol. 2018;16(1):1–7. doi: 10.5604/01.3001.0011.6504
58. Loriette C, Ziane C, Ben Hamed S. Neurofeedback for cognitive enhancement and intervention and brain plasticity. Revue Neurologique. 2021;177(9):1133–1144.
59. Luckos M, Cielebak K, Kaminski P. EEG neurofeedback in the treatment of cognitive dysfunctions after the infection of SARS-CoV-2 and long COVID-19. Acta Neuropsychol. 2021;19(3):361–372. doi: 10.5604/01.3001.0015.2464
60. Pazooki K, Leibetseder M, Renner W, Gougleris G, Kapsali E. Neurofeedback Treatment of Negative Symptoms in Schizophrenia: Two Case Reports. Appl Psychophysiol Biofeedback. 2019;44(1):31–39. doi: 10.1007/s10484-018-9417-1