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CLINICAL NEUROCHEMISTRY

The Relationship of Inflammation with Energetic Metabolism and Mitochondrial Dysfunction in Autistic Spectrum Disorders

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Abstract—Using a set of inflammatory markers, we identified a cluster of patients with autism spectrum disorders (ASD) and high levels of inflammation (n = 35, 70%). Discriminant analysis showed that quinolic acid, the catabolite of the kynurenine pathway, makes the greatest contribution to the pathogenesis of ASD. Dysregulation of energy metabolism has been revealed, including both the main pathways of catabolism (glycolysis, the Krebs cycle) and alternative pathways (beta-oxidation, ketogenesis, branched amino acid metabolism) with impaired mitochondrial function (meglutol) in patients with ASD. Correlations have been revealed for a number of energy exchange metabolites with indices of inflammation. Inflammation in ASD is associated with dysregulation of energy metabolism and mitochondrial dysfunction. These data can serve as the basis for new treatment protocols, including anti-inflammatory therapy, metabolic correction, and restoration of mitochondrial function.

Keywords: Autism Spectrum Disorders (ASD), neuroinflammation, energy metabolism, mitochondrial dysfunction, metabolic markers

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INTRODUCTION

Autism spectrum disorders (ASD) are a clinically heterogeneous group of neuropsychiatric developmental disorders characterized by qualitative deviations in social interaction and communication skills, as well as a limited, stereotypical, repetitive set of interests and activities (ICD 10: F84 General disorders of psychological (mental) development) [1]. The incidence of ASD is about 1% of the child population (according to WHO data as of November 2022) and is characterized by a progressive increase in recent decades [2]. The pathogenetic mechanisms of ASD have not been fully studied, however, modern scientific data indicate the key role of neuroinflammation in the pathogenesis of these disorders [3–8].

The main immunocompetent cells in the brain responsible for the development of neuroinflammation are microglial cells. Neuroinflammation is associated with systemic inflammation and increased activity of blood immune cells, which is reflected in changes in systemic inflammatory markers of both innate and acquired immunity. In ASD, there is an increase in the production of proinflammatory cytokines such as tumor necrosis factor alpha (a-TNF), B-cell activating factor, interleukins 1β, 4, 5, 6, 8, 12p40 interferon-γ, etc. [9, 11], as well as an increase in other inflammation markers, in particular, the activity of leukocyte elastase (LE) and alpha-1 proteinase inhibitor (a-1PI) and the content of autoimmune antibodies to neuroantigens in the blood [10].

The process of inflammation/neuroinflammation is associated with impaired neurotransmission, neurodegeneration, mitochondrial dysfunction, dysmetabolic changes in energy metabolism, etc., which determines the behavioral and psychopathological features in ASD, and can also serve as a basis for new diagnostic markers and targets for therapeutic intervention.

The study of the relationship between inflammation and energy metabolism, as a key factor ensuring the functioning of the immune and other body systems, is of considerable interest. This relationship has been studied in various diseases (obesity, cancer, diabetes) [12], but in ASD this issue remains unstudied.

An example of metabolic changes during inflammation is cancer cachexia. Tumors produce proinflammatory cytokines (a-TNF, interleukin-6, etc.); proinflammatory status causes increased gluconeogenesis from lactic acid, associated with increased synthesis of a factor that enhances proteolysis, including muscle tissue, and a lipid-mobilizing factor that enhances beta-oxidation of fatty acids and lipolysis. In obesity and type II diabetes mellitus, adipocytes undergo metabolic changes with an abnormal increase in anabolism processes and fat accumulation. This

process is associated with increased synthesis of adipokines (a-TNF, interleukin-6), which directly affect regional immunocytes, causing infiltration into adipose tissue [13].

During inflammation, changes in energy metabolism also occur directly in immunocytes. Thus, microglial cells begin to receive energy predominantly from either anaerobic glycolysis or oxidative phosphorylation, whereas normally these processes are balanced [14]. Another example is the T-regulatory cells of visceral adipose tissue: in the activated state, these cells obtain energy from the process of fatty acid oxidation [15].

The most studied are changes in neutrophil metabolism in inflammatory somatic diseases. The activated neutrophil obtains energy mainly as a result of anaerobic glycolysis, since the foci of inflammation have a hypoxic environment which is unfavorable for mitochondrial function, while it turned out that neutrophils, like hepatocytes, are able to accumulate glycogen and carry out gluconeogenesis. In inflammatory lung diseases such as chronic obstructive pulmonary disease, the ability of neutrophils to maintain energy stores is disrupted due to impaired glycogen synthesis. Diabetes mellitus paradoxically determines a decrease in the rate of glycolysis and the function of the pentose phosphate cycle, while neutrophils begin to accumulate carbohydrates through the sorbitol pathway [16, 17].

The aim of this work was to study the relationship between inflammation, energy metabolism and mitochondrial function disorders in ASD, as well as to assess the correlation between the indices characterizing these processes.

MATERIALS AND METHODS

Laboratory data from 50 pediatric patients (1 to 12 years old, 40 boys and 10 girls) with an established diagnosis of ASD (severe degree according to CARS). The data were obtained from the clinical and laboratory data base of the "PlanetaMed" Center for Complex Diagnostics. The diagnosis "autism spectrum disorder" (F84) established by the psychiatrist met the criteria for this disorder developed in domestic child psychiatry [18]. The corresponding data of 60 conditionally healthy, normotypic children of the corresponding age and gender were used as a control group.

In blood plasma, we measured the parameters included in the neuroimmunotest which characterize the state of both the cellular and humoral components of immunity: the activity of leukocyte elastase (LE) and alpha-1 proteinase inhibitor (a1PI) and the level of autoimmune antibodies [10].

Enzymatic activity of LE was determined by an enzymatic spectrophotometric method using a specific substrate *N*-tert-butoxy-carbonyl-alanine-β-nitrophenyl ester (BOC-Ala-ONp), and were estimated in nmol/min mL (the sensitivity of the method was 40 nmol/min mL) [19].

Functional activity of α1-PI was determined by a spectrophotometric method and estimated in IU/mL (inhibitory units/mL) (the sensitivity of the method was 5 IU/mL) [20].

Level of autoantibodies (AAT) to neuroantigens S100b and myelin basic protein (MBP) in blood serum was determined by the standard solid-phase enzyme-linked immunosorbent assay (ELISA) method and estimated in optical density units (ODU).

The level of neuron-specific enolase (NSE) and S100b protein [21]; the level of kynurenine pathway metabolites: kynurenic (KYNA), xanthurenic (XAN), quinoline (QUIN), and picolinic (PIC) acids were used as other inflammatory markers reflecting damage to nervous tissue, neuronal death and changes in BBB permeability. The concentration of neurospecific proteins was determined in blood plasma using the ELISA method. Metabolites of the kynurenine pathway were determined in urine by gas chromatography with a mass spectrometric detector in the Chromolab laboratory.

The state of energy metabolism and mitochondrial function were assessed in urine based on the following substrates: Krebs cycle (citric, *cis*-aconitic, isocitric, 2-ketoglutaric, succinic, fumaric, malic acids), glycolysis (lactic and pyruvic acids), beta-oxidation (glutaric, adipic, sebacic, suberic, ethylmalonic acids), ketogenesis (acetoacetic, 3-hydroxybutyric and malonic acids), branched-chain amino acids (BCAA) (2-ketoisovaleric, 3-methyl-2-oxovaleric, 4-methyl-2-oxovaleric, 3-hydroxyisovaleric, 2-hydroxyisovaleric, 3-methylglutaric acids, isovalerylglycine, 3-methylcrotonylglycine), meglutol.

Energy metabolism is a catabolic part of metabolism, therefore urine, which contains products of catabolism, is the optimal biomaterial for the analysis of these metabolites. They were measured by gas chromatography with a mass spectrometric detector in the Chromolab laboratory.

Statistical analysis of neuroimmunotest parameters and kynurenine pathway metabolites was performed using control group data. The remaining indices were assessed using reference intervals provided by laboratories. Laboratory data were processed using STATISTICA 10.0 and MS Excel software.

RESULTS AND DISCUSSION

Table 1 presents data on the inflammatory status indices of patients in the study and control groups.

The data presented in the Table 1 show a significant increase in the activity of LE and a-1PI, as well as MBP AAB and S100b AAB in the group of patients compared to the control group. Statistical analysis of kynurenine pathway metabolites showed significant differences from the control group for KYNA and

Table 1. Inflammatory indices in patients with ASD and controls

Inflammation index	Control group, Me (Q1–Q3)	Study group, Me (Q1–Q3)	Reference limits
LE, nmol/min mL	194.4 (172.8–231)	**259.75 (231.1–279.7)***	150–200
a1PI, IU/mL	32.2 (30–38.2)	**44.25 (34–49.3)***	25–35
S100b AAB, OD units	0.565 (0.5–0.77)	0.815 (0.7–0.96)*	0.6–0.9
MBP AAB, OD units	0.625 (0.5–0.73)	0.79 (0.69–0.95)*	0.6–0.9
KYNA, mmol/mol creatinine	2.63 (1.7–3.1)	1.316 (0.4–2.12)*	1.032–4.487
XAN, mmol/mol creatinine	0.847 (0.496–1.231)	0.707 (0.375–1.407)	0.2082–2.2592
QUIN, mmol/mol creatinine	2.535 (1.957–3.016)	3.881 (2.278–5.564)*	1.4805–5.056
PIC, mmol/mol creatinine	1.372 (0.855–1.587)	1.368 (0.71–2.417)	0.390–2.543

*, Mann–Whitney test, $p < 0.05$ compared to the control group. Values outside the reference limits are shown in bold.

Table 2. Results of the analysis of discriminant functions of neuroimmunotest indicators

Inflammation indicators	Wilks lambda	Private lambda	F-excl. (1.75)	*p*-value	Toler	1-toler(R^2)
LE, nmol/min mL	0.528	0.904	7.297	0.009	0.584	0.416
S100b AAB, OD units	0.615	0.776	19.889	0.001	0.873	0.127

Note. Statistically significant coefficients are presented

QUIN. Thus, in the group of patients, the KYNA value decreased in comparison with the control group. Whereas the QUIN concentration in the patient group exceeded the values in the control group. It is known that catabolites of the kynurenine pathway have different functional activities: QUIN and XAN are neurotoxic, while KYNA and PIC exhibit neuroprotective properties. QUIN is a glutamate receptor (NMDA) agonist, which leads to their hyperactivation and reciprocally to a decrease in dopamine neurotransmission, which is associated with behavioral changes in patients with mental illness. The protective properties of KYNA are associated with the antagonistic effect on NMDA receptors and its increase regulates glutamate receptor conductivity. In addition, kynurenic acid is an antagonist of alpha-7-nicotinic acetylcholine receptors (a7nAch-R) and a regulator of the expression of other types of acetylcholine receptors—non-a7nAch-R [22, 23]. The obtained data indicate that the catabolism of tryptophan, shifted to the kynurenine pathway due to inflammation in patients with ASD, occurs predominantly before the formation of QUIN, which contributes to an increase in neurotoxic potential.

In general, the data characterizing the inflammatory status of patients indicate the presence of an inflammatory/neuroinflammatory process with an increase in neurotoxic metabolites of the kynurenic pathway in studied children and are consistent with the data of other authors [24–26].

At the same time, there is a wide range in the studied indices, which served as the basis for clustering patients. A preliminary discriminant analysis of systemic inflammation indices (Neuroimmunotest) was conducted in order to identify indices making the greatest contribution to the pathogenesis of ASD, for their further use as clustered data sets. The results of the discriminant analysis are presented in Table 2.

The results of discriminant analysis indicate that the greatest statistically significant contribution to the pathogenesis of ASD is made by the LE activity and the level of S100b AAB. Using discriminatory indices, a cluster of patients with a high level of inflammation was identified, the boundary of which was taken as the median values of LE activity and/or the level of S100b AAB in the patients. The cluster with a high level of inflammation consisted of 35 (70%) subjects.

Further study of the relationship between inflammatory indices and energy metabolism metabolites was performed using a selected cluster of patients with high levels of inflammation.

Statistical analysis of inflammatory status and energy metabolism parameters are presented in Table 3.

The data presented in Table 3 indicate that the control group and the group of patients were significantly different in the activity of LE and a-1PI with an excess of the upper reference limit in 30 (93%) and 19 (60%) children, respectively, as well as the level of S100b AAB (increased in 9 (28%) subjects) and MBP AAB (increased in 7 (22%) subjects). An excess of reference limits in the isolated clusters of patients was also noted for the concentration of NSE in the blood—24 (75%) children, while an increase in the level of the S100b protein was detected in 15 (47%) children.

Table 3. Comparison of inflammation indices in the selected group and the control group, as well as energy metabolism metabolites

Inflammation indicators	Control group, Me (Q1–Q3), $N = 30$	Cluster of patients, Me (Q1–Q3), $N = 35$	Reference limits
LE, nmol/min*mL	194.4 (172.8–231)	**272.2 (260.3–289.4)***	150–200
a1PI, IU/mL	32.2 (30–38.2)	**38.55 (32.35–48.85)***	25–35
S100b AAB, OD units	0.565 (0.5–0.77)	0.77 (0.67–0.93)*	0.6–0.9
MBP AAB, OD units	0.625 (0.5–0.73)	0.77 (0.67–0.86)*	0.6–0.9
KYNA, mmol/mol creatinine	2.63 (1.7–3.1)	**1.005 (0.394–2.277)***	1.032–4.487
XAN, mmol/mol creatinine	0.847 (0.496–1.231)	0.6059 (0.265–1.407)	0.2082–2.2592
QUIN, mmol/mol creatinine	2.535 (1.957–3.016)	4.084 (1.897–6.253)*	1.4805–5.056
PIC, mmol/mol creatinine	1.372 (0.855–1.587)	1.250 (0.631–2.405)	0.390–2.543
Neurospecific proteins	Cluster of patients, Me (Q1–Q3)	Reference limits	
Protein S100b, μg/L	0.101 (0.084–0.120)	<0.105	
NSE for, μg/L	**20.655 (16.31–27.38)**	<16.3	
Energy metabolism indicators, mmol/mol creatinine	Cluster of patients, Me (Q1–Q3)	Reference limits	
Glycolysis indices			
Lactic acid	20.835 (12.134–37.12)	7.428–35.605	
Pyruvic acid	28.349 (21.509–52.03)	4.764–45.100	
Krebs cycle indices			
Citric acid	100.251 (41.129–189.468)	33.445–305.495	
Cis-aconitic acid	33.938 (17.630–54.133)	16.250–65.820	
Isocitric acid	31.031 (19.461–58.035)	22.214–82.300	
2-ketoglutaric acid	2.174 (0.877–4.159)	1.260–9.432	
Succinic acid	9.638 (2.730–18.170)	2.807–26.470	
Fumaric acid	**1.612 (0.590–4.645)**	0.178–1.595	
Malic acid	0.708 (0.375–1.739)	0.209–2.710	
Ketogenesis indices			
Acetoacetic acid	0.022 (0.010–0.041)	0.0026–0.0915	
3-hydroxybutyric acid	15.877 (8.423–84.407)	0.707–31.518	
Malonic acid	0.684 (0.217–1.600)	0.297–1.832	
Beta-oxidation indices			
Glutaric acid	0.407 (0.272–1.041)	0.227–1.707	
Sebacic acid	0.202 (0.074–0.411)	0.018–0.359	
Adipic acid	3.166 (1.652–6.616)	1.176–7.747	
Suberic acid	1.431 (0.875–2.318)	0.794–4.154	
Ethylmalonic acid	3.830 (1.582–7.039)	2.290–20.290	
Coenzyme Q10 Metabolism Index			
Meglutol	9.318 (3.425–20.460)	5.216–15.919	
BCAA Metabolism Rates			
2-ketoisovaleric acid	0.978 (0.486–2.286)	0.292–1.451	
3-methyl-2-oxovaleric acid	1.504 (0.780–2.307)	0.523–3.843	
4-methyl-2-oxovaleric acid	1.362 (1.048–2.369)	0.243–1.971	
3-hydroxyisovaleric acid	9.728 (5.459–15.728)	4.047–20.001	

Table 3. (Contd.)

Inflammation indicators	Control group, Me (Q1–Q3), $N = 30$	Cluster of patients, Me (Q1–Q3), $N = 35$	Reference limits
2-hydroxyisovaleric acid	0.198 (0.132–0.377)	0.071–0.730	
3-methylcrotonylglycine	0.337 (0.097–0.822)	0.410–5.629	
3-methylglutaric acid	0.244 (0.144–0.454)	0.500–2.680	
Isovalerylglycine	0.835 (0.391–2.184)	0.163–3.877	

*, Mann–Whitney test, $p < 0.05$. Values outside the reference limits are shown in bold.

Among the metabolites of the kynurenine pathway, reliable differences in the isolated cluster of patients were found for KYNA and QUIN in urine, however, the median value of KYNA was outside the reference interval: a decrease in concentration was characteristic of 18 (50%) children. The obtained data indicate a reduced neuroprotective potential in this group of patients.

The next step of statistical evaluation was to identify the kynurenine pathway metabolite/s that contribute most to the pathogenesis of ASD. To identify this metabolite(s), we performed discriminant analysis. We found that QUIN makes a statistically significant contribution ($p < 0.05$): Wilks' coefficients lambda = 0.848, partial lambda = 0.665, F-exclusion (1.75) = 24.192. Urine QUIN concentrations exceeded the reference interval in 18 (50%) children. The observed result of elevated quinolinic acid levels in children with ASD is consistent with scientific literature data [27, 28].

Table 4 shows the results of the analysis of the content of energy metabolites in the selected cluster of patients with high levels of inflammation. Due to the lack of data on the indices in the control group, the number of patients with an altered level of the index (increase/decrease) was determined in comparison with the reference intervals.

The studied glycolysis indices had different functional and indicator values. An increase in lactic acid levels was detected in 10 (30%) children, as well as a decrease in 10 (30%) children. Lactic acid is a product of anaerobic glycolysis, and therefore an increase in its concentration may reflect a shift in the overall energy metabolism to the anaerobic ATP production with activation of anaplerotic (compensatory) pathways. Conversely, a decrease in lactic acid levels suggests depletion of its metabolic substrates. Pyruvic acid is a product of aerobic glycolysis, which is subsequently converted into acetyl-CoA and transferred to the mitochondria, where it is included in the Krebs cycle. An increased concentration of pyruvic acid in the urine of 18 (51%) children in the study group indicates a loss of energy substrate for the Krebs cycle. The obtained result indicates a change in anaerobic metabolism, which has a multidirectional character in children with ASD, accompanied by energy losses.

The glycolytic breakdown of glucose involves anaplerotic pathways of catabolism of branched-chain amino acids (BCAA—valine, leucine, isoleucine). Therefore, a change in the concentration of these metabolites that goes beyond the reference limits can be interpreted as an increase in catabolism (an increase in their concentration) or substrate decompensation (a decrease in their concentration). Among the analyzed group of patients, multidirectional changes in the level of branched-chain amino acid metabolites were revealed, reflected in Table 4. The results revealed indicate a disturbance in BCAA metabolism in ASD at different levels of their catabolic conversion.

Energy metabolism also includes beta-oxidation of fatty acids. Beta oxidation occurs in the mitochondria, so changes in urinary fatty acid concentrations may indicate mitochondrial dysfunction. This position is confirmed by the results presented in Table 4, reflecting the multidirectional change in the concentration of glutaric, sebacic, adipic, suberic and ethylmalonic acids.

The end products of beta-oxidation are ketone bodies, which are further converted into acetyl-CoA, which is the main substrate of the Krebs cycle. An increase in the concentration of ketogenesis indices (acetoacetic, 3-hydroxybutyric, malonic acid) is an indicator of the activation of alternative pathways for substrate supply of the Krebs cycle. The results presented in Table 4 indicate an increase in the concentration of 3-hydroxybutyric and malonic acids in 15 (43%) and 11 (31%) patients.

Among the Krebs cycle parameters, the median value was above the upper reference limit for fumaric acid, and 22 (63%) patients had elevated concentrations. A change in the concentration of fumaric acid that goes beyond the reference interval indicates a disturbance in the Krebs cycle at the level of converting fumaric acid into malic acid. Since there was a tendency for other Krebs cycle substrates to decrease, the increase in fumaric acid is likely due to its entry from the urea cycle, which may reflect hyperammonemia in patients in the studied cluster.

Meglutol is an indicator of coenzyme Q10 metabolism. Coenzyme Q10 is a biologically active molecule involved in oxidative phosphorylation of the mito-

Table 4. Content of energy metabolism indices in urine in studied children

Energy indicators exchange, mmol/mol creatinine	Number of patients with elevated concentration in urine, %	Number of patients with decreased concentration in urine, %
Glycolysis indices		
Lactic acid	29	29
Pyruvic acid	51	9
BCAA Metabolism Rates		
2-ketoisovaleric acid	34	20
3-methyl-2-oxovaleric acid	17	14
4-methyl-2-oxovaleric acid	37	6
3-hydroxyisovaleric acid	23	3
2-hydroxyisovaleric acid	20	37
3-methylcrotonylglycine	0	71
3-methylglutaric acid	0	77
Isovalerylglycine	17	23
Beta-oxidation indices		
Glutaric acid	14	29
Sebacic acid	37	20
Adipic acid	26	20
Suberic acid	20	20
Ethylmalonic acid	9	46
Ketogenesis indices		
Acetoacetic acid	14	–
3-hydroxybutyric acid	43	–
Malonic acid	31	–
Krebs cycle indicators		
Citric acid	9	43
Cis-aconitic acid	23	34
Isocitric acid	20	31
2-ketoglutaric acid	11	34
Succinic acid	17	29
Fumaric acid	63	20
Malic acid	20	26
Coenzyme Q10 Metabolism Index		
Meglutol	43	–

chondrial electron transport chain. Elevated meglutol levels in urine are additional evidence of impaired mitochondrial function.

When conducting a correlation analysis between markers of inflammatory status and energy metabolism, the relationships presented in Table 5 were revealed.

The data in Table 5 indicate multiple correlations between inflammatory markers and indicators of energy metabolism, as well as mitochondrial function.

Among the correlation links between inflammation indices of neuroimmunotest (LE, a-1PI, S100b AAB, MBP AAB) and energy metabolism indices, we found inverse correlation links. These correlations probably indicate energy expenditure and the activation of compensatory catabolic pathways.

A direct correlation was found between the average degree of NSE and the Krebs cycle, BCAA metabolism and beta-oxidation, indicating dysregulation of energy metabolism in the nervous tissue of children with ASD.

The kynurenine pathway is a catabolic pathway associated with inflammation and energy metabolism. We revealed a direct correlation between kynurenine pathway metabolites and energy metabolism indices. The strongest correlation was found for metabolites of

Table 5. Correlation coefficients between inflammatory status and energy metabolism indicators

Indices	Correlation coefficient
Inflammation	
a-1PI&PIC	–0.36
QUIN&NSE	0.41
Inflammation and Energy Metabolism	
LE&*cis*-aconitic acid	–0.37
LE&isovalerylglycine	–0.37
a-1PI&*cis*-aconitic acid	–0.45
a-1PI &isocitric acid	–0.35
a-1PI &malic acid	–0.36
a-1PI &3-hydroxybutyric acid	–0.41
a-1PI &adipic acid	–0.41
a-1PI &meglutol	–0.47
a-1PI &3-methyl-2-oxovaleric acid	–0.38
S100b AAB&citric acid	–0.35
S100b AAB&3-hydroxyisovaleric acid	–0.41
MBP AAB&fumaric acid	–0.35
MBP AAB&malic acid	–0.36
MBP AAB&meglutol	–0.42
MBP AAB&3-methyl-2-oxovaleric acid	–0.37
MBP AAB&3-hydroxyisovaleric acid	–0.41
NSE&*cis*-aconitic acid	0.43
NSE&isocitric acid	0.36
NSE&adipic acid	0.36
NSE&3-methylglutaric acid	0.37
KYNA&isocitric acid	0.45
KYNA&2-ketoglutaric acid	0.86
KYNA&acetoacetic acid	0.36
KYNA&suberic acid	0.52
KYNA&ethylmalonic acid	0.68
KYNA&isovalerylglycine	0.47
XAN&citric acid	0.38
XAN&*cis*-aconitic acid	0.45
XAN&isocitric acid	0.45
XAN&2-ketoglutaric acid	0.70
XAN&malic acid	0.48
XAN&acetoacetic acid	0.38
XAN&suberic acid	0.40
XAN&ethylmalonic acid	0.47
XAN&meglutol	0.56
XAN&3-methylcrotonylglycine	0.35
XAN&isovalerylglycine	0.82
QUIN&pyruvic acid	0.40
QUIN&citric acid	0.60
QUIN&cis-aconitic acid	0.61
QUIN&isocitric acid	0.60

Table 5. (Contd.)

Indices	Correlation coefficient
QUIN&succinic acid	0.45
QUIN&fumaric acid	0.37
QUIN&malic acid	0.50
QUIN&acetoacetic acid	0.40
QUIN&meglutol	0.58
QUIN&3-methylglutaric acid	0.35
Glycolysis	
Lactic acid&succinic acid	0.68
Lactic acid & malic acid	0.37
Lactic acid & 3-hydroxybutyric acid	0.81
Lactic acid & sebacic acid	0.93
Lactic acid&adipic acid	0.35
Lactic acid & suberic acid	0.46
Lactic acid & meglutol	0.57
Lactic acid & 2-ketoisovaleric acid	0.95
Lactic acid&3-methyl-2-oxovaleric acid	0.93
Lactic acid & 4-methyl-2-oxovaleric acid	0.83
Lactic acid & 3-hydroxyisovaleric acid	0.95
Lactic acid & 2-hydroxyisovaleric acid	0.37
Pyruvate&...	No
Krebs cycle	
Cis-aconitic acid&acetoacetic acid	0.38
Cis-aconitic acid&3-methyl-2-oxovaleric acid	0.45
Cis-aconitic acid&4-methyl-2-oxovaleric acid	0.35
Cis-aconitic acid&3-hydroxyisovaleric acid	0.38
Cis-aconitic acid&3-methylcrotonylglycine	0.38
Isocitric acid & acetoacetic acid	0.37
2-ketoglutaric acid&acetoacetic acid	0.60
2-ketoglutaric acid&adipic acid	0.41
2-ketoglutaric acid&suberic acid	0.64
2-ketoglutaric acid&ethylmalonic acid	0.75
2-ketoglutaric acid&3-methylglutaric acid	0.54
2-Ketoglutaric acid&isovalerylglycine	0.39
Succinic acid & acetoacetic acid	0.35
Succinic acid & 3-hydroxybutyric acid	0.56
Succinic acid&glutaric acid	0.57
Succinic acid&sebacic acid	0.65
Succinic acid&adipic acid	0.42
Succinic acid & suberic acid	0.50
Succinic acid&2-ketoisovaleric acid	0.59
Succinic acid&3-methyl-2-oxovaleric acid	0.60
Succinic acid&4-methyl-2-oxovaleric acid	0.52
Succinic acid&3-hydroxyisovaleric acid	0.70
Succinic acid & 3-methylglutaric acid	0.44
Fumaric acid&acetoacetic acid	0.75

Table 5. (Contd.)

Indices	Correlation coefficient
Fumaric acid&adipic acid	0.68
Fumaric acid&suberic acid	0.57
Fumaric acid & 3-methylglutaric acid	0.66
Malic acid & acetoacetic acid	0.45
Malic acid&adipic acid	0.61
Malic acid & suberic acid	0.46
Malic acid & ethylmalonic acid	0.47
Malic acid & 2-ketoisovaleric acid	0.37
Malic acid&3-methyl-2-oxovaleric acid	0.41
Malic acid & 4-methyl-2-oxovaleric acid	0.40
Malic acid & 3-hydroxyisovaleric acid	0.38
Malic acid & 3-methylglutaric acid	0.68
Ketogenesis	
Acetoacetic acid&adipic acid	0.54
Acetoacetic acid & suberic acid	0.56
Acetoacetic acid & 3-methylglutaric acid	0.60
3-hydroxybutyric acid&sebacic acid	0.85
3-hydroxybutyric acid&adipic acid	0.67
3-hydroxybutyric acid&suberic acid	0.71
3-hydroxybutyric acid&2-ketoisovaleric acid	0.78
3-hydroxybutyric acid&3-methyl-2-oxovaleric acid	0.84
3-hydroxybutyric acid&4-methyl-2-oxovaleric acid	0.77
3-hydroxybutyric acid&3-hydroxyisovaleric acid	0.77
3-hydroxybutyric acid&2-hydroxyisovaleric acid	0.65
3-hydroxybutyric acid&3-methylcrotonylglycine	0.37
3-hydroxybutyric acid & 3-methylglutaric acid	0.45
Malonic acid & 2-hydroxyisovaleric acid	0.88
Malonic acid & 3-methylcrotonylglycine	0.70
Beta oxidation	
Sebacic acid&2-ketoisovaleric acid	0.93
Sebacic acid&3-methyl-2-oxovaleric acid	0.91
Sebacic acid&4-methyl-2-oxovaleric acid	0.82
Sebacic acid&3-hydroxyisovaleric acid	0.91
Sebacic acid&2-hydroxyisovaleric acid	0.39
Adipic acid&3-methyl-2-oxovaleric acid	0.40
Adipic acid & 2-hydroxyisovaleric acid	0.44
Adipic acid & 3-methylglutaric acid	0.88
Suberic acid & 2-ketoisovaleric acid	0.51
Suberic acid&3-methyl-2-oxovaleric acid	0.52
Suberic acid & 4-methyl-2-oxovaleric acid	0.45
Suberic acid & 3-hydroxyisovaleric acid	0.51
Suberic acid & 2-hydroxyisovaleric acid	0.60
Suberic acid & 3-methylcrotonylglycine	0.45
Suberic acid & 3-methylglutaric acid	0.66
Ethylmalonic acid & 3-methylglutaric acid	0.43

Table 5. (Contd.)

Indices	Correlation coefficient
Coenzyme Q10	
Meglutol & *cis*-aconitic acid	0.47
Meglutol&succinic acid	0.59
Meglutol&fumaric acid	0.46
Meglutol&malic acid	0.48
Meglutol & Acetoacetic Acid	0.44
Meglutol&3-hydroxybutyric acid	0.54
Meglutol & Sebacic Acid	0.52
Meglutol&adipic acid	0.45
Meglutol & Suberic Acid	0.40
Meglutol&2-ketoisovaleric acid	0.48
Meglutol&3-methyl-2-oxovaleric acid	0.60
Meglutol&4-methyl-2-oxovaleric acid	0.61
Meglutol&3-hydroxyisovaleric acid	0.58
Meglutol&3-methylglutaric acid	0.39
Meglutol&isovalerylglycine	0.64

Only statistically significant correlations are presented.

the kynurenine pathway (KYNA, XAN, QUIN) with substrates of the Krebs cycle. The same connections between kynurenine pathway metabolites were found with indices of beta-oxidation, ketogenesis, and BCAA metabolism. This relationship between indices reflects the intensification of catabolic processes with the involvement of compensatory catabolic pathways in this group of patients.

A correlation analysis between the energy metabolism indices revealed no statistically significant relationships between pyruvic acid and energy metabolism indicators (data not shown in Table 5), which indicates the predominance of anaerobic glycolysis over aerobic glycolysis. At the same time, direct correlations were found between lactic acid and the Krebs cycle parameters, ketogenesis, beta-oxidation, and BCAA metabolism, which probably indicates the activation of compensatory pathways for the formation of glycolysis substrates and the Krebs cycle.

The results of the correlation analysis of Krebs cycle substrates, reflected in Table 5, show direct correlations with the indices of BCAA metabolism, beta-oxidation, and ketogenesis, which indicates the relationship between compensatory catabolic pathways in patients with ASD with high levels of inflammation.

Direct correlations between meglutol and the Krebs cycle, beta-oxidation, ketogenesis and BCAA metabolism parameters reflect a deficiency in coenzyme Q10 metabolism and, accordingly, mitochondrial dysfunction.

In general, the set of results obtained in this study indicate that inflammation, dysregulation of energy metabolism, and mitochondrial dysfunction are pathogenetic interrelated links of ASD and therefore can serve as diagnostic markers of ASD, as well as targets for therapeutic intervention in personalized therapy of patients (anti-inflammatory therapy and metabolic correction).

CONCLUSIONS

The most significant inflammatory indicators contributing to the pathogenesis of ASD are LE activity and the level of AAB to the neuroantigen S100b, as well as the level of the neurotoxic metabolite QUIN, formed as a result of inflammation-associated catabolism of tryptophan. In patients with ASD with a pronounced proinflammatory status, we found multidirectional changes in energy metabolism and mitochondrial function indicators, which were, probably, related to different degrees of compensation of impaired processes and their depletion. We also found statistically significant correlations in these patients. The obtained data reflect the most important pathogenetic links of ASD (inflammation/neuroinflammation, mitochondrial dysfunction, energy dysmetabolism); markers of these processes can serve as laboratory indicators for the diagnosis of ASD, as well as the basis for the development of innovative approaches associated with reducing the level of inflammation and correcting metabolic dysfunction in the complex therapy of patients with ASD.

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ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Ethical approval. Informed voluntary consent was obtained from each participant included in the study.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

REFERENCES

1. Balandina, O.V., Bondar, E.A., Vlasov, V.V., Grigorenko, E.L., Kuznetsova, Yu.E., Kuzyukova, A.A., Lebedeva, E.I., Martynikhin, I.A., Mendelevich, V.D., Meshkova, E.S., Namazova-Baranova, L.S., Novikov, A.Yu., Osin, E.K., Portnova, A.A., Rachin, A.P., Romanova, R.S., Sivolap, Yu.P., Talantseva, O.I., Ustinova, N.V., and Sharkov, A.A., *Clin. Recomm.* "Autism Spectrum Disorders" 2024, ID: 594_3.
2. Zeidan, J., Fombone, E., Scorah, J., Ibrahim, A., Durkin, M.S., Saxena, S., Yusuf, A., Shih, A., and Elsabbagh, M., *Autism Res.*, 2022, vol. 15, no. 5, pp. 778–790.
3. Hughes, H.K., Moreno, R.J., and Ashwood, P., *Brain Behav. Immun.*, 2023, vol. 108, pp. 245–254.
4. Matta, S.M., Hill-Yardin, E.L., and Crack, P.J., *Brain Behav. Immun.*, 2019, vol. 79, pp. 75–90.
5. Toscano, C.V.A., Barros, L., Lima, A.B., Nunes, T., Carvalho, H.M., and Gaspar, J.M., *Neurosci. Biobehav. Rev.*, 2021, vol. 129, pp. 63–74.
6. Liao, X., Yang, J., Wang, H., and Li, Y., *J. Psychiatr. Res.*, 2020, vol. 130, pp. 167–176.
7. Liao, X., Liu, Y., Fu, X., and Li, Y., *Mol. Neurobiol.*, 2020, vol. 57, pp. 3424–3438.
8. Shen, Y., Zhong, J.G., Lan, W.T., Li, Y.H., Gong, J.H., Zhao, B.X., and Hou, X.H., *Front. Psychiatry*, 2023, vol. 14, p. 1086068.
9. Than, U.T.T., Nguyen, L.T., Nguyen, P.H., Nguyen, X.H., Trinh, D.P., Hoang, D.H., Nguyen, P.A.T., and Dang, V.D., *Sci. Rep.*, 2023, vol. 13, no. 1, p. 22587.
10. Klyushnik, T.P., Androsova, L.V., Simashkova, N.V., Zozulya, S.A., Otman, I.N., and Koval-Zaitsev, A.A., *Zh. Nevrol. Psikhiatr. im. S.S. Korsakova*, 2011, vol. 111, no. 8, pp. 41–45.
11. Hughes, H.K., Moreno, R.J., and Ashwood, P., *Brain Behav. Immun.*, 2023, vol. 108, pp. 245–254.
12. Wang, H., and Ye, J., *Rev. Endocr. Metab. Disord.*, 2015, vol. 16, pp. 47–54.
13. Divella, R., Gadaleta Caldarola, G., and Mazzocca, A., *J. Clin. Med.*, 2022, vol. 11, no. 8, p. 2191.
14. Liu, X., Jiang, N., and Zhou, W., *Molecules*, 2023, vol. 28, no. 11, p. 4501.
15. Elkins, C., and Li, C., *Immunometabolism*, 2022, vol. 4, no. 4.
16. Morrison, T., Watts, E.R., Sadiku, P., and Walmsley, S.R., *Immunol. Rev.*, 2023, vol. 314, no. 1, pp. 427–441.
17. Budikhina, A.S., and Pashchenkov, M.V., *Immunologiya*, 2021, vol. 42, no. 1, pp. 5–20.
18. Simashkova, N.V., Klyushnik, T.P., and Yakupova, L.P., *Psikhiatriya*, 2018, vol. 78, no. 2, pp. 17–24.
19. Dotsenko, V.L., Neshkova, E.A., and Yarovaya, G.A., *Vopr. Med. Khim.*, 1994, vol. 40, no. 3, pp. 20–25
20. Nartikova, V.F., and Paskhina, T.S., *Vopr. Med. Khim.*, 1979, vol. 25, no. 4, pp. 494–499.
21. Skripchenko, N.V., and Shirokova, A.S., *Pediatr. Neurosurg. Neurol.*, 2016, no. 4, pp. 16–25.
22. Abou Chahla, M.N., Khalil, M.I., Comai, S., Brundin, L., Erhardt, S., and Guillemin, G.J., *Brain Sci.*, 2023, vol. 13, no. 3, p. 505.
23. Shilov, Yu.E. and Bezrukov, M.V., *Vestn. Ross. Akad. Med. Nauk*, 2013, vol. 68, no. 1, pp. 35–41.
24. Klyushnik, T.P., Ivanov, A.B., and Petrov, V.G., *Zh. Nevrol. Psikhiatr. im. S.S. Korsakova*, 2014, vol. 114, no. 2, p. 37.
25. Zozulya, S.A., Smirnova, L.K., and Kuznetsov, N.E., *Psikh. Zdorov.*, 2017, vol. 15, no. 1, pp. 11–18.
26. Santana-Coelho, D., *Brain Behav. Immun. Health*, 2024, p. 10083.
27. Gevi, F., Zolla, L., Gabriele, S., and Persico, A.M., *Mol. Autism*, 2016, vol. 7, pp. 1–11.
28. Saade, M.C., Clark, A.J., and Parikh, S.M., *Front. Nutr.*, 2022, vol. 9, p. 1070435.
29. Kominsky, D.J., Campbell, E.L., and Colgan, S.P., *J. Immunol.*, 2010, vol. 184, no. 8, pp. 4062–4068.
30. Anashkina, A.A., and Erlykina, E.I., *Sovrem. Tekhnol. Med.*, 2021, vol. 13, no. 1, pp. 78–92.
31. Ye, Z., Wang, S., Zhang, X., Li, H., Xu, Z., and Zuo, Y., *Front. Endocrinol.*, 2020, vol. 11, p. 617.

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