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Investigation of serum galanin, alarin, meteorin-like protein, and ischemia modified albumin levels in patients with schizophrenia

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Original Article

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Abstract

Objective: The objective of the current research is to study the serum levels of ischemia modified albumin (IMA), a new oxidative stress indicator, and various peptides (galanin, alarin, and meteorin-like protein) that may affect the pathophysiology of schizophrenia, determine their relationship with clinical features and each other, and compare them to those in healthy controls. Materials and Methods: This is a cross-sectional study consisting of 45 individuals with schizophrenia who are in remission and 45 healthy individuals. The differences and relationships between categorical variables and serum protein levels of the patient and control groups were statistically analysed. Receiver operating characteristics analysis was used for the diagnostic decision-making properties of serum protein levels to predict the presence of the disease. Results: In comparison with the control group, the median levels of serum proteins galanin and alarin were statistically lower in the patient group, whereas METRNL and IMA levels were higher. Considering the predictive values of serum proteins in the diagnosis of the disease, it was observed that serum galanin, alarin, and IMA levels had a sensitivity and specificity higher than 80%, followed by METRNL with 73.3% sensitivity and 66.7% specificity. Conclusion: Our findings reveal galanin, alarin, meteorin-like protein, and IMA are important molecules with high sensitivity and specificity in diagnosing schizophrenia. Furthermore, we think that further studies are needed to use them as reliable parameters in terms of clinical course, classification, and prognosis in explaining the etiopathogenesis of the disease.

Significant outcomes

- Investigations of molecules that may affect the pathophysiology of schizophrenia.
- The potential of the relevant molecules to be early biomarkers in the diagnosis of the disease.

Limitations

- The small sample size and the cross-sectional character of the study individuals' age, sex, smoking status, dietary habits, personal lifestyles, and metabolic factors may impact the results.
- The potential effects of drugs could not be excluded.

Introduction

Schizophrenia is probably the most severe among psychiatric disorders. It is one of the most disabling and economically devastating medical disorders. The World Health Organization listed it among the top 10 diseases that contribute to the global disease burden (Getinet Ayano, 2016).

Although several molecular-based hypotheses, involving neurodevelopmental and neurochemical hypotheses, have been suggested to elucidate the neuropathology of schizophrenia, the search for molecules in various neuropeptide structures still continues to clarify the pathophysiology of schizophrenia and contribute to diagnosing and treating the disease (Zamanpoor, 2020). Furthermore, the role of immunological and immunopathological mechanisms in the etiopathogenesis of schizophrenia has recently become a subject of research with increasing popularity (Rothermundt *et al.*, 2001).

Galanin represents a 29/30 amino acid-long neuropeptide that is widely expressed in the brains of numerous mammals (Freimann *et al.*, 2015). It was discovered by Tatemoto *et al.* (1983) by detecting the C-terminal amide structure in the porcine intestinal extract using a

chemical method (Tatemoto *et al.*, 1983). Since its discovery a long time ago, numerous physiological functions have been attributed to galanin, and active studies are ongoing to confirm these functions and reveal their significance for pathology and physiology (Lang *et al.*, 2007).

The galanin peptide family consists of the 'parent' galanin derived from the same peptide precursor gene product as galanin, the galanin message-associated peptide, the galanin-like peptide (GALP) that is encoded by various gene, and the newly discovered peptide alarin, encoded by a splice variant of the GALP gene. Hormones of the galanin family are bound to GalRs, such as GalR1, GalR2, and GalR3. The receptors in question are commonly distributed in the mammalian brain (Lang *et al.*, 2007). It has been considered that galanin may take a part in the pathophysiology of neuropsychiatric diseases, for example, schizophrenia and eating disorders, Parkinson's, and Alzheimer's diseases (Teixeira Mendes *et al.*, 2001; GÜL *et al.*, 2021; Gul *et al.*, 2021). Galanin functions as an endogenous 'painkiller' following nerve damage, encouraging survival and regeneration (Hökfelt and Wiesenfeld-Hallin, 2024).

The release of growth hormone, prolactin, and luteinizing hormone from the pituitary gland is stimulated by galanin. Peripherally, galanin causes the inhibition of insulin secretion from pancreatic β-cells and contracts or relaxes different gastrointestinal smooth muscles. Hence, it has therapeutic potential in various areas, including nociception, anxiety, depression, obesity, memory disorders, diabetes, cardiovascular diseases, digestive disorders, sexual dysfunction, and growth disorders (Wang *et al.*, 2005). In an animal model of depression, a study found that galanin (1-15) [GAL(1-15)] enhanced the antidepressant effects of FLX in naïve rats; this may indicate a new method of strengthening depression (Flores-Burgess *et al.*, 2022).

Alarin represents a 25-amino acid peptide belonging to the galanin peptide family. Alarin was detected for the first time in the gangliocytes of neuroblastic tumours and later demonstrated to play a vasoactive function in the skin. It has been recently shown that alarin stimulates the hypothalamic-pituitary-gonadal axis, in addition to food intake in rodents, which has suggested that it may be a neuromodulatory peptide in the brain (Eberhard *et al.*, 2010).

Meteorin-like protein represents a small (28 kDa) secreted protein similar to meteorin. M2 macrophages and barrier tissues (skin, mucosa) express METRNL. METRNL is overexpressed in rheumatoid arthritis lesions and is a cytokine that takes part in inflammation and innate immunity (Ushach *et al.*, 2015a). There are studies available in the literature that research METRNL levels in rheumatoid arthritis (Zhang *et al.*, 2022), obesity (Schmid *et al.*, 2021), and type 2 diabetic patients (Khajebishak *et al.*, 2022).

Albumin is an important determinant of the antioxidant capacity of human serum (Roche *et al.*, 2008). It is thought that structural alterations in the N-terminus of serum albumin, reducing cobalt- and nickel-binding capacity, are associated with the generation of reactive oxygen species (ROS) during ischemia (Bar-Or *et al.*, 2001). The said decrease in metal-binding capacity is measurable and known as ischemia modified albumin (IMA) (Can *et al.*, 2013). Recent *in vitro* research has stated that ROS, particularly hydroxyl radicals, can chemically change human serum albumin, leading to the formation of IMA (Gurumurthy *et al.*, 2014). IMA levels have been assessed in diseases, for example, autism spectrum disorder (Ceylan *et al.*, 2020a), acute cerebrovascular diseases (Elshony *et al.*, 2021), and hypertensive retinopathy(Pavlovschi *et al.*, 2021).

The objective of the present research is to examine the serum levels of IMA, a new oxidative stress indicator, and various peptides such as galanin, alarin, and METRNL, which may affect the pathophysiology of schizophrenia, to determine their relationship with clinical features and each other, and compare them with those in healthy controls. Although there is no study in the literature assessing galanin, alarin, METRNL, and IMA together in the same patient group, the research studying these parameters separately in patients with schizophrenia is also limited. Accordingly, it was aimed to contribute to the literature by researching the potential of the relevant molecules to be early biomarkers in the diagnosis of the disease.

Material and Methods

Patient groups

This study was conducted with individuals in remission who were treated with a diagnosis of schizophrenia and whose schizophrenia diagnosis was confirmed according to the DSM-5 criteria, who presented to Zeytinburnu Community Mental Health Center, one of the community mental health centres providing services under Bakırköy Prof. Dr. Mazhar Osman Mental Health and Neurological Diseases Training and Research Hospital. The study sample comprised 2 groups, 45 individuals with schizophrenia and 45 healthy individuals among hospital employees who volunteered to take part in the research, and the study was performed as cross-sectional. Ethics committee approval was received from Dr. Sadi Konuk Training and Research Hospital to start the study (date 19.04.2021 and decision number 2021-08-32). Signed consent was acquired from all individuals enrolled in the research by explaining to them the form and purpose of the study in detail. The research complies with the Helsinki Declaration.

Study inclusion criteria

Inclusion criteria of the patient group

1. Being diagnosed with schizophrenia
2. Being in remission (not being hospitalised in the last 6 months, no having drug or drug dose changes due to exacerbation of symptoms, and CGI <= 3 were considered being in remission)
3. Being aged between 18 and 65 years
4. Not having a psychiatric disorder in addition to the diagnosis of schizophrenia
5. Not having an alcohol and/or substance use disorder in the last 1 year
6. Not having additional chronic medical (hypertension, diabetes, chronic obstructive pulmonary disease, rheumatological disease, etc.) or neurological (epilepsy, multiple sclerosis, etc.) diagnoses
7. Not having fever, malaise, and similar signs or symptoms of infectious disease during sampling

Inclusion criteria of the control group

1. Being aged between 18 and 65 years
2. Not having a current or past psychiatric disorder
3. Not having an alcohol and/or substance use disorder in the last 1 year
4. Not having chronic medical (hypertension, diabetes, chronic obstructive pulmonary disease, rheumatological disease, etc.) or neurological (epilepsy, multiple sclerosis, etc.) diseases

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5. Not having any signs or symptoms of an infectious disease during sampling

Exclusion criteria for the control and patient groups

1. Using drugs that will affect the immune-endocrine system
2. Heavy exercise or fasting status for more than 24 h
3. Using antioxidant preparations

Tools used in the study

A sociodemographic and clinical data form prepared by the researchers, the Clinical Global Inventory (CGI), the Brief Psychiatric Rating Scale (BPRS), and the Positive and Negative Syndrome Scale (PANSS) were employed.

Biochemical analyses

In the research, 5 ml blood samples were voluntarily collected from the brachial vein of fasting individuals into biochemistry tubes between 09:00 and 10:00 in the morning for biochemical investigations (by receiving the informed consent form). The collected samples were centrifuged for a period of 10 min at 4000 r.p.m. at a temperature of + 4°C at Zeytinburnu Community Mental Health Center, and serum was obtained. The obtained serum samples were divided into Eppendorf tubes and stored at a temperature of –80°C until analysis was performed. The samples were sent to Kafkas University Faculty of Medicine, Department of Medical Biochemistry in a special vehicle under cold chain conditions. All ELISA analyses in the current research were conducted in the Medical Biochemistry R&D laboratory of Kafkas University on the same day, as soon as the samples arrived.

Measurement of serum peptide levels by ELISA analyses

GAL, GALP (Alarin), METRNL, and IMA levels were examined in serum samples with human ELISA kits (catalog no; E1332Hu, E4655Hu, E3941Hu, and E1172Hu; lot no; 202111018, 202111018, 202111018, and 202111018, respectively, Bioassay Technology Laboratory [BT-LAB]., Shanghai, China), following the kit procedures. The absorbances were read spectrophotometrically at a wavelength of 450 nm in the ELx800 ELISA reader (BioTek® Instruments, Inc., USA). Plates were washed using the BioTek ELX50 washer as an automatic washer. The samples were not diluted, and their concentrations were calculated according to standard curves. The results were reported as ng/L for GAL, ng/mL for GALP, ng/mL for METRNL, and ng/mL for IMA. The kits' measuring ranges were 0.5–100 ng/L, 0.05–10 ng/mL, 0.05–15 ng/mL, and 2–600 ng/mL, respectively. The kits' minimum measurable levels were 0.26 ng/L, 0.022 ng/mL, 0.023 ng/mL, and 1.08 ng/mL. The intra-assay coefficient variables (CV%) of all kits were < 8%, while their inter-assay CV% was < 10%.

Application

Ethics committee approval was received from Dr. Sadi Konuk Training and Research Hospital to start the study (date 19.04.2021 and decision number 2021-08-32). Signed consent was acquired from all individuals enrolled in the research by explaining to them the form and purpose of the study in detail.

Statistical analysis

The data obtained from the research were transferred to the computer environment and analysed using the SPSS (Statistical Package for Social Sciences) 22.0 package programme. In descriptive analyses, frequency data were given as number (n) and percentage (%), and numerical data were given using arithmetic mean ± standard deviation (sd) and median (1–3rd quartile). The chi-square (χ^2) test and Fisher's exact χ^2 test were carried out for the purpose of comparing categorical data. The compliance of numerical data with normal distribution was investigated by the Kolmogorov–Smirnov and Shapiro–Wilk tests. For non-normally distributed numerical variables, the Mann–Whitney *U*-test was utilised in the comparison of the two groups. The relationship between two non-normally distributed numerical variables was studied by Spearman's correlation analysis. Concerning the correlation relationships, $r = 0.05$–0.30 was considered a low correlation, $r = 0.30$–0.40 a low-moderate correlation, $r = 0.40$–0.60 a moderate correlation, $r = 0.60$–0.70 a good correlation, $r = 0.70$–0.75 a very good correlation, and $r = 0.75$–1.00 was considered an excellent correlation. The diagnostic decision-making properties of serum protein levels in predicting the presence of the disease were tested by receiver operating characteristics (ROC) curve analysis. The statistical significance level was considered to be $p < 0.05$.

Results

A total of 90 individuals, 45 patients with schizophrenia in remission and 45 healthy individuals, were enrolled in the present research carried out at Zeytinburnu Community Mental Health Center. Table 1 presents the distribution of sociodemographic characteristics and serum protein levels between the patient and control groups. The age of the patient group was statistically higher in comparison with the control group ($p < 0.001$). The education level of the control group was determined to be higher than that of the patient group ($p < 0.001$). The rates of being single and unemployed were identified to be statistically higher in the patient group than in the control group (p-values; $p = 0.06$, $p < 0.001$, respectively). Galanin and alarin levels, among the serum proteins, were lower in the patient group in comparison with the control group, whereas METRNL and IMA levels were higher ($p < 0.001$).

The distribution of the disease and treatment-related characteristics of the patient group enrolled in the research is listed in Table 2. Of the patients, 84.40% (n = 38) were using oral antipsychotics (AP), while 48.90% (n = 22) were using long-acting APs. The mean duration of the disease was 16.37 ± 8.57 years. The mean PANSS total score was 35.82 ± 4.75, and the mean BPRS score was 3.51 ± 2.72.

The relationship between chlorpromazine equivalent AP dose and patient and disease characteristics was studied in Table 3. There was a moderately significant negative correlation between the chlorpromazine equivalent AP dose and the disease onset age, and a moderately significant positive relationship was identified with the CGI score (r and p values; $r = -0.306$, $p = 0.041$; $r = 0.391$, $p = 0.008$, respectively). A positive and low-level significant correlation was identified between the chlorpromazine equivalent AP dose and PANSS negative symptoms score ($r = 0.296$, $p = 0.048$).

ROC analysis was carried out for the diagnostic decision-making properties of serum protein levels in predicting the presence of the disease. The ROC analysis showed that the serum galanin threshold value of 41.73 ng/L or less could predict

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Table 1. Distribution of sociodemographic characteristics and serum proteins in the patient and control groups

	Patient (n = 45)	Control (n = 45)	Test value	*p*
Age (years)	43.00 (35.00–50.00)	31.00 (27.00–39.00)	**4.630***	**< 0.001**
Sex				
Female	19 (42.20)	27 (60.00)	2.846**	0.092
Male	26 (57.80)	18 (40.00)		
Education level				
Secondary school and lower	33 (73.30)	5 (11.10)	**35.709****	**< 0.001**
High school and higher	12 (26.70)	40 (88.90)		
Marital status				
Married	13 (28.90)	26 (57.80)	**7.647****	**0.006**
Single	32 (71.10)	19 (42.20)		
Living alone				
No	44 (97.80)	38 (84.40)	4.939***	0.058
Yes	1 (2.20)	7 (15.60)		
Children				
Yes	16 (35.60)	18 (40.00)	0.189**	0.664
No	29 (64.40)	27 (60.00)		
Employment status				
Employed	13 (28.90)	44 (97.80)	**45.981****	**< 0.001**
Unemployed	32 (71.10)	1 (2.20)		
Working duration				
None****	14 (31.10)	0 (0.00)		
≤5 years	10 (22.20)	12 (26.70)	**16.848****	**< 0.001**
>5 years	21 (46.70)	33 (73.30)		
Smoking				
Yes	19 (42.20)	14 (31.10)	1.196**	0.274
No	26 (57.80)	31 (68.90)		
Family history of psychiatric disease				
Yes	2 (4.40)	1 (2.20)	0.345***	1.000
No	43 (95.60)	44 (97.80)		
Serum galanin (ng/L)	26.85 (21.02–34.86)	52.11 (42.22–68.78)	**6.363***	**< 0.001**
Serum alarin (ng/mL)	2.37 (2.23–2.78)	4.88 (3.47–6.92)	**7.993***	**< 0.001**
Serum METRNL (ng/mL)	6.01 (2.39–8.77)	2.22 (1.93–3.38)	**4.257***	**< 0.001**
Serum IMA (ng/mL)	273.03 (135.88–414.52)	93.48 (85.33–118.01)	**5.354***	**< 0.001**

METRNL, meteorin-like protein; IMA, ischemia modified albumin.
*: Mann–Whitney *U*-test.
**: Pearson's chi-square test.
***: Fisher's exact test.
****: The group from which the difference originated.

schizophrenia with 82.2% sensitivity and 80.0% specificity compared to healthy individuals. The area under the curve was determined to be 0.889 ($p < 0.001$, confidence interval: 0.822–0.957) (Fig. 1). The ROC analysis demonstrated that the serum alarin threshold value of 3.195 ng/mL or lower could predict schizophrenia with 86.7% sensitivity and 84.4% specificity compared to healthy individuals. The area under the curve was found to be 0.873 ($p < 0.001$, confidence interval: 0.796–0.949) (Fig. 2). According to the ROC analysis, the serum METRNL threshold value of 2.63 ng/mL or higher could predict schizophrenia with 73.3% sensitivity and 66.7% specificity compared to healthy individuals. The area under the curve was found to be 0.760 ($p < 0.001$, confidence interval: 0.659–0.862) (Fig. 3). In the ROC analysis, it was revealed that the serum IMA threshold value of 128.16 ng/mL or higher could predict schizophrenia with 80.0% sensitivity and 80.0% specificity compared to healthy individuals.

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Table 2. Distribution of disease and treatment-related characteristics in the patient group

	n	%
History of suicide attempt		
Yes	3	6.70
No	42	93.30
History of homicide attempt		
Yes	1	2.20
No	44	97.80
Use of oral APs		
Yes	38	84.40
No	7	15.60
Use of long-acting APs		
Yes	22	48.90
No	23	51.10
	Mean ± SD	**Median (1–3rdquartile)**
Disease onset age (years)	25.66 ± 8.39	25.00 (18.00–30.50)
Disease duration (years)	16.37 ± 8.57	15.00 (9.00–24.00)
Duration of regular AP use	67.57 ± 47.67	48.00 (30.00–108.00)
Number of hospitalizations	2.06 ± 2.25	1.00 (1.00–2.00)
Chlorpromazine equivalent AP dose	581.26 ± 449.77	400.00 (275.00–841.50)
CGI score	2.35 ± 0.60	2.00 (2.00–3.00)
PANSS positive symptoms	10.97 ± 3.24	10.00 (8.00–13.50)
PANSS negative symptoms	10.97 ± 3.24	10.00 (8.00–13.50)
PANSS general psychopathology	17.60 ± 2.08	17.00 (16.00–19.00)
PANSS total score	35.82 ± 4.75	34.00 (31.50–39.00)
BPRS score	3.51 ± 2.72	3.00 (2.00–5.00)

AP, antipsychotic; CGI, Clinical Global Inventory, PANSS; Positive and Negative Syndrome Scale; BPRS, Brief Psychiatric Rating Scale.

The area under the curve was determined to be 0.828 ($p < 0.001$, confidence interval: 0.737–0.919) (Fig. 4).

Discussion

In this study, the median galanin and alarin levels were determined to be statistically lower, and METRNL and IMA were revealed to be statistically higher in the patient group than in the control group. Our findings suggest that galanin, alarin, METRNL, and IMA have high sensitivity and specificity in the diagnosis of schizophrenia.

The role of galanin as a co-transmitter in rodent and human noradrenergic locus coeruleus neurones has been discussed (Hökfelt *et al.*, 2018). Galanin inhibits the secretion of insulin, acetylcholine, serotonin, and noradrenaline (Teixeira Mendes *et al.*, 2001). While galanin exerts its effects through galanin receptors 1–3 (GALR 1–3), it has been stated to control basic emotional, metabolic, and behavioural processes in various species, from zebrafish to humans (Mills *et al.*, 2020). It regulates many physiological actions in the nervous system of mammals, involving mood, cognition, nociception, neuroendocrine regulation, energy, and osmotic homeostasis (Freimann *et al.*, 2015). It inhibits acetylcholine (ACh) release in the hippocampus, which suggests a potential role in the modulation of memory and learning. Galanin receptors have become popular drug targets for the pharmaceutical and biotechnology industries (Wang *et al.*, 2005).

A postmortem study investigated galanin, corticotropin-releasing factor, arginine vasopressin (AVP), delta-sleep-inducing peptide, neuropeptide Y (NPY) and peptide YY (PYY) concentrations in the gray matter of the temporal cortex and hypothalamus of 14 patients with schizophrenia and 21 healthy controls, matched for age and autopsy delay, and detected that galanin, AVP, NPY, and PYY decreased considerably in the temporal cortex of patients with schizophrenia. Upon comparing patients with schizophrenia receiving and not receiving neuroleptic therapy, it was seen that the treatment factor was not able to explain decreased neuropeptide concentrations in the temporal lobe. A significant difference was not reported in hypothalamic neuropeptide concentrations when comparing healthy controls to patients with schizophrenia (Frederiksen *et al.*, 1991). In the research investigating the impacts of galanin on forebrain monoamine synthesis and spontaneous locomotor activity in rats, it was concluded that galanin modulated forebrain dopaminergic neurotransmission. The impact is observed to mediate at the somatodendritic level of the meso-neostriatal pathway and can be used for normalising disturbances that are probably attributed to dysfunction in the said neuronal pathway, including schizophrenia (Ericson and Ahlenius, 1999). It has been argued that galanin has

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Table 3. Relationship between chlorpromazine equivalent AP dose and patient and disease characteristics

	Chlorpromazine equivalent AP dose	
	r	p^*
Age (years)	−0.252	0.095
Disease onset age (years)	**−0.306**	**0.041**
Disease duration (years)	0.042	0.786
Number of hospitalizations	0.121	0.427
CGI score	**0.391**	**0.008**
PANSS positive symptoms	0.204	0.180
PANSS negative symptoms	**0.296**	**0.048**
PANSS general psychopathology	0.111	0.467
PANSS total score	0.291	0.052
BPRS score	0.239	0.114
Serum galanin	−0.287	0.056
Serum alarin	−0.018	0.908
Serum METRNL	0.064	0.674
Serum IMA	0.098	0.524

AP, antipsychotic; CGI, Clinical Global Inventory, PANSS; Positive and Negative Syndrome Scale; BPRS, Brief Psychiatric Rating Scale; METRNL, meteorin-like protein; IMA, ischemia modified albumin.
*: Spearman's correlation test.

an anti-inflammatory function in a number of cases and a proinflammatory function in others. Studies in the literature on galanin yield disputable results. The reason for this may be the complex nature of metabolic network signalling caused by interactions between galanin and galanin receptors (Oliveira Volpe *et al.*, 2020). On the contrary, it has been indicated that galanin can be considered an immunomodulatory peptide since it is capable of sensitising polymorphonuclear neutrophils and natural killer cells to proinflammatory cytokines (Kofler *et al.*, 2015).

It is argued that neuropeptides take part in the mechanism of action of ECT (electroconvulsive therapy). Considering that ECT has therapeutic effects in both schizophrenia and depression and lithium, in addition to antipsychotic drugs and psychotomimetics, selectively affects certain neuropeptides, it is assumed that different combinations of monoamine and neuropeptide changes in the chosen neuronal populations possibly form the basis for the impacts of ECT on particular disease symptoms, regardless of diagnosis (Mathé, 1999).

A study examining the impacts of single and repeated ECT, neurokinin A, substance P, vasoactive intestinal polypeptide, neurotensin, neuropeptide Y, and galanin on brain regional distribution in mice detected the highest neuropeptide Y and galanin concentrations in the hypothalamus and pituitary gland and revealed that peptide levels were fairly equally distributed in the frontal and occipital cortex, as well as in the striatum and hippocampus (Stenfors *et al.*, 1989).

As far as we know, there is no research in the literature assessing serum alarin levels in subjects with schizophrenia. One of the many possible biological functions of the pleiotropic peptide alarin is that it regulates behaviours that resemble depression. Alarin has been demonstrated to have antidepressant-like effects by animal-based cumulative data, albeit a more clarifying study is required. Alarin appears to be a viable antidepressant for use in the treatment of depression in the future, given its regulatory role in the disease (Abebe *et al.*, 2022).

In a study investigating the correlation between the positive antidepressant-like impacts of alarin and the hypothalamic pituitary adrenal axis activity and brain-derived neurotrophic factor (BDNF) levels in various brain regions in the chronic unpredictable mild stress (CUMS) paradigm, alarin ensured a dose-dependent decrease in the immobility time in the tail suspension test in mice. In the CUMS paradigm, the administration of alarin (1.0 nmol, intracerebroventricular) significantly enhanced murine behaviour, associated with decreased corticotropin-releasing hormone mRNA levels in the hypothalamus and a decrease in serum levels. Furthermore, alarin up-regulated BDNF mRNA levels in the prefrontal cortex and hippocampus (Wang *et al.*, 2014). Another study demonstrated that alarin might exert antidepressant-like impacts by targeting receptor-mediated signalling systems that might help identify the alarin receptor (Wang *et al.*, 2015).

An association has been found between circulating alarin levels with metabolic syndrome and insulin resistance (Fang *et al.*, 2018). In our study, the median galanin and alarin levels, among the serum proteins, were identified to be statistically lower (26.85 ng/L and 1.40 ng/mL, respectively) in the patient group than in the control group (52.11 ng/L and 4.88 ng/mL, respectively) ($p < 0.001$). Psychotomimetics, lithium, and antipsychotic drugs influence the brain synthesis, tissue concentrations, and release of particular neuropeptides (Mathé, 1999). These findings show the necessity of researching neuropeptides in terms of determining treatment targets. In a study investigating epileptiform activities in rat brain slices; it has been observed that alarin plays a role in regulating excitability in hypothalamic neurones and has a modulatory effect on synchronised neuronal discharges (Kalkan and Abidin, 2023).

Cytokines take part in numerous functions of the immune system, such as initiating, potentiating, and resolving immune responses. Meteorin-like/Meteorin-β represents a new immunoregulatory cytokine related to inflammation. METRNL is a small (approximately 28 kDa) secreted protein that is expressed by activated macrophages and barrier tissues (mucosa and skin). Various cytokines,for example, TNF-α, IL-12, IL-17a, and IL-4, induce METRNL production by bone marrow macrophages, whereas IFN-y and TGF-y inhibit it. Moreover, the generation of different cytokines and chemokines in macrophages is regulated by METRNL (Ushach *et al.*, 2018).

Clinical trials have provided reliable pieces of evidence that there is an association between METRNL and inflammation-related diseases, coronary heart disease, etc., and demonstrated that METRNL remains unclear in a number of diseases, for example, nervous system diseases (Miao *et al.*, 2020). METRNL is a new cytokine taking part in both innate and acquired immune responses (Ushach *et al.*, 2015). In our study, METRNL levels were identified to be higher in the sera of subjects with schizophrenia ($p < 0.001$). Pieces of evidence from first-episode psychosis and first-degree relatives of patients with schizophrenia support the idea that increased inflammation is associated with schizophrenia. The results of this research increase the possibility that further research on inflammation will ensure a better understanding of the aetiology and pathophysiology of schizophrenia (Kirkpatrick et al., n.d.).

Apart from the neuropeptides and inflammatory agents mentioned above, another issue that should be clarified in the pathophysiology of various psychiatric disorders is oxidative stress.

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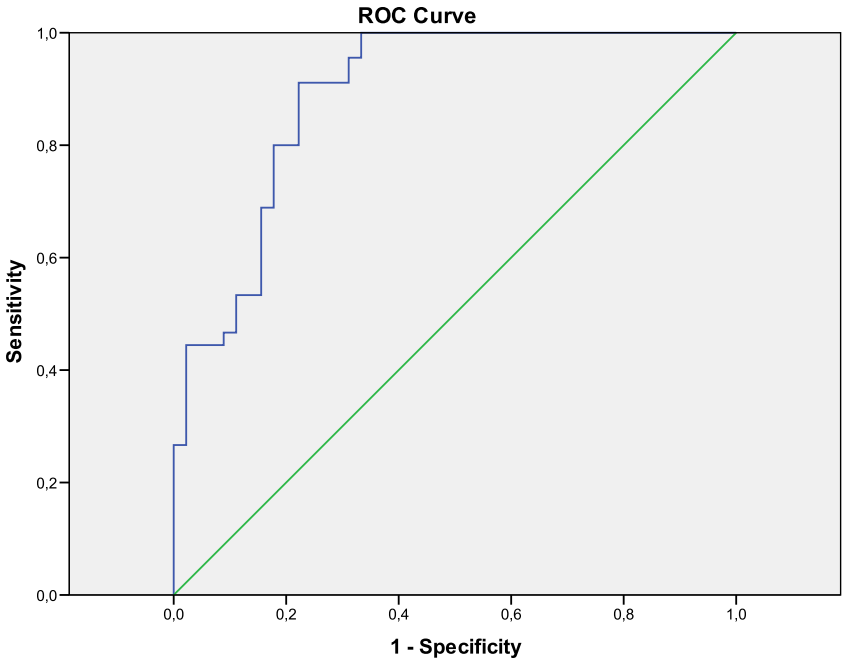

Figure 1. ROC analysis of serum galanin level.

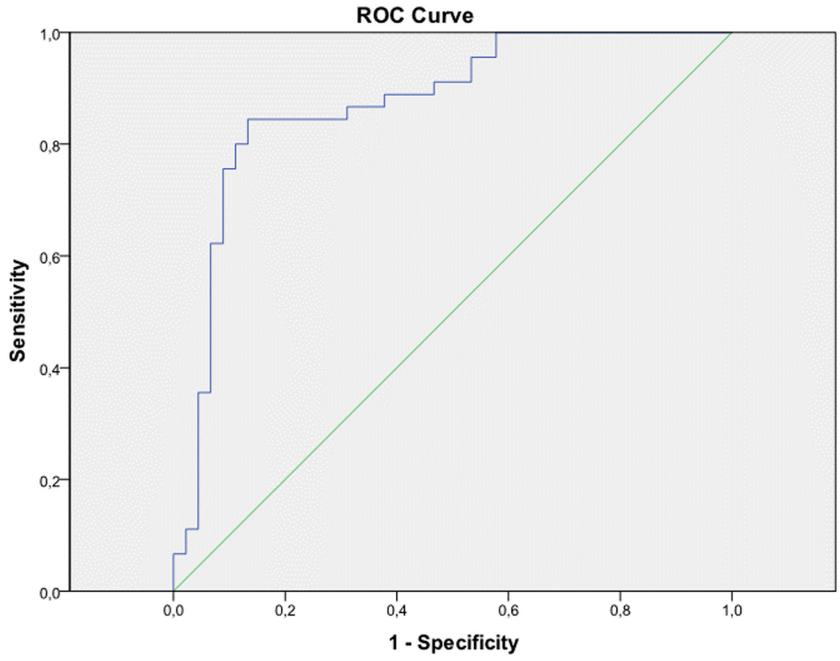

Figure 2. ROC analysis of serum alarin level.

Brain tissue is very sensitive to oxidative stress since it depends on oxygen a lot. The loss of selective cells in the brain is one of the ultimate common pathways that lead to progressive cognitive and behavioural impairment in schizophrenia. However, the factors causing selective cell loss in the brain are still unknown. There are pieces of evidence indicating reduced oxidative metabolism in certain brain regions in schizophrenia, and studies have associated these mechanisms with mitochondrial dysfunction. As a result of mitochondrial dysfunction, chain reactions are formed in which a number of ROS (free radicals) participate. Free radicals can damage all components of cells if their generation is higher than the capacity of antioxidant systems. Damage occurs in basic macromolecular structures, such as DNA, lipids, and proteins, as a result of these reactions (Yao *et al.*, ; Tunç *et al.*, 2018). Albumin, an acute phase reactant, is one of these molecules modified by free radicals. Albumin is a binding antioxidant with free radical scavenging properties. Moreover, the excess of free radicals can make a contribution to the psychopathology of psychiatric disorders and depressive symptoms (Soriani *et al.*, 1994). There are accumulated pieces of evidence indicating that antioxidant capacity is altered in schizophrenia. The membrane dysfunction may be secondary to free radical-mediated pathology and can make a contribution to certain aspects of schizophrenia symptomatology and complications of its treatment. Particularly, free radical-mediated abnormalities can make a contribution to developing clinically significant outcomes, such as

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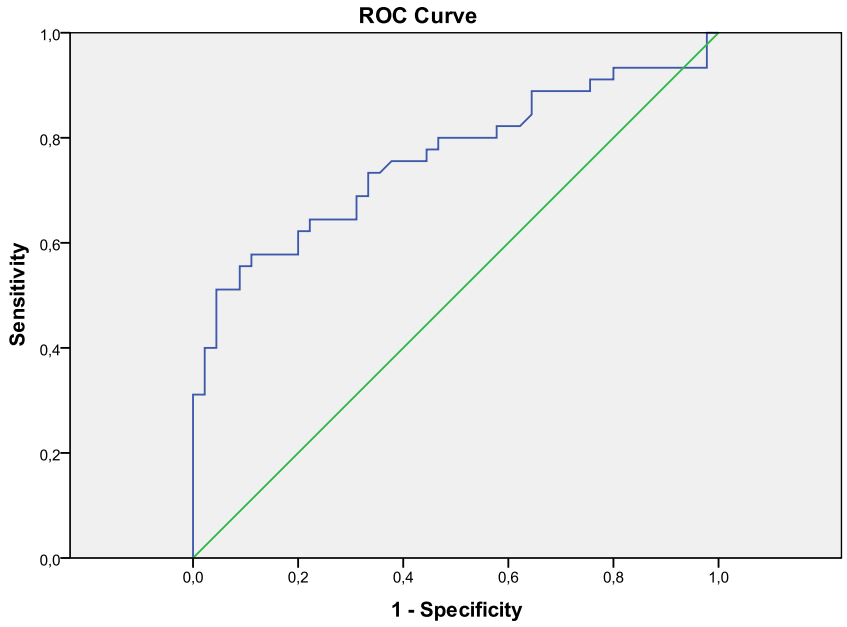

Figure 3. ROC analysis of serum METRNL level.

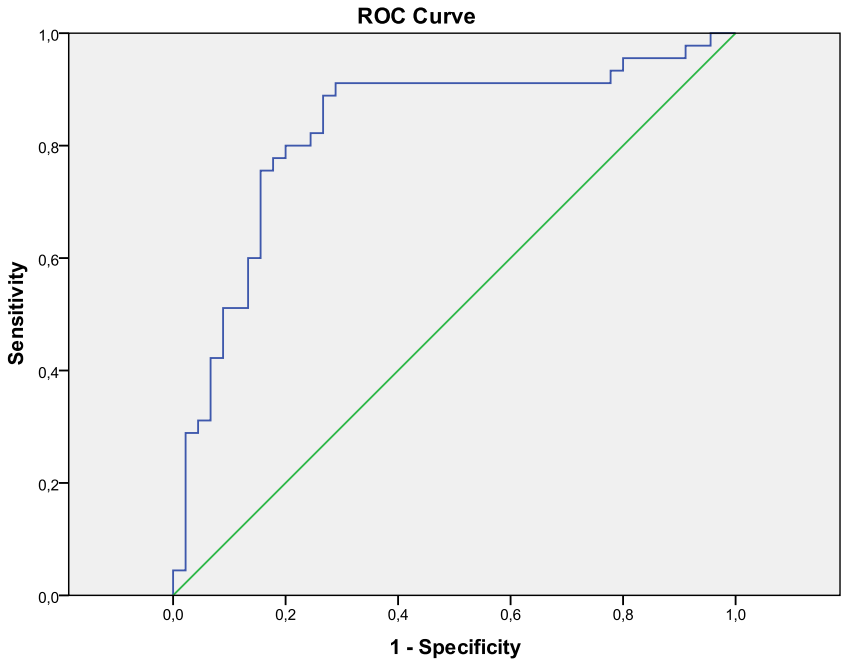

Figure 4. ROC analysis of serum IMA level.

tardive dyskinesia, prominent negative symptoms, and parkinsonian symptoms. Previous findings indicating the changed membrane dynamics and antioxidant enzyme activities in schizophrenia are in line with the concept of free radical-mediated neurotoxicity in schizophrenia. The above-mentioned results provide a theoretical basis for developing new therapeutic strategies in the future, including fatty acid and antioxidant supplementation (Yao *et al.*, 2012).

A study researching serum IMA levels in subjects with bipolar disorder in remission and healthy controls revealed no significant difference between age, sex, and IMA levels (Unal *et al.*, 2018).

In a study comparing myeloperoxidase activity, serum IMA levels, and peripheral blood mononuclear cell count in subjects with autism spectrum disorder, they were detected to be significantly higher in comparison with control subjects (Ceylan *et al.*, 2020a). In a study conducted to reveal whether changes in plasma IMA levels could assist with identifying the global metabolic risk in unipolar depression, bipolar disorder, and schizophrenia, it was found that although IMA levels were higher in all patient groups, statistical significance emerged only in the bipolar disorder group. Whereas increased IMA level was dependent on oxidative stress in the schizophrenia group, it was indicated to be due to immunity in the bipolar disorder group (Tunç *et al.*, 2018). In our research, METRNL and IMA levels in the schizophrenia group (6.01 ng/mL and 273.03 ng/mL, respectively) were identified to be higher compared to the control

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group (2.22 ng/mL and 93.48 ng/mL, respectively) ($p < 0.001$). These findings may argue that increased IMA levels may be related to the pathogenesis of neurodegenerative disorders such as autism and schizophrenia.

Recent studies have shown that IMA may be a potential biomarker for both haemorrhagic and ischemia stroke (Mohamed *et al.*, 2023). It has been stated that the interictal IMA levels of migraineurs are significantly higher than the control group and that IMA may be an indicator of oxidative stress in migraine patients and may lead to antioxidant agent studies in the prophylaxis of migraine (Say et al., 2020).

The small sample size and the cross-sectional character of the study were the most important limitations of this research. Individuals' age, sex, smoking status, dietary habits, personal lifestyles, and metabolic factors may impact the results. Patients in remission were enrolled in the research, but patients in the attack period were not included. The potential effects of drugs could not be excluded since patients were taking drugs and many were even receiving combination therapies.

As a result, galanin and alarin levels, among serum proteins, were detected to be lower in the patient group than in the control group, whereas METRNL and IMA levels were determined to be higher. To the best of our knowledge, the current research is the first study stating galanin and alarin and METRNL and IMA levels in subjects with schizophrenia and comparing them to healthy controls. Our findings demonstrate that galanin, alarin, METRNL, and IMA have high sensitivity and specificity in diagnosing schizophrenia and the potential of the relevant molecules to be early biomarkers in the diagnosis of the disease. The better the roles of the mentioned peptides in the pathogenesis of schizophrenia are explained, the more their potential roles in diagnosing and treating the disease can be utilised. Comprehensive clinical and experimental studies with larger sample sizes are required to reveal whether these biochemical parameters can be used as prognostic markers and whether they will be among the treatment targets.

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/neu.2024.20.

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Competing interests. All authors declare that they have no conflict of interest.

Ethical standards. Ethics committee approval was received from Dr. Sadi Konuk Training and Research Hospital to start the study (date 19.04.2021 and decision number 2021-08-32). Signed consent was acquired from all individuals enrolled in the research by explaining to them the form and purpose of the study in detail.

References

Abebe EC, Mengstie MA, Seid MA and Dejenie TA (2022) Regulatory effects and potential therapeutic implications of alarin in depression, and arguments on its receptor. *Frontiers in Psychiatry* **13**, 1051235. DOI: 10.3389/fpsyt.2022.1051235.

Ayano Getinet (2016) Schizophrenia: a concise overview of etiology, epidemiology diagnosis and management: review of literatures. *Journal of Schizophrenia Research* **3**(2), 2–7.

Bar-Or D, Curtis G, Rao N, Bampos N and Lau E (2001) Characterization of the Co $^{2+}$ and Ni $^{2+}$ binding amino-acid residues of the N-terminus of human albumin. *European Journal of Biochemistry* **268**(1), 42–48. DOI: 10.1046/j.1432-1327.2001.01846.x.

Can M, Varlibas F, Guven B, Akhan O and Yuksel GA (2013) Ischemia modified albumin and plasma oxidative stress markers in Alzheimer's disease. *European Neurology* **69**(6), 377–380. DOI: 10.1159/000339006.

Ceylan MF, Tural Hesapcioglu S, Yavas CP, Senat A and Erel O (2020) Serum ischemia-modified albumin levels, myeloperoxidase activity and peripheral blood mononuclear cells in autism spectrum disorder (ASD). *Journal of Autism and Developmental Disorders* **51**(7), 2511–2517. DOI: 10.1007/S10803-020-04740-9.

Eberhard N, Mayer C, Wagner A, Bauer HC, Sperk G, Boehm U and Kofler B (2010) Distribution of the neuropeptide alarin in murine brain. *Regulatory Peptides* **164**(1), 35–36. DOI: 10.1016/j.regpep.2010.07.087.

Elshony HS, Okda MA and El-Kabany RA (2021) Ischemia-modified albumin and fibulin-5 as diagnostic and prognostic markers for acute cerebrovascular disease. *Egyptian Journal of Neurology, Psychiatry and Neurosurgery* **57**(1), 1–8. DOI: 10.1186/s41983-020-00264-2.

Ericson E and Ahlenius S (1999) Suggestive evidence for inhibitory effects of galanin on mesolimbic dopaminergic neurotransmission. *Brain Research* **822**(1-2), 200–209. DOI: 10.1016/S0006-8993(99)01144-0.

Fang X, Zhang T, Yang M, Li L, Zhang C, Hu W, Fan X, Liu H, Zhu Z, Liu D, Zheng H, Gu HF, Liu R and Yang G (2018) High circulating alarin levels are associated with presence of metabolic syndrome. *Cellular Physiology and Biochemistry* **51**(5), 2041–2051. DOI: 10.1159/000495823.

Flores-Burgess A, Millón C, Gago B, Garciá-Durán L, Cantero-Garciá N, Puigcerver A, Narvaéz JA, Fuxe K, Santín L and Diáz-Cabiale Z (2022) Galanin (1-15) enhances the behavioral effects of fluoxetine in the olfactory bulbectomy rat, suggesting a new augmentation strategy in depression. *International Journal of Neuropsychopharmacology* **25**(4), 307–318. DOI: 10.1093/IJNP/PYAB089.

Frederiksen SO, Ekman R, Gottfries C-G, Widerlöv E and Jonsson S (1991) Reduced concentrations of galanin, arginine vasopressin, neuropeptide Y and peptide YY in the temporal cortex but not in the hypothalamus of brains from schizophrenics. *Acta Psychiatrica Scandinavica* **83**(4), 273–277. DOI: 10.1111/j.1600-0447.1991.tb05539.x.

Freimann K, Kurrikoff K and Langel Ü. (2015) Galanin receptors as a potential target for neurological disease. *Expert Opinion on Therapeutic Targets* **19**(12), 1665–1676. DOI: 10.1517/14728222.2015.1072513.

Gul HF, Yildirim C, Erdogan CE, Gul O and Koc N (2021) The role of galanin, alarin, irisin, PGC1-A and BDNF in the pathophysiology of Alzheimer's disease. *International Journal of Medical Science and Clinical Invention* **8**(06), 5498–5507. DOI: 10.18535/ijmsci/v8i06.09

Gül HF, Yıldırım C, Erdoğan C, Gül Ö. and Arslantürk İ. (2021) Investigation of plasma PGC1-α, irisin, BDNF, GAL, and GALP levels in Parkinson's disease. *Cumhuriyet Üniversitesi Sağlık Bilimleri Enstitüsü Dergisi* **6**(2), 145–161. DOI: 10.51754/cusbed.948015.

Gurumurthy P, Borra SK, Yeruva RKR, Victor D, Babu S and Cherian KM (2014) Estimation of ischemia modified albumin (IMA) levels in patients with acute coronary syndrome. *Indian Journal of Clinical Biochemistry* **29**(3), 367–371. DOI: 10.1007/s12291-013-0367-3.

Hökfelt T, Barde S, Xu ZQD, Kuteeva E, Rüegg J, le Maitre E, Risling M, Kehr J, Ihnatko R, Theodorsson E, Palkovits M, Deakin W, Bagdy G, Juhasz G, Prud'homme HJ, Mechawar N, Diaz-Heijtz R and Ögren SO (2018) Neuropeptide and small transmitter coexistence: fundamental studies and relevance to mental illness. *Frontiers in neural circuits* **12**, 106. DOI: 10.3389/fncir.2018.00106.

Hökfelt T and Wiesenfeld-Hallin Z (2024) The therapeutic potential of galanin in the management of pain – a review article. *Danish Medical Journal* **71**(2), 1–11. DOI: 10.61409/A10230653.

Kalkan ÖF, Öztürk H, Şahin Z, Başoğlu H, Aydin Abidin S and Abidin I (2023) Alarin potentiates ongoing epileptiform activity in rat brain slices: an in vitro electrophysiological study. *Acta Neurobiol Exp (Wars)* **84**(1), 35–42. DOI: 10.55782/ane-2024-2520.

Khajebishak Y, Faghfouri AH, Soleimani A, Madani S and Payahoo L (2022) Exploration of meteorin-like peptide (metrnl) predictors in type 2 diabetic

patients: the potential role of irisin, and other biochemical parameters. *Hormone Molecular Biology and Clinical Investigation* **0**(0), 127–135. DOI: 10.1515/HMBCI-2022-0037.

Kirkpatrick B and Miller BJ (2013) Inflammation and schizophrenia. *Schizophrenia Bulletin*. **39**(6), 1174–1179. DOI: 10.1093/schbul/sbt141.

Kofler B, Brunner S, Koller A, Wiesmayr S, Locker F, Lang R, Botz B, Kemény À and Helyes Z (2015) Contribution of the galanin system to inflammation. *SpringerPlus* **4**(1), 1–32. DOI: 10.1186/2193-1801-4-S1-L57/METRICS.

Lang R, Gundlach AL and Kofler B (2007) The galanin peptide family: Receptor pharmacology, pleiotropic biological actions, and implications in health and disease. *Pharmacology and Therapeutics. Pergamon* **115**(2), 177–207. DOI: 10.1016/j.pharmthera.2007.05.009.

Mathé AA (1999) Neuropeptides and electroconvulsive treatment. *The Journal of ECT* **15**(1), 60–75. DOI: 10.1097/00124509-199903000-00006.

Miao Z-W, Hu W-J, Li Z-Y and Miao C-Y (2020) Involvement of the secreted protein Metrnl in human diseases. *Acta Pharmacologica Sinica* **41**(12), 1525–1530. DOI: 10.1038/s41401-020-00529-9.

Mills EG, Izzi-Engbeaya C, Abbara A, Comninos AN and Dhillo WS (2020) Functions of galanin, spexin and kisspeptin in metabolism, mood and behaviour. *Nature Reviews Endocrinology* **17**(2), 97–113. DOI: 10.1038/s41574-020-00438-1.

Mohamed OS, Darwish MM, Mousa MM, Abd Elaziz AEH and Mohamed AK (2023) Impact of mode of anesthesia on ischemia modified albumin, operative conditions, and outcome in emergency craniotomies. *Egyptian Journal of Anaesthesia* **39**(1), 1–10. DOI: 10.1080/11101849.2022.2154011.

Oliveira Volpe CM, Vaz T, Rocha-Silva F, Villar-Delfino PH and Nogueira-Machado JA (2020) Is galanin a promising therapeutic resource for neural and nonneural diseases? *Current Drug Targets* **21**(9), 922–929. DOI: 10.2174/1389450121666200225112055.

Pavlovschi E, Pantea V, Borovic D and Tagadiuc O (2021) Study of ischemia modified albumin (IMA) as a biomarker in hypertensive retinopathy. *Medicine and Pharmacy Reports* **94**(2), 185–190. DOI: 10.15386/MPR-1815.

Roche M, Rondeau P, Singh NR, Tarnus E and Bourdon E (2008) The antioxidant properties of serum albumin. *FEBS Letters* **582**(13), 1783–1787. DOI: 10.1016/j.febslet.2008.04.057. No longer published by Elsevier.

Rothermundt M, Arolt V and Bayer TA (2001) Review of immunological and immunopathological findings in schizophrenia. *Brain, Behavior, and Immunity* **15**(4), 319–339. DOI: 10.1006/brbi.2001.0648.

SAY B, Dindar Badem N and Ergun U (2020) Serum levels of ischemic modified albumin (IMA) and prolidase in migraine subjects. *Journal of Health Sciences and Medicine* **3**(1). DOI: 10.32322/jhsm.638466.

Schmid A, Karrasch T and Schäffler A (2021) Meteorin-like protein (Metrnl) in obesity, during weight loss and in adipocyte differentiation. *Journal of Clinical Medicine* **10**(19), 4338. DOI: 10.3390/JCM10194338.

Soriani M, Pietraforte D and Minetti M (1994) Antioxidant potential of anaerobic human plasma: role of serum albumin and thiols as scavengers of carbon radicals. *Archives of Biochemistry and Biophysics* **312**(1), 180–188. DOI: 10.1006/abbi.1994.1297.

Stenfors C, Theodorsson E and Mathé AA (1989) Effect of repeated electroconvulsive treatment on regional concentrations of tachykinins, neurotensin, vasoactive intestinal polypeptide, neuropeptide Y, and galanin in rat brain. *Journal of Neuroscience Research* **24**(3), 445–450. DOI: 10.1002/JNR.490240315.

Tatemoto K, Rökaeus Å., Jörnvall H, McDonald TJ and Mutt V (1983) Galanin - a novel biologically active peptide from porcine intestine. *FEBS Letters* **164**(1), 124–128. DOI: 10.1016/0014-5793(83)80033-7.

Teixeira Mendes C, Forlenza OV and Gattaz WF (2001) Galanin and alzheimer's disease. *Revista de psiquiatria clinica*. Sao Paulo: Univ Sao Paulo, pp. 248–252.

Tunç S, Atagün Mİ., Neşelioğlu S, Bilgin YY, Başbuğ HS and Erel Ö. (2018) Ischemia-modified albumin: a unique marker of global metabolic risk in schizophrenia and mood disorders. *Psychiatry and Clinical Psychopharmacology*, **29**(2), 123–129. DOI: 10.1080/24750573.2018.1517466.

Unal K, Topcuoglu C and Yirun MC (2018) Serum ischemia modified albumin (IMA) levels of patients with bipolar disorder in remission and healthy controls. *Biomedical Research* **29**(12), 2672–2675. DOI: 10.4066/biomedicalresearch.29-18-337.

Ushach I, Arrevillaga-Boni G, Heller GN, Pone E, Hernandez-Ruiz M, Catalan-Dibene J, Hevezi P and Zlotnik A (2018) Meteorin-like/Meteorin-β is a novel immunoregulatory cytokine associated with inflammation. *The Journal of Immunology* **201**(12), 3669–3676. DOI: 10.4049/jimmunol.1800435.

Ushach I, Burkhardt AM, Martinez C, Hevezi PA, Gerber PA, Buhren BA, Schrumpf H, Valle-Rios R, Vazquez MI, Homey B and Zlotnik A (2015a) METEORIN-LIKE is a cytokine associated with barrier tissues and alternatively activated macrophages. *Clinical Immunology* **156**(2), 119–127. DOI: 10.1016/j.clim.2014.11.006.

Wang M, Chen Q, Li M, Zhou W, Ma T, Wang Y and Gu S (2014) Alarin-induced antidepressant-like effects and their relationship with hypothalamus-pituitary–adrenal axis activity and brain derived neurotrophic factor levels in mice. *Peptides* **56**, 163–172. DOI: 10.1016/J.PEPTIDES.2014.04.009.

Wang M, Zhou W, Zhou X, Zhuang F, Chen Q, Li M, Ma T and Gu S (2015) Antidepressant-like effects of alarin produced by activation of trkB receptor signaling pathways in chronic stress mice. *Behavioural Brain Research* **280**, 128–140. DOI: 10.1016/J.BBR.2014.11.039.

Wang S, Hwa J and Varty G (2005) Galanin receptors and their therapeutic potential. **5**(4), 415–440. DOI: 10.1517/14728214.5.4.415.

Yao JK, Reddy RD and van Kammen DP (2012) Oxidative damage and schizophrenia. *CNS Drugs* **15**(4), 287–310. DOI: 10.2165/00023210-200115040-00004.

Yao JK, Reddy RD and van Kammen DP(2001) Oxidative damage and schizophrenia: An overview of the evidence and its therapeutic implications. *CNS Drugs* **15**(4), 287–310. DOI: 10.2165/00023210-200115040-00004.

Zamanpoor M (2020) Schizophrenia in a genomic era: A review from the pathogenesis, genetic and environmental etiology to diagnosis and treatment insights. *Psychiatric Genetics. Lippincott Williams and Wilkins* **30**(1), 1–9. DOI: 10.1097/YPG.0000000000000245.

Zhang S, Lei Y, Sun T, Gao Z, Li Z and Shen H (2022) Elevated levels of metrnl in rheumatoid arthritis: Association with disease activity. *Cytokine* **159**, 156026. DOI: 10.1016/J.CYTO.2022.156026.

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