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ASSESSMENT OF THE TRACE ELEMENT BLOOD CONDITION IN CHILDREN WITH CONGENITAL DEFORMITIES OF THE THORACIC AND LUMBAR SPINE (Preliminary report)

© *T.V. Lobachevskaya, D.M. Talova, M.V. Sogoyan, A.V. Ovechkina*

The Turner Scientific Research Institute for Children's Orthopedics, Saint Petersburg, Russia

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Introduction. Since the end of the last century, dysfunction of the trace element composition of blood in various forms of scoliosis has been an urgent problem in several studies. Hidden deficiency of trace elements, associated with insufficient food consumption or low absorption in the body, can cause progressive bone deformities. In this context, special importance is attached to trace elements, such as copper, selenium, zinc, boron, manganese, and others. The study of the trace element concentrations in patients with congenital spinal deformities currently is an important and significant task.

Aim. We assess the trace element composition of whole blood in children with congenital deformities of the thoracic and lumbar vertebral columns.

Materials and methods. We analyzed the trace element status of blood in 108 patients (aged 2–16 years) with congenital deformities of the thoracic and lumbar spine (CSD). The congenital vertebral anomalies included disorders of formation, fusion, and/or segmentation of the vertebrae. The control group consisted of 35 healthy children of identical age. Blood ethylenediaminetetraacetic acid (EDTA) was examined using mass spectrometry with inductively coupled plasma (ICP-MS ThermoScientific, iCAP RQ).

Results and discussion. The content of 33 essential and conditionally essential trace elements in the whole blood of patients with CSD was determined. In 37% of patients the zinc, copper, selenium, and chromium levels were decreased compared with the controls. In 7% and 89% of patients the selenium and of chromium levels, respectively, were especially low, below the sensitivity of the device.

Conclusion. The statistically significantly low content of zinc, copper, selenium, and chromium in the whole blood of patients with CSD may have a role in the pathogenesis of the disorders. Further investigations are needed to evaluate their importance as a marker of disease progression.

Keywords: children; congenital deformities; selenium; cupper; zinc; chromium.

ОЦЕНКА МИКРОЭЛЕМЕНТНОГО СОСТАВА КРОВИ У ДЕТЕЙ С ВРОЖДЕННЫМИ ДЕФОРМАЦИЯМИ ГРУДНОГО И ПОЯСНИЧНОГО ОТДЕЛОВ ПОЗВОНОЧНИКА (предварительное сообщение)

© *Т.В. Лобачевская, Д.М. Талова, М.В. Согоян, А.В. Овечкина*

ФГБУ «НИДОИ им. Г.И. Турнера» Минздрава России, Санкт-Петербург

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Введение. Дисфункции микроэлементного состава крови при различных формах сколиоза представляют собой актуальную проблему. Начиная с конца прошлого столетия был опубликован ряд исследований, посвященных этой теме. Скрытый дефицит микроэлементов, обусловленный недостаточным потреблением их с пищей или пониженным всасыванием в организме, может стать одной из причин возникновения прогрессирующих костных деформаций. В данном контексте особое значение придается таким микроэлементам, как медь, селен, цинк, бор, марганец и др. Исследование концентрации микроэлементного состава у пациентов с врожденными деформациями позвоночника (ВДП) в настоящее время является важной и значимой задачей.

Цель данного исследования заключалась в оценке микроэлементного состава цельной крови у детей с ВДП грудной и поясничной локализации.

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Материалы и методы. Проведен анализ микроэлементного статуса крови 108 пациентов в возрасте от двух до 16 лет с врожденными деформациями грудного и поясничного отделов позвоночника на фоне врожденных аномалий развития позвонков: нарушения формирования, слияния и/или сегментации. Контрольную группу составили 35 здоровых детей идентичного возраста. Исследовали кровь, стабилизированную ЭДТА, с использованием масс-спектрометрии с индуктивно связанной плазмой (прибор «ИСП-МС», компании ThermoScientific, iCAP RQ).
Результаты и обсуждение. Было определено содержание 33 эссенциальных и условно эссенциальных микроэлементов в цельной крови больных ВДП. У 37 % пациентов выявлено снижение содержания ряда микроэлементов крови — цинка, меди, селена и хрома по сравнению с контролем. При этом у 7 % пациентов содержание селена и у 89 % содержание хрома было особенно значительно снижено и находилось вне пределов определяемости (чувствительности) прибора.
Заключение. Достоверное снижение в крови больных с ВДП содержания хрома, цинка и селена может вносить свой вклад в патогенез этих пороков развития и требует дальнейшего изучения для оценки их значения как маркера прогрессирования деформации.

Ключевые слова: дети; врожденные деформации позвоночника; цинк (Zn); селен (Se); медь (Cu); хром (Cr); масс-спектрометрия.

Introduction

According to literature data, approximately 50% of congenital malformations of the spine have a progressive course [1], which leads to severe and rigid curvatures of the spinal column at school age, often accompanied by vertebral–medullary conflict [1–3]. One of the methods of preventing the development of neurological deficit and gross congenital spinal deformities (CSD) in pediatric patients is the intrauterine correction of mineral imbalance. Based on the initial clinical examination and radiological examination data among young pediatric patients, predicting the course of CSD during the growth process of the child is extremely difficult.

Many studies have established that supplementation of vitamins, protein, and trace elements [4–6] helps in the growth process and development of bone tissues among young individuals. Trace elements are components of immune complexes, hormones, and enzymes and participate actively in metabolic processes. As they are cofactors of most biochemical reactions in the body, they affect the functional state of various organs and systems, as well as the structure and quality of bone and cartilage tissue [7]. Insufficient intake of trace elements with food or impairment of the absorption of a number of trace elements can be one of the possible causes of progressive deformities of the skeletal system. In this context, special significance is attached to trace elements such as copper, selenium, zinc, boron, and manganese.

In clinical practice, patients with CSD often simultaneously suffer from several chronic diseases that negatively affect bones and cartilage tissues. Diabetes mellitus, celiac disease, rheumatoid arthritis, chronic renal failure, and thyroid, liver, and pancreas diseases represent a noninclusive list of pathologies associated with congenital spinal curvatures [8].

Copper and zinc are cofactors of enzymes responsible for the synthesis of glycosamines and collagen. Zinc is a component of approximately 250 enzymes and is involved in carbohydrate, protein, and fat metabolism. Zinc deficiency leads naturally to anemia, secondary immunodeficiency, sexual dysfunction, and fetal malformations. Zinc in combination with cysteine is especially important for gene expression, as the so-called zinc fingers are the central structure of the DNA-binding domains of the receptors of hormones, vitamin D, estrogen, and progesterone. The level of steroid and peptide hormones, such as cortisol, insulin, and somatotropin (insulin-like growth factor-1), depends on the zinc level in the body. A direct correlation was noted between the absorption of zinc in the intestine in patients with various forms of scoliosis and the serum level of circadian hormone melatonin [4–5]. The bone tissue contains approximately 30% of the zinc reserves of the whole body. Many studies have established that the level of zinc in bone tissue decreases rapidly with insufficient intake or impairment of its uptake [6–10]. The average daily requirement of zinc is 10–15 mg [7].

Copper is a component of enzymes involved in iron metabolism, which, in turn, stimulates the absorption of proteins and carbohydrates. Copper is involved in tissue oxygenation. This trace element

is a cofactor for lysyl oxidase and is required for the intermolecular bond of collagen and elastin. In addition, it is involved in collagen formation, red blood cell synthesis, skin pigment formation, and skeleton mineralization, and it is the main component of the myelin sheath [11, 12]. Clinical manifestations of copper deficiency in the body are represented by impaired skeletal formation and development of connective tissue dysplasia. Affected patients often have an impairment of the cardiovascular system. Copper deficiency inhibits bone growth. The daily demand for copper on average ranges from 0.9 to 3.0 mg/day. Physiologically, children require 0.5–1.0 mg of copper per day [7]. Meanwhile, the trace element selenium is involved in bone tissue formation by activation of calcitonin. On average, children need 20–100 μg of selenium daily [7]. Previous studies have repeatedly mentioned a decreased concentration of selenium in patients with idiopathic forms of scoliosis in both blood serum and hair [8].

In our opinion, the determination of the level of microelements in the whole blood of pediatric patients with a progressive variant of the CSD would reveal a number of criteria typical for this pathological condition at an early age. Russian and foreign studies provide data on estimates of the levels of such trace elements in patients with somatic pathology. To our knowledge, no study has focused on the level of trace elements in patients with congenital skeletal deformities.

This study aimed to assess the compositions of trace elements in the whole blood of pediatric patients with congenital deformities of the thoracic and lumbar spine.

Materials and methods

In this study, 108 patients aged 14 months to 16 years with congenital deformities of the thoracic and lumbar spine, whose diagnosis was confirmed using standard methods of clinical and radiological diagnostics, were examined. In patients with congenital spinal curvature, different variants of vertebral development abnormalities have been identified, such as impaired formation (lateral and posterolateral hemivertebrae, posterior and lateral wedge-shaped vertebrae), impaired fusion (asymmetrical butterfly-like vertebrae), impaired vertebral segmentation (blocking of lateral surfaces and anterior surfaces of vertebral bodies), and synostosis of the ribs. Of the children examined, 32% had isolated malformations of the thoracic or lumbar spine, and 68% had multiple and combined malformations of the corresponding parts of the spinal column. All patients had clinical scoliosis and/or kyphosis of the thoracic and/or lumbar spine and marked asymmetry of the shoulder girdle, waist triangles, and pelvic distortion. In all pediatric patients, congenital spinal curvature progressed with the growth and development process.

The control group consisted of 35 healthy children of identical age. The study was performed with whole blood stabilized with ethylenediaminetetraacetic acid (EDTA) using the most innovative method of analyte quantification, inductively coupled plasma mass spectrometry (ICP-MS instrument, ThermoScientific, iCAP RQ, UK).

The experimental method was as follows: the processed blood sample was supplied using a peristaltic pump to a device nebulizer; it was converted into aerosol through an argon stream. The aerosol causes the dissociation of blood into atoms under high temperature through the central channel of the device. The formed positively charged ions pass through the ion optics system into the device itself, where the ions were filtered by mass to charge (m/z), and the intensity of the ion flux was detected. Thereafter, the spectrometer showed the signal intensity in a given range [11, 13]. After a preliminary long-term preparation of the blood samples, in order to destroy the organic matrix of the examined material and release trace elements from a bound state, the concentration of analytes was detected on an ICP-MS instrument.

The material of each patient was examined in duplicate blood samples. To reduce the likelihood of random errors, each patient sample was subjected to a threefold analysis. The average value of the analyte concentration was then calculated. For each trace element, a calibration graph was preliminarily constructed with the control-standardized solutions.

Statistical analysis of the results was performed using Statistics 6.0 program. The significance of differences between the groups was assessed by non-parametric paired Student's t-test with a two-sided distribution and the determination of the statistical confidence indicator. Indicator differences were considered significant at a significance level of $p < 0.05$.

Results and discussion

In this study, 37% of patients with CSD had a moderate decrease in zinc concentration in whole blood (4.4–7.5 mg/l) (Fig. 1). The reference range of values was estimated according to the data of Aftanas et al. [7, 9]. In 13% of the patients, low concentrations of copper, as well as normal zinc levels, were recorded in the blood. The remaining 87% of patients with CSD had a copper concentration within the reference range of 0.80–1.30 mg/l (Fig. 2). In 29% of patients, selenium levels were lower than the reference values (0.058–0.234 mg/l) (Fig. 3). These patients belonged to the group of pediatric patients with low zinc levels, of which 7% had selenium values below the detectable level (beyond the sensitivity) of the device. In 89% of the patients, the chromium concentration was below the detection threshold of this analyte (0.006–0.045 mg/l) (Fig. 4). The remaining trace elements were within the reference values in both the CSD group and control group. In the control group, indicators of zinc and copper were within the reference range (Table 1). The deviations detected in the study group are graphically displayed in Figs. 1–4.

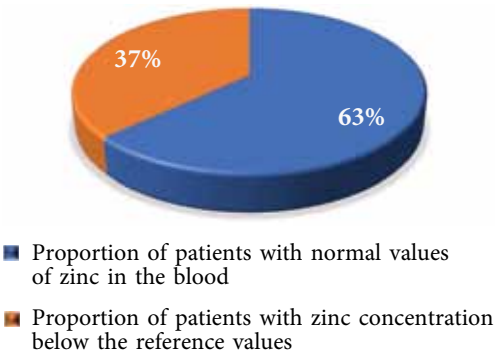

Fig. 1. Distribution of patients based on zinc concentrations inside and outside the reference interval among patients with congenital spinal deformities

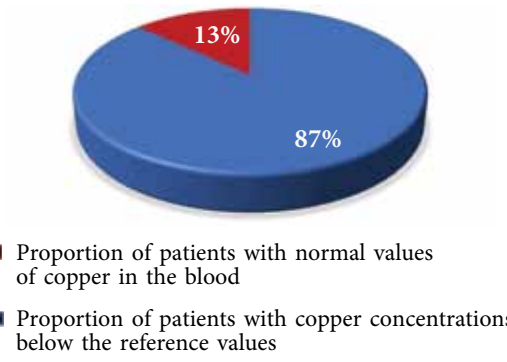

Fig. 2. Distribution of patients based on the copper concentration inside and outside the reference interval in patients with congenital spinal deformities

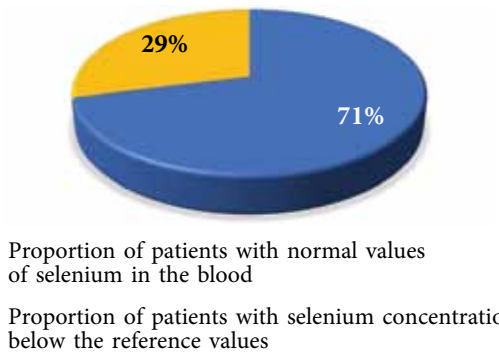

Fig. 3. Distribution of patients based on selenium concentration inside and outside the reference interval in patients with congenital spinal deformities

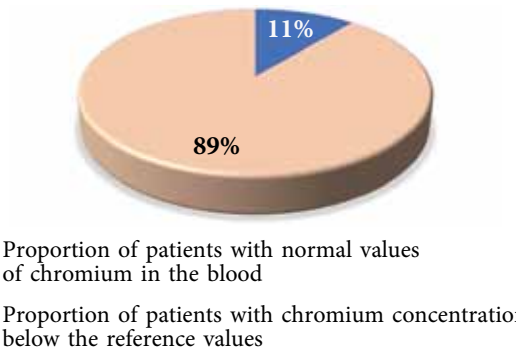

Fig. 4. Distribution of patients based on the chromium concentration inside and outside the reference interval in patients with congenital spinal deformities

Table 1

Average values of trace elements in the CSD group and control group

Trace elements	CSD group	Control group	*p*
Zinc, mg/l	5.5 ± 1.9	6.2 ± 0.9	<0.05
Selenium, mg/l	0.071 ± 0.029	0.15 ± 0.06	<0.01
Copper, mg/l	0.91 ± 0.25	1.14 ± 0.09	<0.1
Chromium, mg/l	<0.005	0.009 ± 0.001	<0.0005

Note. CSD, congenital spinal deformities.

Based on the biochemical study, statistically significant changes were found in the concentrations of trace elements in the EDTA-stabilized blood in patients with CSD.

Conclusion

Based on the analysis results of 33 essential and conditionally essential indicators of trace elements in the whole blood of patients with CSD, reductions in chromium (89%), zinc (37%), and selenium (29%) levels in EDTA-stabilized blood were statistically significant. The concentrations of these trace elements in a significant number of patients with CSD aged 2–16 years were significantly lower than the reference range. These data suggest that the low levels of these trace elements may help in the pathogenesis of CSD development and possibly serve as a marker of the progressive nature of spinal deformity during the growth and development process of the child. Based on the obtained data, developing an algorithm of diagnostic measures to identify the most reliable criteria for predicting the course of congenital deformities of the thoracic and lumbar spine in young pediatric patients with vertebral abnormalities is warranted.

Additional information

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Conflict of interest. The authors declare no obvious and potential conflicts of interest related to the publication of this article.

Ethical review. The study was conducted in accordance with the ethical standards of the Helsinki Declaration of the World Medical Association, as amended by the Ministry of Health of Russia, approved by the ethics committee of the Turner Scientific Research Institute for Children's Orthopedics.

Contribution of authors

T.V. Lobachevskaya performed the development of the research methodology and data processing, wrote all sections of the article, and performed the collection of literature data and their processing.

D.M. Talova was involved in research and data processing.

M.V. Sogoyan was involved in research and data processing.

A.V. Ovechkina edited the article for grammar.

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Information about the authors

Tatiana V. Lobachevskaya — MD, PhD, Assistant Professor, member of AACC, doctor of Clinical Laboratory Diagnostics of the Consultative Diagnostic Center. The Turner Scientific Research Institute for Children's Orthopedics, Saint Petersburg, Russia. https://orcid.org/0000-0002-1158-9573. E-mail: lobachevskayatatiyana@yahoo.com.

Татьяна Владимировна Лобачевская — канд. мед. наук, врач клинической лабораторной диагностики КДЦ ФГБУ «НИДОИ им. Г.И.Турнера» Минздрава России, Санкт-Петербург. https://orcid.org/0000-0002-1158-9573. E-mail: lobachevskayatatiyana@yahoo.com.

Daria M. Talova — MD, doctor of Clinical Laboratory Diagnostics of the Consultative Diagnostic Center. The Turner Scientific Research Institute for Children's Orthopedics, Saint Petersburg, Russia. https://orcid.org/0000-0002-2627-5120. E-mail: dasha_talova@mail.ru.

Дарья Михайловна Талова — врач клинической лабораторной диагностики КДЦ ФГБУ «НИДОИ им. Г.И.Турнера» Минздрава России, Санкт-Петербург. https://orcid.org/0000-0002-2627-5120. E-mail: dasha_talova@mail.ru.

Marina V. Sogoyan — MD, Research Associate of the Genetic Laboratory of the Center for Rare and Hereditary Diseases in Children. The Turner Scientific Research Institute for Children's Orthopedics, Saint Petersburg, Russia. https://orcid.org/0000-0001-5723-8851. E-mail: sogoyanmarina@mail.ru.

Марина Ваниковна Согоян — научный сотрудник генетической лаборатории Центра редких и наследственных заболеваний у детей ФГБУ «НИДОИ им. Г.И. Турнера» Минздрава России, Санкт-Петербург. https://orcid.org/0000-0001-5723-8851. E-mail: sogoyanmarina@mail.ru.

Alla V. Ovechkina — MD, PhD, Associate Professor, Honored Doctor of the Russian Federation, Academic Secretary. The Turner Scientific Research Institute for Children's Orthopedics, Saint Petersburg, Russia. https://orcid.org/0000-0002-3172-0065. E-mail: ovechkina.spb@mail.ru.

Алла Владимировна Овечкина — канд. мед. наук, доцент, заслуженный врач РФ, ученый секретарь ФГБУ «НИДОИ им. Г.И. Турнера» Минздрава России, Санкт-Петербург. https://orcid.org/0000-0002-3172-0065. E-mail: ovechkina.spb@mail.ru.