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11 EFFICACY OF INFLIXIMAB IN AUTISM SPECTRUM DISORDERS IN CHILDREN ASSOCIATED WITH GENETIC DEFICIENCY OF THE FOLATE CYCLE

JUSTIFICATION

The notion of systemic inflammation in autism spectrum disorders in children has been established. A recent meta-analysis of randomized controlled trials published in 2019, which included a systematic review of 25 case-control studies, suggests an association between genetic deficiency of the folate cycle and autism spectrum disorders in children [18]. This evidence is consistent with an earlier meta-analysis of randomized controlled trials from 2013, which included data from 8 studies [17].

The encephalopathy that develops in children with genetic deficiency of the folate cycle and manifests as autism spectrum disorders is associated with oxidative stress. The reason for the latter can be seen in the suppression of the immune system with the development of a special form of immunodeficiency, which is based on the deficiency of natural killers, natural killer T lymphocytes and CD8 + cytotoxic T cells [11]. Immunodeficiency mediates all three known mechanisms of brain damage in children with genetic deficiency of the folate cycle, namely the development of opportunistic infections [2, 15], autoimmune reactions against neuronal antigens [3, 6] and manifestations of systemic inflammation, which is based on the phenomenon of hypercytokinemia [13, 20].

Children with autism spectrum disorders have been shown to have overproduction of several proinflammatory cytokines, including tumor necrosis factor alpha (TNF-alpha), interleukin-1beta, and interleukin-6. A recent systematic review and meta-analysis of controlled clinical trials showed increased serum concentrations of the proinflammatory mediators interleukin-1beta ($P < 0.001$), interleukin-6 ($P = 0.03$), interleukin-8 ($P = 0.04$), interferon-gamma ($P = 0.02$), eotaxin ($P = 0.01$), monocyte chemotactic factor 1 ($P < 0.05$), and decreased levels of the anti-inflammatory cytokine transforming growth factor beta 1 ($P < 0.001$) in children with autism spectrum disorders ($n = 743$) compared with healthy controls ($n = 592$) [13].

The results of the latest meta-analysis of randomized controlled clinical trials prepared by Saghazadeh A. et al., which includes 38 studies involving 2487 children, show a significant increase in serum concentrations of TNF-alpha, gamma-interferon, interleukin 1 beta and interleukin 6 in children with autism spectrum disorders compared with healthy individuals [19].

Each of the mechanisms of damage, as well as the causal immunodeficiency, can be the subject of therapeutic interventions. The neurotropic effects of proinflammatory cytokines are well known [19, 20], and persistent hypercytokinemia can lead to impaired cerebral metabolism, neurogenesis, bioelectric activity and disorganization of mental activity in children with genetic deficiency of the folate cycle.

This study aims to investigate the clinical effects of anti-inflammatory therapy aimed at eliminating abnormal hypercytokinemia. It is necessary to determine whether anti-inflammatory therapy will have the desired neuroprotective effect, contributing to the progress of mental development in children with autism spectrum disorders. As is known, TNF-alpha is a master cytokine, the production of which depends on the implementation of the entire pro-inflammatory cytokine cascade in the human body.

Targeted inhibition of TNF-alpha using specific monoclonal antibody preparations is a therapeutic strategy that has proven itself in a number of severe immunoinflammatory diseases, including rheumatoid arthritis, Crohn's disease, autoimmune spondyloarthropathies and psoriatic arthritis [14]. This strategy may also be useful for suppressing persistent systemic inflammation in children with autism spectrum disorders associated with genetic deficiency of the folate cycle.

The aim of the research: to study the clinical efficacy of infliximab in children with autism spectrum disorders associated with genetic deficiency of the folate cycle, in whom elevated serum TNF-alpha concentrations were noted.

Materials and methods. This prospective controlled single-center non-randomized clinical study included 225 children diagnosed with autism spectrum disorders associated with genetic deficiency of the folate cycle. The diagnosis of autism spectrum disorders was made by psychiatrists from regional hospitals or specialized departments according to DSM–IV–TR (Diagnostic and Statistical Manual of mental disorders) and ICD–10 criteria. Children were recruited into the study group (SG) in 2019–2020. These were patients from different regions of Ukraine aged 2 to 9 years, in whom elevated serum TNF-alpha concentrations were observed. As is known, the phenotype of genetic deficiency of the folate cycle includes 5 main syndromes: autism spectrum disorders, intestinal syndrome (persistent enteritis/colitis) [7], PANDAS [4, 9], epileptic syndrome [5] and signs of pyramidal tract damage.

SG children were administered intravenously drip infliximab (a monoclonal antibody preparation to TNF-alpha) at a dose of 3 mg/kg twice a month for 1–3 consecutive months to achieve an anti-inflammatory effect by targeted neutralization of the indicated pro-inflammatory cytokine, according to the cytokine profile.

The control group (CG) consisted of 51 similar children with a similar age and gender distribution, with a corresponding increase in serum TNF-alpha concentration. These patients did not receive infliximab therapy, but underwent only conventional rehabilitation measures, which included work with a speech therapist/specialist, specially trained teachers and psychiatrists. The dynamics of mental symptoms of autism spectrum disorders during the study was assessed using the Aberrant Behavior Checklist (ABC) scale [1].

Folate cycle gene polymorphisms were detected by polymerase chain reaction (PCR) with restriction analysis in three centers: Neurological Research Institute (USA), Kharkiv Specialized Medical Genetic Center and a commercial laboratory in Kyiv. The nucleotide substitutions MTHFR 677 C > T, MTHFR + 1298 A > C, MTRR 66 A > G, and MTR 2756 A > G were detected in various combinations in the homozygous and heterozygous states.

The concentration of the cytokine TNF-alpha in the blood serum was determined by solid-phase enzyme-linked immunosorbent assay (reagents «Vector-Best», RF; norm – 0-8 ng/ml). The average content of this cytokine in the blood serum in SG was 13.4 ± 0.21 ng/ml, and in CG – 12.7 ± 0.24 ng/ml.

All study participants underwent a serial comprehensive immunological examination, which, in addition to a general blood test, included the study of the subpopulation composition of lymphocytes using laser flow cytofluorimetry (cytofluorimetry Epics XI, USA) and the indirect immunofluorescence method with monoclonal antibodies to CD markers with two or three labels (CD3 + , CD3 + CD4 + , CD3 + CD8 + , CD3–CD19 + , CD3–CD16 + CD56 + , CD3 + CD16 + CD56 +) (Beckman Coulter reagents, USA). Phagocytosis was

assessed according to the latex test with the determination of the phagocytosis index, phagocytic index, number of active phagocytes and phagocytic capacity of the blood, as well as the activity of the enzymes myeloperoxidase (cytofluorimetry) and NADP-oxidase (NST test). Serum concentrations of immunoglobulins of the main classes (M, G, A) were determined by the results of simple radial immunodiffusion according to Mancini. The concentration of IgE, IgD and IgG subclasses (IgG1, IgG2, IgG3, IgG4) in serum was measured using solid-phase enzyme immunoassay (VectorBEST reagents, RF). SG patients had immunodeficiency associated with genetic deficiency of the folate cycle, which was considered to be the cause of the increased content of TNF-alpha in serum.

In addition, the diagnosis of reactivated viral infection was performed based on the results of quantitative PCR of blood serum with species-specific primers for herpesviruses (herpes simplex viruses 1 and 2 types (HSV-1 and HSV-2), varicella-zoster virus (VZV), Epstein-Barr virus (EBV), cytomegalovirus (CMV), human herpes viruses 6, 7 and 8 types (HHV-6, HHV-7, HHV-8)), TTV, measles and KSDS viruses (DNA-Technology reagents, RF). Serological tests were also performed by performing solid-phase enzyme-linked immunosorbent assay to identify specific IgM and IgG in blood serum to *Candida albicans*, *Mycoplasma pneumoniae* and *Chlamydia pneumoniae* (Vector-BEST reagents, RF). Western blot was used to identify borreliosis infection.

Intracellular neurotropic pathogens predominated in SG children: viruses with opportunistic properties, especially human lymphotropic herpesviruses (EBV, CMV, HHV-6, HHV-7), which is consistent with the typical deficiency of NK and NKT cells in such patients. The second most frequently detected was reactivated TTV infection, the neurotropic nature of which was recently reported [8]. The third most frequently identified was beta-hemolytic streptococcus group A, involved in the induction of autoimmune complications, including PANDAS. Candida, Borrelia, Mycoplasma, and Chlamydia were less frequently detected. The high microbial load was consistent with the data on the presence of immunodeficiency in children with a genetic disorder of the folate cycle.

Serum concentrations of known biomarkers of genetic deficiency of the folate cycle – homocysteine, folic acid, vitamins B12 and B6, creatinine, uric acid – were also assessed using available biochemical methods.

All children underwent control MRI scans of the brain in conventional modes (T1 and T2-weighted, FLAIR) on tomographs with a magnetic induction value of at least 1.5 T, at least twice: before and after participation in the study. Typical were signs of leukoencephalopathy of varying severity with a predominant violation of white matter myelination in the parietal lobes of the cerebral hemispheres (**Fig. 11.4**). Also in 57 % of cases there was an additional picture of temporal median sclerosis with the phenomenon of hyperintensity from the hippocampi and insula. Mostly such children suffered from epileptic syndrome of the temporal median epilepsy type or had pathological epileptiform activity on EEG. In 19 % of cases, typical signs of congenital cytomegalovirus neuroinfection were noted in the form of ventriculomegaly, periventricular foci, cysts in the poles of the temporal lobes, hypogenesis of the corpus callosum and zones of delayed myelination of white matter in the parietal lobes of the cerebral hemispheres. These data correspond to the results of an 18-year retrospective study by Pinillos-Pisón R. et al. [16]. Such children usually had symptoms of damage to the pyramidal tracts, and therefore were often diagnosed with cerebral palsy, although autistic mental disorders and other symptoms were also observed.

The criteria for inclusion of the patient in the study were the presence of 2–4 polymorphisms of folate cycle genes, NK and/or NKT cell deficiency, reactivated infection caused by lymphotropic herpesviruses and/or measles virus, signs of leukoencephalopathy/temporal median sclerosis on brain MRI, clinical symptoms of autism spectrum disorders. The criteria for exclusion of the patient from the study were: refusal of the child's parents to participate in the study, the presence of additional genetic pathology involved in the development of the picture of existing mental disorders, the absence of a phenotype of NK and/or NKT cell deficiency and signs of leukoencephalopathy, as well as the development of side effects of immunotherapy that make it impossible to continue the approved treatment. The study endpoints were the main clinical manifestations of autism spectrum disorders, serum TNF-alpha concentration, and the dynamics of the main additional clinical manifestations associated with genetic deficiency of the folate cycle, including PANDAS, persistent enteropathy/colitis, temporal median epilepsy, and symptoms of pyramidal tract damage.

Statistical analysis of the obtained information was processed using the Microsoft Excel electronic program using structural and comparative analysis methods. In order to establish the reliability of the differences in the results, the Student's T-test was used with the calculation of the confidence probability coefficient p (parametric criterion) and the number of signs Z according to Urbach (non-parametric criterion). Pearson's chi-square (χ2) was calculated with the Fisher's exact test and the determination of the Yates correction to study the relationship between the appointment of immunotherapy and the studied indicators' dynamics. The χ criterion, the Pearson correlation coefficient (C) and its normalized value were also calculated to determine the strength of the detected relationships.

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Research results and their discussion. This chapter discusses the clinical effects of anti-inflammatory therapy aimed at eliminating abnormal hypercytokinemia. It is necessary to establish whether anti-inflammatory therapy will have the desired neuroprotective effect, contributing to the progress of mental development in children with ASD. As is known, TNF-alpha is a master cytokine, the production of which depends on the implementation of the entire pro-inflammatory cytokine cascade in the human body. Targeted inhibition of TNF-alpha using specific monoclonal antibody preparations is a therapeutic strategy that has proven itself in a number of severe immunoinflammatory diseases, including rheumatoid arthritis, Crohn's disease, autoimmune spondyloarthropathies and psoriatic arthritis [14]. This strategy may also be useful for suppressing persistent systemic inflammation in children with ASD associated with GDFC.

According to the results obtained, infliximab was effective in reducing the clinical symptoms of ASD disorders in 69 of 92 SG children who had elevated serum TNF-alpha concentrations at the beginning of the study (76 % of cases), but the severity of the achieved clinical effect varied in different patients and in relation to different indicators of mental activity (**Table 11.1; Fig. 11.5**). In the CG, during the same observation period, improvement in mental status was noted in only 17 of 47 patients (36 % of cases), which is half as much as in the SG ($p < 0,05$; $Z_{0,05}$). These significant differences indicate a certain positive modifying effect of infliximab on the state of the psyche in children with ASD associated with GDFC. Adding infliximab to standard in clinical practice specialized educational programs provides tangible additional benefits,

primarily in stabilizing the emotional state of the child, normalizing everyday activity and increasing the resistance of the psychics to stress factors.

In SG, there was a pronounced positive dynamics in the direction of hyperactivity, hyperexcitability and stereotyped behavior, but no significant effect was noted on the stability of eye contact and the development of expressive-receptive language, while in CG some positive changes were achieved specifically in terms of expressive language and the level of eye contact, which indicates different points of action of infliximab and specialized educational programs (**Table 11.1**). The psychotropic effect obtained with infliximab differs from that of intravenous immunoglobulin, which has also demonstrated clinical efficacy in ASD associated with GDFC [10, 12]. The changes induced by infliximab are more pronounced and develop in a shorter time frame, but they are significantly narrower in terms of the spectrum of positive psychotropic effects compared to high-dose immunoglobulin therapy, which has a total modifying effect on the psyche of such children.

● **Table 11.1.** ABC scale scores in SG (n = 225) and CG patients (n = 51)

Subscales	SG	CG
ABC		
Irritability	5.2 ± 0.6 *	11.5 ± 0.9
Hyperactivity	9.1 ± 0.9 *	19.7 ± 1.4
Inadequate eye contact	7.3 ± 1.8	9.5 ± 1.3
Inappropriate speech	7.9 ± 0.8	8.5 ± 1.4
Symptom checklist		
Drowsiness	8.4 ± 0.8	12.5 ± 1.7
Decreased activity	4.8 ± 0.6	4.9 ± 0.9

*Note. * – p < 0,05: Z < Z0,05*

The average total ASD score in SG children according to the ABC scale decreased slightly, which indicates a moderate positive overall effect of infliximab on the state of the respondents' psyche. However, the achieved positive effect as a result of the immunotherapy was uneven and concerned only individual indicators of the ABC scale. Significant and rapid positive dynamics were achieved in the level of hyperactivity and hyperexcitability. Thus, the average hyperactivity score for a three-month course of immunotherapy decreased by 2.5 times, and the total excitability score – almost three times. At the same time, there was a small and statistically insignificant dynamics in the indicators of language development and the level of eye contact. These data allow us to assume that infliximab acts on the psyche of children with autism spectrum disorders selectively, having mainly a psychostabilizing effect, calming the child by reducing the manifestations of hyperactivity and hyperexcitability. This effect is quite pronounced and develops in a short period of time (**Fig. 11.1**).

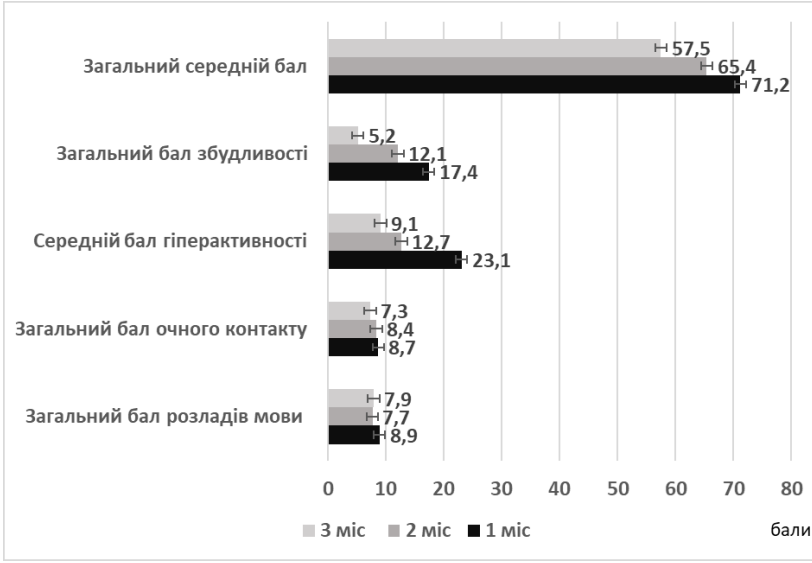
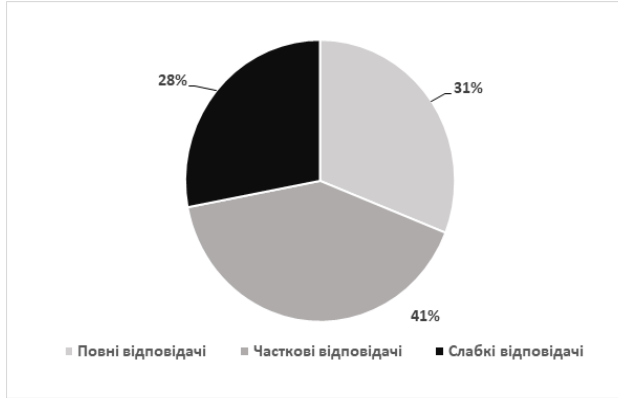


Fig. 11.1. Dynamics of the main indicators of the ABC scale in SG children (n = 225) during a 3-month course of infliximab immunotherapy

If we talk about the structure of psychotropic effects when using infliximab in SG children, then almost complete elimination of manifestations of hyperactivity and hyperexcitability was observed in 21 out of 69 responders to immunotherapy (31 % of cases) among SG patients (**Fig. 11.2**). Such children, in terms of daily motor activity and mental excitability, approached similar levels in healthy children at the end of the immunotherapy course. Thus, every third child among the responders to treatment and every fourth among all SG participants was a complete responder to infliximab immunotherapy in terms of daily motor activity and mental excitability. In another 28 SG children (41 % of cases among responders), the levels of hyperactivity and hyperexcitability during the period of infliximab immunotherapy decreased by at least half compared to the initial ones, which indicates a partial positive response to the treatment. In the remaining 19 patients from the subgroup of responders to immunotherapy among SG participants (28 % of cases), there was a less pronounced decrease in clinical manifestations of hyperactivity and hyperexcitability according to the ABC scale, which allows us to consider them as weak responders to treatment. As noted above, 9 of 38 SG children (24 %) did not respond to infliximab immunotherapy with changes in mental activity and were considered non-responders to the treatment. Their daily motor activity and mental arousal remained at the same level despite immunotherapeutic interventions according to the study protocol.



⬡ **Fig. 11.2.** Structure of responders to infliximab therapy by indicators of hyperactivity and hyperexcitability according to the ABC scale among SG patients (n = 225)

The study of the dynamics of serum TNF-alpha concentration in SG and CG children showed normalization of the amount of this pro-inflammatory cytokine in the blood serum of SG children and a significant decrease in this indicator compared to CG, where the average serum TNF-alpha concentration remained elevated at the end of the observation period, as at the beginning of the study (**Fig. 11.3**). These differences can be explained by the direct therapeutic effect of infliximab, whose monoclonal antibodies directly neutralize TNF-alpha molecules by specific recognition and binding [14]. If we talk about the structure of the anti-inflammatory effect of infliximab among SG patients, then the normalization of serum TNF-alpha concentration occurred in the majority of respondents (in 61 of 92 patients; 66 % of cases), and a decrease in this concentration compared to the initial level was observed in almost all children who received infliximab (in 90 of 92 people). At the same time, in the CG, normalization of serum TNF-alpha concentration occurred in only 7 of 47 patients (14 % of cases), and a decrease in the concentration was noted in another 11 respondents (23 % of cases), which was a striking contrast with the SG data ($p < 0,05$; $Z_{0,05}$).

These data suggest that the achieved positive psychotropic effects in SG on hyperactivity and hyperexcitability were associated with the ability of infliximab to targetably reduce serum TNF-alpha concentration, i.e. to have a systemic anti-inflammatory effect. To verify this conclusion, we additionally studied the association between changes in serum TNF-alpha concentration and the dynamics of mental disorders in SG children during the observation period. To study the association between changes in serum TNF-alpha concentration and hyperactivity and hyperexcitability indicators on the ABC scale, we calculated the Pearson chi-square (χ^2) index. We proceeded from the data that there were 69 clinical responders to immunotherapy out of 92 SG patients, but only 61 SG participants had normalization of serum TNF-alpha concentration on the background of infliximab administration. All patients with normalized serum TNF-alpha levels, except for one child, demonstrated a clinical response to immunotherapy according to the ABC scale. The calculation results of conjugation indices are shown in **Tabs. 11.2** and **11.3**.

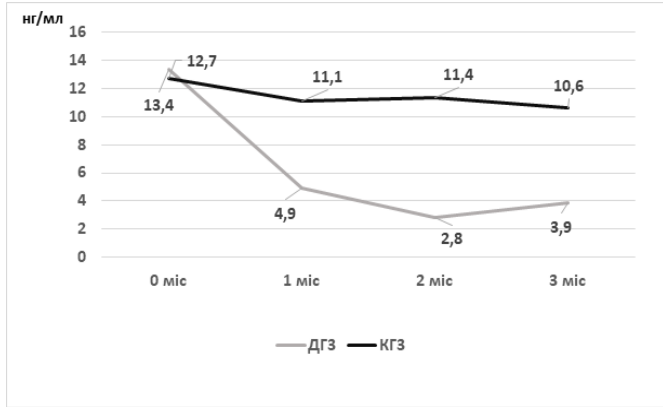


Fig. 11.3. Dynamics of serum TNF-alpha concentration in SG (n = 225) and CG (n = 51) during the observation period

Table 11.2. Results of calculating the Pearson chi-square (χ^2) test in SG (n = 225) when studying the association between serum TNF-alpha concentration and clinical outcome according to the ABC scale

	SG	CG	χ^2	χ^2 at p = 0,05	χ^2 at p = 0,01	significance	Yates' correction	significance	Fisher's exact test	significance
TNF-alpha/ hyperactivity-hyperexcitability	38	22	18,768	3,841	6,635	p < 0,001	15,555	< 0,001	0,00004	p < 0,05

Table 11.3. Calculation of the strength of the relationship between serum TNF-alpha concentration and hyperactivity and hyperexcitability indicators according to the ABC scale in SG children (n = 225)

Criterion name	TNF-alpha / hyperactivity-hyperexcitability	
	value of the criterion	connection strength
criterion φ	0,703	strong
Pearson correlation coefficient (C)	0,575	relatively strong
Normalized value of Pearson coefficient (C')	0,813	very strong

The obtained data prove the existence of a connection between the normalization of serum TNF-alpha concentration and a decrease in the severity of hyperexcitability and hyperactivity in children with ASD SG. This gives grounds to assert that it is the suppression of TNF-alpha and the achievement of a systemic anti-inflammatory

effect in this connection that is the reason for the achieved positive dynamics of mental status indicators in SG children. As is known, TNF-alpha, like other pro-inflammatory cytokines, has certain neurotropic properties, affecting the human psyche [13, 19, 20]. An increase in serum TNF-alpha concentration in children with autism spectrum disorders changes their mental state, potentiating the manifestations of hyperactivity and hyperexcitability, and a targeted effect on TNF-alpha molecules using a drug of specific monoclonal antibodies, leveling the effects of TNF-alpha, contributes to improving the levels of everyday motor activity and mental excitability in children with ASD.

In addition to autism, the effect of infliximab immunotherapy on other manifestations of GDFC, which are also based on an immunoinflammatory reaction that can be attenuated or blocked by a tested immunobiological agent, was evaluated. A decrease in clinical manifestations of such syndromes as intestinal dysfunction, PANS/PITANDS/PANDAS and epileptiform activity of the cerebral cortex was noted, but no positive dynamics were recorded in the movement disorders caused by damage to the pyramidal tracts of the CNS.

Thus, the manifestations of PANS/PITANDS/PANDAS in SG, namely tic hyperkinesis and obsessive-compulsive syndrome, decreased in 77 % of cases among SG patients diagnosed with streptococcal-induced autoimmunity with CNS involvement. There were significant differences from the CG, in which a decrease in PANS/PITANDS/PANDAS symptoms was observed in only 14 % of cases ($p < 0,05$; $Z < Z_{0,05}$). An increase in the number of days with normal bowel movements by at least one third of the initial level in SG children, indicating partial compensation of the intestinal syndrome, occurred in 82 % of cases among patients who had signs of intestinal dysfunction in the form of persistent diarrhea and/or constipation. The differences with the data in the CG were also significant, since similar positive dynamics in intestinal symptoms among these children was observed only in 23 % of cases ($p < 0,05$; $Z < Z_{0,05}$). Reduction or disappearance of epileptiform bioelectric activity of the cerebral cortex according to EEG data in SG was registered in 81 % of cases among patients who had epileptic seizures or, at least, pathological epileptiform activity of the cerebral cortex during EEG. In CG children, positive dynamics of the epileptic syndrome was found only in 27 % of cases, which indicates significant differences with SG data both when calculating the parametric and nonparametric criteria ($p < 0,05$; $Z < Z_{0,05}$) (**Table 11.4**). However, no significant differences were found in the observation groups regarding the dynamics of clinical manifestations of damage to the pyramidal nerve pathways of the CNS ($p > 0,05$; $Z > Z_{0,05}$).

● **Table 11.4.** Differences in study endpoints between SG (n = 225) and CG patients (n = 51)

End point	SG, %		CП, %		t-test	Number of characters Z
	+	–	+	–		
ASD	76	24	36	64	p < 0,05 *	Z < Z0,05 *
PANDAS	77	23	14	86	p < 0,05 *	Z < Z0,05 *
Epileptic syndrome	81	19	27	73	p < 0,05 *	Z < Z0,05 *
Intestinal syndrome	82	18	23	77	p < 0,05 *	Z < Z0,05 *
Pyramid disorders	24	76	26	74	p > 0,05	Z > Z0,05

* – *reliable differences*

Thus, infliximab helps to reduce not only some manifestations of ASD, but also has a broader, systemic effect, allowing to reduce the manifestations of the main clinical syndromes of the broad GDFC phenotype. Educational programs and corrective exercises, which are still the basis of therapy for children with ASD, do not allow to achieve the indicated positive changes in the work of the brain, intestines and immune system, as evidenced by significant differences with the results in CG.

Conclusions. Infliximab leads to significant improvements in hyperactivity and hyperexcitability, as well as stereotypic behavior in children with autism spectrum disorders associated with genetic deficiency of the folate cycle. Responders to immunotherapy are 76 % of patients with this pathology, which is twice as high as with standard therapy. However, there is no effect of infliximab on such manifestations of autism as the level of eye contact and language development. Psychotropic effects of infliximab immunotherapy are closely related to the normalization of previously elevated serum TNF-alpha concentrations and are probably due to the elimination of the pathological activating effect of this pro-inflammatory cytokine on CNS neurons. In parallel, there is an improvement in other clinical syndromes of genetic deficiency of the folate cycle in children with autism spectrum disorders – intestinal pathology, epileptic syndrome and PANDAS, in the pathogenesis of which, as is known, TNF-alpha and the systemic and intracerebral inflammation induced by this cytokine are involved. However, under the influence of immunotherapy, there is no change in the dynamics of motor deficit in children with symptoms of pyramidal tract damage. Further clinical studies in this direction with a larger number of participants and randomization are necessary to obtain more convincing data.

REFERENCES

1. Amman, M. G., Singh, N. N., Stewart, A. W., Field, C. J. (1985). Psychometric characteristics of the aberrant behavior checklist. Journal of Mental Deficiency, 89, 492–502.
2. Binstock, T. (2001). Intra-monocyte pathogens delineate autism subgroups. Medical Hypotheses, 56 (4), 523–531. https://doi.org/10.1054/mehy.2000.1247
3. Cabanlit, M., Wills, S., Goines, P., Ashwood, P., Van de Water, J. (2007). Brain-Specific Autoantibodies in the Plasma of Subjects with Autistic Spectrum Disorder. Annals of the New York Academy of Sciences, 1107 (1), 92–103. https://doi.org/10.1196/annals.1381.010
4. Esposito, S., Bianchini, S., Baggi, E., Fattizzo, M., Rigante, D. (2014). Pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections: an overview. European Journal of Clinical Microbiology & Infectious Diseases, 33 (12), 2105–2109. https://doi.org/10.1007/s10096-014-2185-9
5. Frye, R. E. (2015). Metabolic and mitochondrial disorders associated with epilepsy in children with autism spectrum disorder. Epilepsy & Behavior, 47, 147–157. https://doi.org/10.1016/j.yebeh.2014.08.134
6. Frye, R. E., Sequeira, J. M., Quadros, E. V., James, S. J., Rossignol, D. A. (2012). Cerebral folate receptor autoantibodies in autism spectrum disorder. Molecular Psychiatry, 18 (3), 369–381. https://doi.org/10.1038/mp.2011.175
7. Furlano, R. I., Anthony, A., Day, R., Brown, A., McGarvey, L., Thomson, M. A. et al. (2001). Colonic CD8 and γδ T-cell infiltration with epithelial damage in children with autism. The Journal of Pediatrics, 138 (3), 366–372. https://doi.org/10.1067/mpd.2001.111323

8. Ikuta, Y., Oba, K., Nai, E., Katori, T., Hashino, M., Inamine, Y. et al. (2019). Aseptic meningitis caused by torque teno virus in an infant: a case report. Journal of Medical Case Reports, 13 (1). https://doi.org/10.1186/s13256-019-2233-2
9. Jyonouchi, H., Geng, L., Streck, D. L., Toruner, G. A. (2012). Immunological characterization and transcription profiling of peripheral blood (PB) monocytes in children with autism spectrum disorders (ASD) and specific polysaccharide antibody deficiency (SPAD): case study. Journal of Neuroinflammation, 9 (1). https://doi.org/10.1186/1742-2094-9-4
10. Maltsev D.V. Efficiency of a high dose of intravenous immunoglobulin in children with autistic spectrum disorders associated with genetic deficiency of folate cycle enzymes // Journal of global pharma technology. – 2019. – Vol. 11(05). – P. 597–609.
11. Maltsev, D. (2020). Features of folate cycle disorders in children with ASD. Bangladesh Journal of Medical Science, 19 (4), 737–742. https://doi.org/10.3329/bjms.v19i4.46634
12. Maltsev D.V. High-dose intravenous immunoglobulin therapy efficiency in children with autism spectrum disorders associated with genetic deficiency of folate cycle enzymes // Psychiatry, Psychotherapy and Clinical Psychology. – 2016. – Vol. 2. – P. 63–80.
13. Masi, A., Quintana, D. S., Glozier, N., Lloyd, A. R., Hickie, I. B., Guastella, A. J. (2014). Cytokine aberrations in autism spectrum disorder: a systematic review and meta-analysis. Molecular Psychiatry, 20 (4), 440–446. https://doi.org/10.1038/mp.2014.59
14. Melsheimer, R., Geldhof, A., Apaolaza, I., Schaible, T. (2019). Remicade®(infliximab): 20 years of contributions to science and medicine. Biologics: Targets and Therapy, 13, 139–178. https://doi.org/10.2147/btt.s207246
15. Nicolson, G. L., Gan, R., Nicolson, N. L., Haier, J. (2007). Evidence for Mycoplasma ssp., Chlamydia pneunomiae, and human herpes virus-6 coinfections in the blood of patients with autistic spectrum disorders. Journal of Neuroscience Research, 85 (5), 1143–1148. https://doi.org/10.1002/jnr.21203
16. Pinillos Pisón, R., Llorente Cereza, M. T., López Pisón, J., Pérez Delgado, R., Lafuente Hidalgo, M., Martínez Sapiñá, A., Peña Segura, J. L. (2009). Congenital infection by cytomegalovirus. A review of our 18 years' experience of diagnoses. Revista de Neurología, 48 (7), 349–353. https://doi.org/10.33588/rn.4807.2008391
17. Pu, D., Shen, Y., Wu, J. (2013). Association between MTHFR Gene Polymorphisms and the Risk of Autism Spectrum Disorders: A Meta-Analysis. Autism Research, 6 (5), 384–392. https://doi.org/10.1002/aur.1300
18. Sadeghiyeh, T., Dastgheib, S. A., Mirzaee-Khoramabadi, K., Morovati-Sharifabad, M., Akbarian-Bafghi, M. J., Poursharif, Z. et al. (2019). Association of MTHFR 677C > T and 1298A > C polymorphisms with susceptibility to autism: A systematic review and meta-analysis. Asian Journal of Psychiatry, 46, 54–61. https://doi.org/10.1016/j.ajp.2019.09.016
19. Saghazadeh, A., Ataeinia, B., Keynejad, K., Abdolalizadeh, A., Hirbod-Mobarakeh, A., Rezaei, N. (2019). A meta-analysis of pro-inflammatory cytokines in autism spectrum disorders: Effects of age, gender, and latitude. Journal of Psychiatric Research, 115, 90–102. https://doi.org/10.1016/j.jpsychires.2019.05.019
20. Smith, S. E. P., Li, J., Garbett, K., Mirnics, K., Patterson, P. H. (2007). Maternal Immune Activation Alters Fetal Brain Development through Interleukin-6. The Journal of Neuroscience, 27 (40), 10695–10702. https://doi.org/10.1523/jneurosci.2178-07.2007