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IDENTIFICATION MARKERS OF CENTRAL NERVOUS SYSTEM DAMAGE SEVERITY OF PERINATAL GENESIS IN CHILDREN

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Abstract

The article is devoted to a topical problem of pediatrics – perinatal damage to the nervous system in young children. The aim of the study was to search clinical and paraclinical markers determining the severity of damage to the nervous system and enabling to timely identify the cohort of children threatened by mental disability. The study outline included clinical assessment of neurological and somatic status of 136 children with perinatal damage to nervous system, aged from 2 to 16 months, as well as neurosonography, magnetic-resonance imaging, global and stimulation electroneuromyography, computer electroencephalography, determining the levels of neurospecific markers of central nervous system damage, i.e. anti-nerve growth factor antibodies, the activity of leukocyte (neutrophil) elastase, myelin basic protein antibodies, protease inhibitor L1. All the children were consulted by an ophthalmologist, orthopedist and defectologist.

The leading factors that cause the perinatal damage to the nervous system are antenatal and intrapartum hypoxia (50%) and fetal infection (27%). The structure of perinatal syndromes has a heterogeneous nature and different incidence rates. The most common syndrome of nervous system perinatal damage recovery period is the syndrome of motor disorders (68.5%), which is in 24% of cases accompanied by a disturbance of psycho-verbal development, in 38% – by dysfunctions of the autonomic nervous system (sleep disturbance, gastrointestinal dysfunction) and in 7% – by paroxysmal syndrome.

According to neuroimaging, the changes in the periventricular region, cavitation and the well-marked cortical/subcortical sub-atrophy are the predictors of unfavorable outcomes. The ophthalmic changes of organic nature may include the presence of pigmented rims and gliosis in fundoscopy, as well as arteriospasms and divergent strabismus. The neuro-immunological indicators are critically important in evaluating the severity of the nervous system damage. The increase of leukocyte elastase indices point out the permeability of the blood-brain barrier and the consequent "launch" of anti-nerve growth factor antibodies promotes to serious disturbances of reparative processes after perinatal heterogeneous stress, thus leading to psycho-neurological disability. Neuro-immunological markers also help to verify the therapeutic efficacy and "separate" the cohort of children born with congenital hereditary diseases of the nervous system. In case of the latter, the presence of severe neurological status is marked with regulatory indices of anti-nerve growth factor antibodies.

Keywords: *perinatal pathology, markers of encephalopathy, fundoscopy, children, neuroimmunology.*

Introduction

Perinatal damage to the nervous system is one of the most topical problems in young children due to the high incidence of this pathology. The implementation and widespread administration of in vitro fertilization, perinatal intensive care advancements as well as expanding the boundaries of perinatal period create preconditions for further growth of this pathology [*Barashnev Yu, 2001; Khachatryan L et al., 2003; Zykov V et al., 2009*]. Perinatal damage to the nervous system is a group of heterogeneous and clinically polymorphic syndromes proceeding from exogenous damage of the nervous system, originated in the perinatal period. In line with WHO recommendations, the perinatal period commences at 22 completed weeks of gestation and ends 7 completed days after birth. The nature and degree of the damage are determined by the volume of clinical manifestations and the prognosis of the disease – from recovery to profound

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disability, and sometimes to death. The most "favorable" is hypoxic damage, the most severe – infectious-toxic damage [*Vatolin K, 1995; Volpe J, 1995; Zykov V et al., 2009; Fox C, Fullerton H, 2010; Palchik A, Shabalov N, 2013*].

After the exposure of the causative factor, the brain develops profound metabolic and vascular disorders, which leads to significant neuronal loss (necrosis). If the compensatory abilities "stop" the disaster in the brain tissue, the recovery process begins. In an unfavorable course, the processes of destruction (suicidality) of neurons with protein-tanatins starts (self-destruction is a genetically determined process) when the damage begins to spread to other zones, involving broader areas of cortical and subcortical structures [*Elkonin B, 2001; Vannucci R et al., 2001; Zhu C et al., 2006; Studenikin V et al., 2008; Petrukhin A, 2009; Kliegman R et al., 2011; Shabalov N, 2011; Ivanova D, 2015*].

Timely and adequate therapeutic measures can "stop" apoptosis, compensate the loss of neurons and switch on the sprouting mechanism (the ability of neurons to give new processes, and thus, a few dozen cells can compensate the functions of thousands). In view of the above, it is crucial to search for predictors allowing to assess the degree of the nervous system damage and the risk of neuropsychiatric disability. Of course, there is a correlation between the degree of medullary substance damage and the final reparation, which ultimately determines the clinical manifestations of perinatal damage. However, clinicians are often faced with the clinical and morphological dissociation when significant structural damage to the brain shown by magnetic-resonance imaging is accompanied by minimal clinical symptoms, and vice versa, minor cavitation can lead to profound neuropsychiatric disability. Thus, it can often be difficult to predict the further development of the child relying on the morphological deficiency.

With regard to the above, it can be concluded, that the search of diagnostic predictors has a crucial importance for assessing the degree of damage to the nervous system and the risk of development of severe motor and mental disorders.

Material and methods

To determine the markers of the nervous system damage severity, 400 children with perinatal damage of nervous system aged from 2 to 16 months underwent examination, from which 136 children were (79 boys and 57 girls) selected for further investigation. The selection criteria were the absence of inflammatory diseases, the availability of complete medical records, and parental consent for the conduction of complex clinical and paraclinical studies. All the patients were observed during the period of more than 5 years for verifying the revealed predictors. The control group consisted of 65 healthy children of the same age.

The study outline included the following tasks: clinical evaluation of neurological and somatic status, neurosonography and magnetic-resonance imaging, global and stimulation electroneuromyography, computer electroencephalography. Laboratory researches allowed determining the levels of neurospecific markers of central nervous system damage, i.e. anti-nerve growth factor antibodies, the activity of leukocyte (neutrophil) elastase, myelin basic protein antibodies, protease inhibitor L1. All the children were consulted by an ophthalmologist, orthopedist and defectologist.

For the convenience of the statistical survey the numerical score of clinical signs was developed, which included the investigation of motor, cognitive, emotional and behavioral spheres (Fig. 1). Each of the 6 sub-spheres was rated as follows: 0 – no disorder, 0.5 – mild disorders, 1 – severe disorders. Thus, the minimum total score was 0 (for the healthy children in the control group), and 6 – for the children with severe organic lesion.

Statistical processing was implemented with the use of Statistica 22.0 software package (Statsoft Inc. USA). Differences were tested using Mann-Whitney test, Fisher's exact test, chi-square, Student's t-test. A two-tailed P-value of less than 0.05 was considered significant. "IBM SPSS Statistics, version 22" and "Microsoft Excel 2010" were used for all statistical analyses.

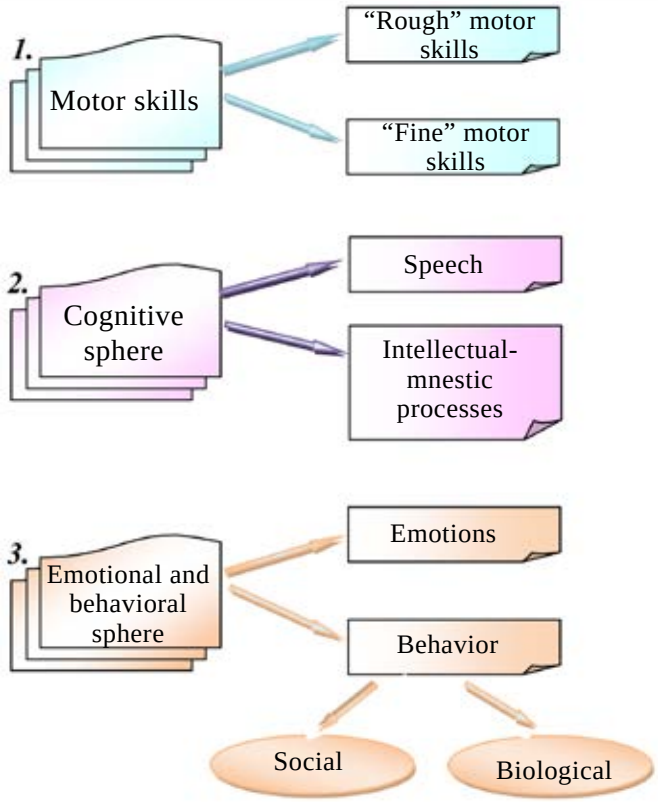

FIGURE 1. Algorithm for evaluation of psychomotor development

RESULTS AND DISCUSSION

According to the results of clinical evaluation, the patients were divided into two groups: I group included patients with the estimation score of 0.5-3 points and II group had an estimation score of 3.5-6 points. The neurological status of I group patients was characterized by delayed development of motor, intellectual, emotional and behavioral skills, and II group patients had a well-marked delay in all studied spheres (Table 1).

According to neuroimaging, I group patients had the following changes: 63% of children had the enlargement of interhemispheric fissure, from which 58% with symptoms of residual ventriculomegaly. In 53% there was a slight sub-atrophy of medullary substance, 26% had cavitation change, 16% had changes in the periventricular region, 47% patients were with thalamic and 16% – with myelination disorders. There were changes in periventricular region in 35.3% and impaired myelination in 38% of the patients of II group ($p<0.05$), which were revealed twice as frequently ($p<0.03$) compared with children of I group. As for other disorders, there were no significant differences between the patients of I and II groups. This was confirmed by the monitoring process which recorded any cases of clinical and morphological picture imbalances (Table 1).

TABLE 1

Identification markers of nervous system damage severity in young children

Characteristics	I group	II group
According to the results of clinical estimation		
Degree of nervous system damage	Mild and moderate	Severe
Clinical estimation (scores)	0.5-3	3.5-6
Anti-nerve growth factor antibodies	norm	↑
Leukocyte elastase levels	↑	↑↑
According to neuroimaging		
Ventriculomegaly	+	+
Enlargement of the interhemispheric fissure	+	++
Sub-atrophy of medulla	+	+
Atrophy of medulla	-	++
Changes in thalamic region	+	+
Changes in periventricular region	±	++
Cavitation changes	±	+
Disorders of myelination	±	+

NOTES: (↑) – increase; (↑↑) – strong increase; (+) – presence of a sign; (-) – absence of a sign; (±) – slight changes; (++) – strong changes

During the global and stimulation myography the patients in both groups recorded changes typical for supra-segmental type of disturbances in motor integration. As compared with the control group there was a significant reduction of the distal latency time ($p<0.02$) and the increase in the amplitude of the M-response in I group reached up to 11.3±0.6 *mV* and in II group – 12.03±0.4 ($p<0.05$ and $p<0.01$ as compared with the control group). The most evident was the difference in the ratios of deviation from the age norm, which was almost twice as high in the children of II group as in I group children and amounted to 24.9±4.1 and 12.4±0.4%, respectively ($p<0.01$ between the groups) (Table 2).

Table 2

Myographic markers of nervous system damage severity in young children

Parameters	I group	II group
Amplitude of the interference curve (*mkV*)	149±6.8	170±12.3
Amplitude of the M-response (*mV*)	1.2 times higher than norm	2 times higher than norm
Speed of impulse by efferent fibers (*m/sec*)	1.3 times lower than norm	1.5 times lower than norm
Coefficient of deviation from the age norm (%)	35.2	46.4
Latency reduction (*m/sec*)	0.24	0.54

Conducted correlation analysis of the indices of stimulation neuromyography and scores of clinical data showed a direct mean correlation ($R=0.34\pm0.007$; $p<0.05$) between the clinical severity and the speed of the efferent fiber impulse along the tibial nerve. This verified the impact of the suprasegmentary structure damage degree in the regulation of motor integration.

The evaluation of ophthalmic status allowed to find partial atrophy of the optic disc in 71% of the patients with organic damage to the nervous system from II group (Table 3).

The refractive disorders in children in both groups did not differ significantly and amounted to 13.2% and 11.8%, respectively. Patients of I group had transient convergent strabismus in 81.6% of cases, and 41% of II group patients had stationary convergent strabismus and 23.5% were patients with steady divergent strabismus (due to the partial atrophy of the optic nerve). The dynamic monitoring revealed that neurological (i.e., transient) strabismus is likely to improve with age while the stationary strabismus tends to develop complications. It's interesting that anisocoria (often unstable) was detected in 63% of patients in I group and in 21% of patients in II group. In 87% of the children in I group and in 53% of patients in II group the optic discs had a light pink coloration; there was a discoloration from temporal sides. The optic discs had a light pink coloration in 87% of the children in I group and in 53% of patients in II group. The dynamic monitoring allowed to reveal the coloration process of optic discs and enabled to judge the pace of myelination. The fundoscopy showed dilatation (74%) and asymmetry (71%) in children of I group. It also revealed essential changes in the artery, such as a narrowing and tortuosity in the children of II group, which certainly indicated severe angiopathy of the retina caused by organic affection of brain.

Table 3

Ophthalmic markers of nervous system damage severity in young children (according to neuroimaging)

Parameters	I group	II group
Sub-atrophy of optic nerve discs	-	+
Transient strabismus	+	+
Constant strabismus	-	+
Anisocoria	+	+
Nystagmus	-	+
Gliosis of vascular funnel	+	+++
Pigmented rim	+	+
Varicose veins	+	±
Arteriospasm	-	+

Notes: *(+) – presence, (-) – absence, (±) – slightly and (+++) – strongly expressed signs*

The basic search of markers that reflect the severity of damage to the nervous system was implemented by laboratory research, such as determining the titer of anti-nerve growth factor antibodies, leukocyte elastase level, myelin basic protein antibodies and anti-protease inhibitor L1 antibodies. A significant increase of leukocyte elastase concentration was observed in blood ($R=0.5$; $p<0.01$) of our patients proceeding from the severity of clinical manifestations. So, the patients of I group had leukocyte elastase activity of 301±17.1 *nmol/min × ml* ($p<0.001$ as compared with the control group). The patients of II group had leukocyte elastase activity of 396±14.2 *nmol/min × ml* ($p<0.002$). The differences in mean values in I and II groups were significant ($p<0.05$) (Fig. 2).

At the same time, the indices of the anti-nerve growth factor antibodies in patients of I group (0.64±0.07; $p<0.02$) varied within normative values (0.58±0.17), whereas the children from II

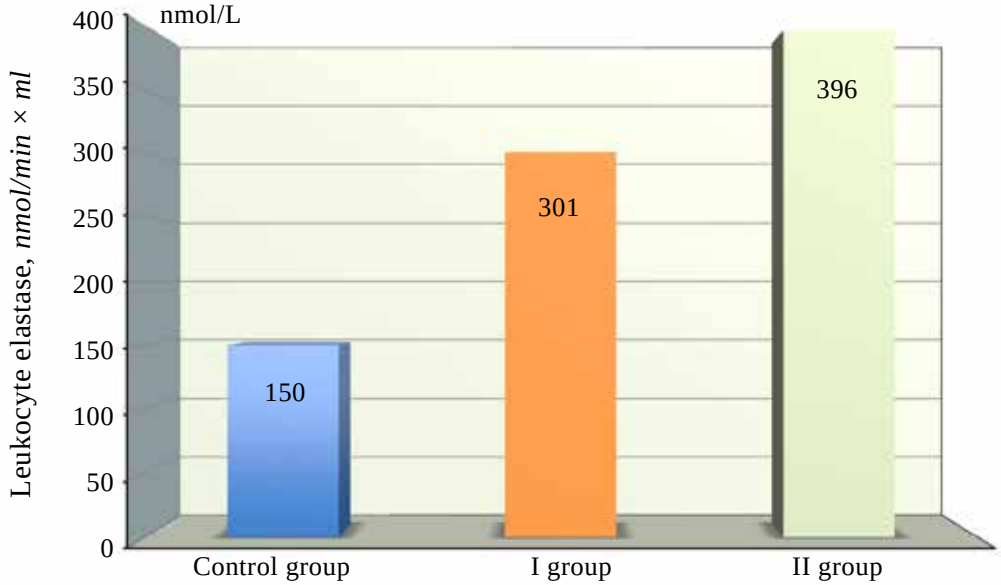

FIGURE 2. *Activity of leukocyte elastase in children with perinatal damage of nervous system*

group (0.92±0.07; p<0.003) had a significant rise in the anti-nerve growth factor antibodies titer, which directly correlated with the clinical severity (R=0.54; p<0.02). With regard to anti-myelin basic protein antibodies, no significant changes were found in titer of the children of I and II groups in contrast to the control group, which can obviously be explained by physiologically incomplete myelination in patients of this age category.

It was curious to detect a high-level protease inhibitor L1 in patients of I and II groups with the absence of significant changes from the norm. Taking into account the protective role of protease inhibitor L1 (inactivator L of blood-brain barrier integrity marker), the obtained results are quite understandable and logical. The level of protease inhibitor-L1 in the children of I group reached 46.3±2.5 *IU/l*, p<0.015, meanwhile, normal indices of the analysis were 29.9±1.2 *IU/L*.

CONCLUSION

The leading factors that cause the perinatal damage to the nervous system are antenatal and intrapartum hypoxia (50%) and fetal infection (27%). The structure of perinatal syndromes has a heterogeneous nature and different incidence rates. The most common syndrome of nervous system perinatal damage recovery period is the syndrome of motor disorders (68.5%), which is in 24% of cases accompanied by a disturbance of psycho-verbal development, in 38% – by dysfunctions of the autonomic nervous system (sleep disturbance, gastrointestinal dysfunction) in 7% – by paroxysmal syndrome. Long-term catamnesis allows to recognize the fact that the character of motor integration disorder is the most important criterion.

Thus, the most favorable type of muscle tonus is dystonia, and the most prognostically unfavorable condition is muscular hypotonia. Children with decreased tonus later developed organic pathology in 39% of cases, and in 61% – functional disorders. Children with dystonia had a recovery in 41% of cases, 3% of the children developed organic changes and 56% had functional changes. Children with 83% hypertonicity had functional disorders, and 17% – organic impairments. According to neuroimaging, the changes in the periventricular region, cavitation and the well-marked cortical/subcortical sub-atrophy are the predictors of unfavorable outcomes. The ophthalmic changes of organic nature may include the presence of pigmented rims and gliosis in fundoscopy, as well as arteriospasms and divergent strabismus.

The neuro-immunological indicators are critically important in evaluating the severity of the nervous system damage. The increase of leukocyte elastase indices point out the permeability of the blood-brain barrier, and the consequent “launch” of anti-nerve growth factor antibodies promotes to serious disturbances of reparative processes after perinatal heterogeneous stress, thus leading to psycho-neurological disability. Neuro-immunological

markers also help to verify the therapeutic efficacy and "separate" the cohort of children born with congenital hereditary diseases of the nervous system. In case of the latter, the presence of severe neurological status is marked with regulatory indices of anti-nerve growth factor antibodies.

So, the markers of organic damage to the central nervous system are:

- Infectious or toxic/metabolic genesis of perinatal damage to nervous system,
- Diffuse muscular hypotonia,
- Damage to the periventricular region and cortical/subcortical brain atrophy,
- Presence of pigmented rims and gliosis in fundoscopy, arteriospasms and divergent strabismus,
- High level of leukocyte elastase and anti-nerve growth factor antibodies against the decreased levels of a protease inhibitor L1.

Thus, the widespread introduction of identification markers of central nervous system damage severity of perinatal genesis into practical medicine will allow to timely identify the children in need of a comprehensive neurological therapy and to prevent the children's disability.

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