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International Journal of Biomedicine (ISSN 2158-0510) is published four times a year by International Medical Research and Development Corp. (IMRDC), 6308, 12 Avenue, Brooklyn, NY 11219 USA

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BRIEF REVIEW

Neurology

INTERNATIONAL JOURNAL OF BIOMEDICINE

The Role of Non-Drug Treatment Methods in the Management of Epilepsy

Ekaterina A. Narodova, PhD*; Natalia A. Shnayder, PhD, ScD; Valeria V. Narodova, PhD, ScD; Diana V. Dmitrenko, PhD, ScD; Ivan P. Artyukhov, PhD, ScD

Krasnoyarsk State Medical University named after Prof. V.F. Voino-Yasenetsky
Krasnoyarsk, the Russian Federation

Abstract

This review is dedicated to the issue of non-pharmacological treatment of epilepsy in the adult population in Russia and other countries. A literature review was conducted using international databases for the period between 2010 and 2017. A total of 64 full-text editions were included in this review, which allowed us to reveal the basic non-pharmacological epilepsy treatment options. However, not all of these options have a sufficient evidence base, and some of them are not always safe. Particularly, methods with a low level of evidence include acupuncture and aromatherapy. Further studies are needed to explore the methods aimed at eliminating the dominant epileptic system through the development of a new, more powerful dominant system. One of the methods that can influence the pathogenesis of epilepsy is physical activity for patients with epilepsy, since epileptiform activity is reported to disappear from the EEG during exercises. The positive results of the application of music therapy are also described in the modern literature. Specifically, according to the results of some studies, the positive effect of reducing the frequency of seizures was achieved in epileptic patients who listened to music during sleep over a year. However, these studies are not numerous, so they cannot constitute a high level of evidence. Therefore, care should be exercised in applying these methods in epileptic patients. **(International Journal of Biomedicine. 2018;8(1):9-14.)**

Key Words: epilepsy • adults • non-pharmacological treatment • music therapy

Introduction

According to the world statistics, epilepsy takes the third place for overall morbidity after cardiovascular diseases and diabetes mellitus, and the third place in neurological morbidity.[1] Therefore, epilepsy is a relevant public health problem in Russia and other countries.[2] In recent years, considerable attention has been paid to the development and implementation of medicinal and alternative (non-pharmacological) methods for epilepsy treatment. However, current epilepsy treatment options allow achieving remission or reducing the number of seizures in only 60%-70% of patients.[3] An important problem of epileptology is ensuring the safety and acceptability of the treatment as well as preventing adverse side effects (ASEs) of antiepileptic drugs (AEDs). ASEs can often decrease patients' quality of life, thereby offsetting the positive effect of the treatment. Moreover, such ASEs as depression and anxiety (the fear of the coming seizure) may aggravate epileptic seizures.[4] Some ASEs are associated with the effects of AEDs on the liver enzymes. These effects cause induction or inhibition of the liver enzymes, displacing other AEDs from protein linkages. These reactions increase the rate of metabolism and cause a reduction of the plasma concentration of AEDs, which may lead to difficulties in the choice of AED dosage. On average, the frequency of ASEs and complications of antiepileptic therapy remains high and varies, according to different authors, from 7% to 25%.[5-7]

Therefore, the presence of ASEs requires the immediate withdrawal of AEDs, even if drug-induced epilepsy remission is achieved. It should be noted that 40% of epileptic patients need polytherapy. This leads to an increase in the frequency of ASEs, adverse drug-drug interactions, and teratogenicity.[8] In addition, there are difficulties in assessing the effectiveness of ASEs of a single drug. Drug-drug interactions often decrease the efficiency of antiepileptic treatment and contribute to the

***Corresponding author:** Ekaterina A. Narodova, PhD. Krasnoyarsk State Medical University named after Prof. V.F. Voino-Yasenetsky, Krasnoyarsk, the Russian Federation. E-mail: nataliashnayder@gmail.com*

development of ASEs.[2,9,10] Consequently, non-drug methods of epilepsy treatment should also be used, both as an additional therapy and (in some cases) as the basic therapy (for example, vagus nerve stimulation) (Table 1).

Table 1.

Non-drug methods of epilepsy treatment

Non-invasive Methods	Invasive Methods
Physical Activity	Vagus Nerve Stimulation
Transcranial Magnetic Stimulation	Deep Brain Stimulation
Psychotherapy	Percutaneous Stimulation of the Trigeminal Nerve
Music Therapy	Surgery
Aromatherapy	
Acupuncture	
Referential Bioadaptation	

The aim of this review is to analyze studies dedicated to non-pharmacological treatment of epilepsy in adults.

Materials and Methods

We have analyzed recent full-text publications in such Russian and international databases as E-Library, PubMed, Scopus, Oxford University Press, Springer, Web of Science Core Collection. The analyzed period comprised 8 years (from 2010 to 2017). The search for publications was conducted using the following keywords: "structural epilepsy", "drug-resistance", "epilepsy", "refractory epilepsy", "focal epilepsy", "intractable epilepsy", "drug-resistant focal epilepsy", and "non-drug therapy".

Results and Discussion

Patients with epilepsy experience a range of social restrictions, leading to their external and internal stigmatization. These limitations include employment problems, driving prohibition, and restriction of physical activity. However, it is a well-known fact that physical exercises lead to better functional adaptation.[11]

Patients with epilepsy who are involved in sport can receive the same benefits from physical activity as healthy people, including an increase in performance efficiency and tolerance, weight loss, and normalization of cardiovascular system functioning. Physical activity is also a critical factor in reducing the risks of diabetes, hypertension, coronary heart disease, obesity, and osteoarthritis. As for psychological advantages, the research in this field found that physically active patients have better mental health than those leading a sedentary lifestyle.[12-14]

Physical activity in early age can cause formation of a neuronal reserve, which then will be used during the life course. Consequently, physically active patients have a lower risk of developing the cognitive impairments associated with epilepsy.[15-17]

The preventive and curative effects of physical activity in cases of epilepsy can be achieved in accordance with several principles, including the principles of consistency, regularity, duration, monitoring, and personalization of the training load. Despite this, it is believed that enhanced muscular activity is accompanied with tachypnea (hyperventilation), which can initiate the seizures.

However, some authors claim that physical activity can reduce the likelihood of seizures. Usually, seizures do not occur while running, swimming, ice-skating, skiing, crossing a crowded street, or during sporting events, although this issue is disputable. On the other side, it is reported that seizures often start when patients are relaxed or sleeping. The described data accounts for the development of new dominant excitation areas in the CNS during vigorous exercises. Due to the negative induction, these areas slow down or inhibit activity in the epileptic area, thereby preventing the occurrence of seizures. It is reported that during physical exercises, seizures occur much more rarely than during relaxation.[11] The disappearance of epileptiform activity in many patients' EEG during physical activity supports this theory.[18-21]

Neurostimulation

Vagus nerve stimulation (VNS) is one of the non-drug epileptic treatment methods. The principle of this method is in the chronic electrical stimulation of the left vagus nerve, using an implantable stimulator.[22] The primary candidates for the application of this method are patients with drug-refractory epilepsy (DRE) who cannot get resection surgery. The main contraindications for this method are pregnancy and lactation, cardiac arrhythmia, bronchial asthma, chronic obstructive pulmonary disease, acute peptic and duodenal ulcer, vasovagal syncope and the type 1 diabetes.[23] Against the background of VNS therapy during the period from 3 months to 3 years, a complete cessation of seizures was revealed in 4.8%-17.6% of patients. A decrease in the number of seizures by 50% or more was detected in 27.3%-47% of patients, while a decrease in the number of seizures by less than 50% was detected in 23.5% of patients.[24-26]

Deep Brain Stimulation

Deep brain stimulation is an effective therapeutic method for DRE treatment, especially for temporal lobe epilepsy. Thus, according to a randomized study, assessing the effectiveness of hippocampal stimulation in patients with temporal lobe DRE, a positive effect in the form of complete disappearance of seizures was found in 50% of patients.[27] Other studies have shown that after 11 years of deep brain stimulation, the attacks were not registered for at least 12 months in only 13.8% of the patients.[28]

The principle of this method lies in electrode implantation into certain brain structures (target-structures); these electrodes are supplied with low voltage and high frequency electric current. Due to the impulses generated by the neurostimulator, the selected brain structures change their functions. Thus, this high frequency stimulation of the target-structures reduces the severity of the symptoms and allows reducing the amount of AEDs taken by the patients as well as bringing the patient back into society.[23]

Transcranial Magnetic Stimulation

Low-frequency repetitive transcranial magnetic stimulation (rTMS) leads to a decrease in cerebral cortex neuronal excitability, while high-frequency rTMS increases the excitability.[2] The mechanisms of rTMS are related to its ability to cause long-term effects of postsynaptic inhibition in excitatory neurotransmitter systems, and a reduction in neuronal excitability through inactivation of the voltage-dependent ion channels.[29]

Based on the data published till now and taking all restrictions into account, a group of European experts assigned evidentiary class C (probably effective) to the low-frequency mode of epileptic focus stimulation in cases where its location is in the cortex or in proximity to cortex dysplasia.[30]

Percutaneous trigeminal nerve stimulation

Percutaneous trigeminal nerve stimulation is a minimally invasive method in which the branches of the first trigeminal nerve are exposed to electricity. To implement this method in practice, a special system is used which consists of an external electric impulse generator and electro-conductive plasters. There are few studies that report the use of this method, but most of those that do consider this method to have a positive clinical effect. During preliminary clinical trials, 57% of patients noticed a 50% or more reduction in the number of seizures.

Psychotherapeutic options

Currently, it is the practice to distinguish three fundamental categories of psychotherapeutic techniques used in epileptology: rewards/sanctions, self-control, and neurofeedback. The rewards/sanctions and self-control categories are used for self-induced seizures and for so-called "reflective attacks" as well as for epileptic seizures that are amplified under the influence of emotional factors. Neurofeedback is a non-pharmacological method of epilepsy treatment with objective registration, amplification and feedback of physiological information to the patient. This method is based on the principle of self-identification of one's own EEG data.

According to information from different authors, neurofeedback can lead to a great reduction in the number of seizures in 50% of cases of patients with epileptic risk factors. From this 50%, in 10% of cases, it is possible to completely discontinue AEDs without the reappearance of epileptic seizures for 2-3 years and more, and in the remaining 40%-50% of cases, after the use of the neurofeedback method, it is possible to halve pharmacological treatment.[31]

There are also art-therapy options for epilepsy treatment. For example, there is an actively developing method, based on the creation of therapeutic music, to reduce the number of epileptic seizures. This method is based on the theory that epileptic seizures occur because of abnormal synchronization of the brain's electrical activity, and that the majority of them stop spontaneously. The effect of structured auditory stimuli provides non-invasive galvanic cortex stimulation, which can reduce epileptiform activity.[32] To prove this hypothesis, researchers conducted a randomized study, which explored the effectiveness of music therapy for patients diagnosed with epilepsy.[33] Patients were exposed to Mozart's music every night for one year. This research resulted in a 17% reduction in the number of epileptic seizures. The achieved effect remained stable during the next year.[34,35]

In another randomized study, which studied both children and adult patients with epilepsy, 85% of patients had a positive response to music therapy with an average reduction of the epileptiform activity index by 31% during listening to the music and by 24% afterwards.[36-44]

The methodology of audiogenic stimuli, proposed by Alfred Tomatis, is another art-therapy method, which is close to music therapy.[45] However, in clinical practice, we meet with an increase in the number of epileptic seizures by 50%-60% after the use of this method. As a result, currently, the effectiveness of this method remains debatable.

Aromatherapy can be useful (for achieving a state of relaxation) as a component of epilepsy behavioral treatment. However, its use is more justified for the treatment of conditions accompanying epilepsy, such as anxiety and depression. In the application of aromatherapy for patients with epilepsy, camphora, sage, and rosemary should be avoided because these substances are known to aggravate patients' condition and increase the number of epileptic seizures.[10]

In the Asian-Pacific region, acupuncture is actively used as a non-pharmacological method of epilepsy treatment. There are data on the use of acupuncture for patients with strokes in order to avoid post-stroke epilepsy. S.Weng and colleagues showed that patients with strokes who received acupuncture had significantly less probability of post-stroke epilepsy, compared to those who did not receive such treatment ($P<0.0001$). However, defensive effects associated with acupuncture need further exploration.[46]

Some authors report neuroprotective, anti-inflammatory and neurotrophic effects of acupuncture and electroacupuncture. These effects are explained by the amplification of recurrent inhibition of the brain cortex and hippocampus with the liberation of different neurotransmitters, including gamma-aminobutyric acid and serotonin. However, due to the lack of controlled clinical trials, those methods cannot be recommended as reliably effective and safe in epileptology.[47]

Conclusion

Based on the results of our literature review, it can be stated that an adequate number of studies of the analyzed period are dedicated to non-pharmacological epilepsy treatment. Methods with both proved clinical effectiveness and low reliable treatment options were found in the studied literature. Most of the authors emphasize a positive influence of physical activity on epileptic patients, including prevention of epileptic seizures. Besides, physical activity is reported to have a positive influence on patients' psychic function, preventing cognitive disorders. However, up until now, physical exercises as an additional therapy are not included in any treatment program for patients with epilepsy. The analysis of the literature showed that this lack of inclusion is due to a current concern among neurologists and epileptologists about the

occurrence of epileptic seizures in a state of hyperventilation. Those concerns are not unfounded, because hyperventilation can provoke epileptic seizures in a certain group of patients with epilepsy. As a result, it is reasonable not to ban physical activity for all epileptic patients, but to limit its intensity for the group of patients in whom hyperventilation can provoke epileptic seizures. Meanwhile, the fact is reported that during physical exercising, epileptic patients' EEGs show reduced epileptiform activity. Finally, regarding music therapy, there are studies that suggest it has a positive effect, but the issue is still under-investigated.

All the options for non-pharmacological epilepsy treatment represented in the present review are based on the classical theory of G.N.Kryzhanivsky about the creation and destruction of pathological systems.[48] The author noted that in early stages of the disease, the elimination of the pathological determinant leads to liquidation of the pathological system.

In late stages, the fixation of a pathological system leads to chronization of the pathological process and corresponding neural disorders. The battle with pathological systems, especially with those with complicated and matured forms, is hard and is not always effective. It requires a complex pathogenetic therapy, focused on elimination of the pathological determinant (for example, the elimination of epileptic focus) and normalization of other links of the pathological system. Activation of the anti-epileptic system, amplification of overall control and other genetic mechanisms are important as well. It is also known that there is a constant countdown in living systems, on which homeostasis is based.[49]

According to the theory of V.A.Rudnev,[50] so-called "internal time" is a genetic core of any motor activity, having characteristics of both populations and individuals. Internal time is expressed as an individual rhythm. Many studies explore individual rhythm, its "maturation" in late ontogenesis, as well as its breaking in different cases of neural disorders.[51] Individual rhythm is a reflection of the harmony of the brain's work, and its breaking is a sign of disintegration in the brain's work. Since it is an established fact that in cases of epilepsy a pathological activation of the brain's neurons occurs, which is a stress for the central neural system, it is possible that the occurrence of an epileptic system can change a patient's individual rhythm.

Consequently, research on individual rhythm changes in patients with symptomatic post-surgery epilepsy, and comparison of these changes with the individual rhythm indicators of healthy persons, can help to create a new dominant in the absence of pathological focus and reset the remaining epileptic system links, imposing the mode of operation closest to the physiological one, and activate the anti-epileptic system. There is also a concept that states "seizures lead to seizures." First proposed by doctor William Gowers (1881) and reflecting the concept of epilepsy as a progressing disease,[52] this concept remains relevant. Therefore, taking into account the prevalence rate of epilepsy and lack of desired effects of pharmacological therapy, the development of new non-pharmacological treatment options, dedicated to creation of a new dominant in the human brain to suppress the formation and activity of a pathological epileptic system, becomes relevant.

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 15-19
http://dx.doi.org/10.21103/Article8(1)_OA1

ORIGINAL ARTICLE

Cardiology

INTERNATIONAL JOURNAL OF BIOMEDICINE

Percutaneous Coronary Interventions in Patients with ST-segment Elevation Myocardial Infarction and Totally Occluded Culprit Artery after Pre-hospital Thrombolysis

Ivan Bessonov, MD, PhD*; Vadim A. Kuznetsov, MD, PhD, FESC, FACC; Igor Zyrianov, MD, PhD; Stanislav Sapozhnikov, MD

Tyumen Cardiology Research Center, Tomsk National Research Medical Center of the Russian Academy of Sciences, the Russian Federation

Abstract

The aim of this study was to evaluate in-hospital outcomes of percutaneous coronary intervention (PCI) in patients with acute STEMI with completely occluded culprit artery after pre-hospital thrombolysis (PT).

Methods: Altogether 1,103 consecutive patients with STEMI admitted to the coronary care unit and submitted to PCI from January 2005 to January 2015 were included in the analysis. The clinical and angiographic characteristics, in-hospital outcomes, as well as predictors of no-reflow phenomenon were analyzed.

Results: Altogether 708 patients (64.2%) with a completely occluded culprit artery (initial TIMI flow ≤ 1) were included in the analysis. Overall, 78(11%) patients who received PCI after PT (PT-group) were compared with 630(89%) patients who received primary PCI (PPCI-group). The rates of no-reflow (14.1% vs 6.8%; *P*=0.02) and recurrent MI (5.1% vs 1.3%; *P*=0.03) were significantly higher in the PT-group. The rates of death (5.2% vs 5.1%; *P*=0.61) and stent thrombosis (3.8% vs 1.3%; *P*=0.11), as well as MACE (9.0% vs 7.0%; *P*=0.52), were comparable between the groups. After univariate analysis, several clinical and procedural characteristics were associated with no-reflow, but only PT was associated with recurrent MI (OR=4.20; 95% CI 1.24-14.3; *P*=0.02). After multivariate analysis, PT remained an independent predictor of no-reflow (OR=2.53; 95% CI 1.17-5.46; *P*=0.015)

Conclusion: PCI in patients with STEMI and completely occluded culprit artery after PT was associated with higher levels of no-reflow and recurrent MI. Completely occluded culprit artery after PT was an independent predictor of no-reflow during PCI. **(International Journal of Biomedicine. 2018;8(1):15-19.)**

Key Words: STEMI • percutaneous coronary intervention • pre-hospital thrombolysis • totally occluded culprit artery

Abbreviations

BMI, body mass index; **CAD**, coronary artery disease; **CKD,** chronic kidney disease; **CABG,** coronary artery bypass grafting; **MI**, myocardial infarction; **MACE**, major adverse cardiac events; **PCI**, percutaneous coronary intervention; **PT**, pre-hospital thrombolysis; **STEMI**, ST-segment elevation MI.

Introduction

Despite the reduction in time to reperfusion with pre-hospital thrombolysis (PT), about 40% of patients do not achieve culprit artery patency.[1] In such cases, rescue percutaneous coronary interventions (PCI) reduce the risks of recurrent MI and death compared to conservative therapy.[2,3] Some studies show no differences between rescue and primary PCI in reducing the composite of death, shock, congestive heart failure, or reinfarction.[4,5] However, the results of PCI in patients with a completely occluded culprit artery after PT are still insufficiently explored. We hypothesized that persistent infarct-related artery occlusion after PT can worsen the prognosis. The aim of this study was to evaluate in-hospital outcomes of PCI in patients with acute STEMI with completely occluded culprit artery after PT.

***Corresponding author:** Ivan Bessonov, MD, PhD. Tomsk NRMC of the RAS. Tomsk, Russia. E-mail: IvanBessnv@gmail.com*

Materials and Methods

Study population

Altogether 1,103 consecutive patients with STEMI admitted to the coronary care unit and submitted to PCI at our center from January 2005 to January 2015 were included in the analysis. Some patients underwent ambulance-based, full-dose tenecteplase thrombolysis, and since 2008, PT has been performed. The decision to conduct treatment with PT was taken by an ambulance physician according to the local standard. All patients received aspirin, 300 mg, and a loading dose of the clopidogrel (300 mg or 600 mg) before catheterization. Patients were anticoagulated with unfractionated heparin (100 IU/kg). Coronary angiography and PCI were performed from either the femoral or radial arterial access, according to the operator's decision. Patients were managed with standard post-PCI care and appropriate secondary prevention (aspirin, clopidogrel, β-blockers, angiotensin-converting-enzyme inhibitors and statins) was commenced unless otherwise contraindicated.

Clinical and procedural data

Clinical data were obtained from the local hospital database, containing information on each PCI in STEMI patients, including demographics, baseline clinical and procedural details as well as in-hospital outcomes. All angiograms of the included patients were reviewed. An occluded culprit artery was defined as a culprit vessel with TIMI 0 to 1 flow, indicating no dye penetration or minimal dye penetration without complete vessel opacification. In patients after PT, non-invasive markers of reperfusion were evaluated and documented within 90 min after thrombolytic administration, according to hospital standard. In cases when ST resolution was partial or absent along with prolonged chest pain and absent of reperfusion arrhythmias, rescue PCIs were performed. The demographic, clinical, angiographic and in-hospital outcomes were analyzed. The composite of in-hospital death, recurrent MI and stent thrombosis were defined as MACE. Recurrent MI was defined as acute MI that occurs following the initial MI within hospitalization. Periprocedural MI was considered as a part of recurrent MI.

Statistical analysis

All analyses were performed with SPSS v21.0 (SPSS Inc., Chicago, Illinois, USA). Continuous variables are presented as mean ±SD and as medians (interquartile ranges). Continuous variables were compared by use of Student's unpaired t-test, or Mann-Whitney U test when data were not normally distributed. Categorical variables were expressed as numbers (percentages) and compared by Chi-squared test or Fisher's exact. To determine relationship between PT with no-reflow and recurrent MI univariate analysis was performed. To assess the impact of PT on no-reflow, multivariate logistic analysis was used. Variables reaching significance, or tendency to significance, on univariate analysis ($p \leq 0.1$) were subsequently incorporated into a multivariate model. Independent variables are presented as odds ratios (OR) with 95% confidence intervals (CI). P-values <0.05 were considered statistically significant.

Results

Out of 1,103 patients who underwent PCI for STEMI, 207(18.8%) received PT and 395(35.8%) were excluded—381 because initial TIMI flow ≥2 and 14 for failure of guidewire lesion crossing or stent delivery. The remaining 708 patients were included in the final analysis. Overall, 78(11%) patients who received PCI after PT (PT-group) were compared with 630(89%) patients who received primary PCI (PPCI-group).

Clinical and procedural data

The characteristics of both patient groups are presented in Table 1. There were no differences in age, BMI, hyperlipidemia, incidence of diabetes, hypertension or CKD.

Table 1.

Baseline patient characteristics

Variable		PT-group	PPCI-group	*P*-value
Age, years		58.3±10.6	59.0±11.6	0.49
Males, gender, n (%)		63 (80.8%)	451 (71.6%)	0.09
BMI, kg/m^2		29.3±4.2	28.9±4.9	0.26
Current smoker, n (%)		38 (48.7)	243 (38.6)	0.08
Hyperlipidemia, n (%)		66 (84.6)	555 (88.1)	0.37
Diabetes mellitus, n (%)		10 (12.8)	110 (17.5)	0.30
Hypertension, n (%)		58 (74.4)	505 (80.2)	0.23
Previous CAD, n (%)		25 (32.1)	229 (36.3)	0.46
Previous MI, n (%)		18 (23.1)	99 (15.7)	0.10
Previous PCI, n (%)		10 (12.8)	58 (9.2)	0.31
Previous CABG, n (%)		1 (1.3)	2 (0.3)	0.30
Previous stroke, n (%)		2 (2.6)	40 (6.3)	0.14
CKD, n (%)		4 (5.1)	39 (6.2)	0.48
Glycemia on admission, (mmol/l)		8.6±3.0	8.5±3.6	0.19
Killip class on admission, n (%)	I	66 (84.6)	576 (91.4)	0.25
	II	7 (9.0)	32 (5.1)	
	III	1 (1.3)	3 (0.5)	
	IV	4 (5.1)	19 (3)	

Angiographic and procedural data are summarized in Table 2. There were no differences between the groups with regard to infarct-related arteries, number of stents implanted, incidence of multivessel disease, use of thrombus aspiration, predilation and a transradial approach. Glycoprotein IIb/IIIa inhibitors were used more often in the PT-group. In the PT-group, pain-to-thrombolysis time was shorter than pain-to-door time in the PPCI-group. The median time from receiving thrombolysis to PCI was 220 (105-480) minutes. The rate of no-reflow was higher in the PT-group.

For PT-group, non-invasive markers of reperfusion were analyzed (Table 3). The absence of post-thrombotic resolution of ST elevation was detected in most patients. One-third of patients had prolonged chest pain and reperfusion arrhythmias.

Table 2.

Angiographic and procedural outcomes

Variable	PT-group	PPCI-group	*P*-value
Infarct artery:			
Left anterior descending, n (%)	34 (43.6)	279 (44.3)	0.91
Left circumflex, n (%)	85 (13.5)	8 (10.3)	0.43
Right, n (%)	240 (38,1)	33 (42.3)	0.47
Left main, n (%)	5 (0.8)	1 (1.3)	0.51
Multi-vessel disease, n (%)	26 (33.3)	183 (29)	0.43
Number of stents implanted, n (%)	1.0±0.5	1.1±0.5	0.08
Predilation, n (%)	46 (59)	404 (64.1)	0.37
Drug-eluting stent, n (%)	22 (31.4)	161 (27.5)	0.49
Transradial approach, n (%)	43 (55.1)	330 (52.4)	0.65
MT, n (%)	4 (5.1)	43 (6.8)	0.57
GPI, n (%)	3 (3.8)	4 (0.6)	0.03
Pain-to-door/pain-to-thrombolysis time*, min	80 [50;225]	120 [64;235]	0.03
No-reflow, n (%)	11 (14.1)	43 (6.8)	0.02

MT - manual thrombus aspiration; GPI - glycoprotein IIb/IIIa inhibitor;-Pain-to-door time calculated for PPCI-group, pain-to-thrombolysis time calculated for PT-group.*

Table 3.

Non-invasive markers of reperfusion in PT-group

Variable		n (%)
Relative ST-segment resolution at 90 minutes	Complete (>70%)	6 (7.7)
	Partial (30%–70%)	17 (21.8)
	Absent (<30%)	55 (70.5)
Chest pain	Prolonged	28 (35.9)
	Absent	50 (64.1)
Reperfusion arrhythmias	AIVR	1 (1.3)
	VPB	19 (24.4)
	VF	5 (6.4)
	Absent	53 (67.9)

AIVR - accelerated idioventricular rhythm; VPB - ventricular premature beats; VF - ventricular fibrillation.

In-hospital clinical outcomes

The rate of recurrent MI was significantly higher in the PT-group (Table 4). All recurrent MIs were due to stent thrombosis in the PPCI-group. In the PT-group, one patient had a recurrent MI without stent thrombosis. A recurrent MI in this case was diagnosed by the presence of typical symptoms of acute myocardial ischemia and a fivefold re-rise of troponin T. The rates of death and stent thrombosis, as well as MACE, were comparable between the groups. After univariate analysis, several clinical and procedural characteristics were associated with no-reflow, but only PT was associated with recurrent MIs (Table 5). After multivariate analysis (Fig.1), PT remained an independent predictor of no-reflow (OR=2.53; 95% CI 1.17-5.46), along with age, body mass index, previous coronary artery disease, glycemia on admission, cardinogenic shock, left main PCI, multivessel disease and pre-dilation.

Table 4.

In-hospital clinical outcomes

Variable	PT-group	PPCI-group	*P*-value
Death, n (%)	4 (5.1)	33 (5.2)	0.61
Recurrent MI, n (%)	4 (5.1)	8 (1.3)	0.03
Stent thrombosis, n (%)	3 (3.8)	8 (1.3)	0.11
MACE, n (%)	7 (9.0)	44 (7.0)	0.52

Table 5.

Univariate logistic analysis for no-reflow and recurrent MI

Variable	No-reflow			Recurrent MI		
	OR	95% CI	*P*	OR	95% CI	*P*
Age (years)	1.04	1.01-1.06	0.05	1.00	0.95-1.05	0.95
Male sex	1.02	0.55-1.90	0.95	0.53	0.11-2.42	0.41
BMI	1.07	1.01-1.12	0.02	0.90	0.78-1.03	0.12
Current smoker	0.68	0.37-1.23	0.20	1.53	0.49-4.80	0.47
Hyperlipidemia	0.68	0.32-1.43	0.47	0.70	1.15-3.23	0.64
Diabetes mellitus	1.62	0.84-3.13	0.15	0.98	0.21-4.53	0.98
Hypertension	1.31	0.63-2.75	0.47	0.77	0.21-2.88	0.70
Previous CAD	2.21	1.27-3.87	0.01	1.28	0.40-4.08	0.67
Previous MI	1.50	0.76-2.93	0.24	2.58	0.76-8.71	0.13
Previous PCI	0.74	0.26-2.11	0.57	0.85	0.12-6.71	0.88
CKD	2.56	1.08-6.06	0.03	3.20	0.69-15.06	0.14
Glycemia on admission	1.15	1.08-1.22	<0.001	0.96	0.79-1.16	0.64
Pain-to-door/ pain-to-thrombo-lysis time*	1.00	0.99-1.01	0.76	0.99	0.94-1.03	0.64
Cardiogenic shock	7.41	2.99-18.38	<0.001	2.79	0.34-22.53	0.34
Left anterior descending PCI	1.01	0.58-1.77	0.97	1.79	0.56-5.68	0.33
Left circumflex PCI	0.66	0.26-1.69	0.38	0.60	0.08-4.68	0.62
Right PCI	0.86	0.48-1.53	0.60	1.14	0.36-3.63	0.82
Left main PCI	12.77	2.51-64.86	0.03	-		
Multi-vessel disease	2.03	1.16-3.56	0.02	1.72	0.54-5.49	0.36
Pre-dilation	3.07	1.48-6.40	0.001	1.74	0.47-6.47	0.41
PT	2.24	1.10-4.55	0.03	4.20	1.24-14.30	0.02

**-Pain-to-door time calculated for PPCI-group, pain-to-thrombolysis time calculated for PT-group.*

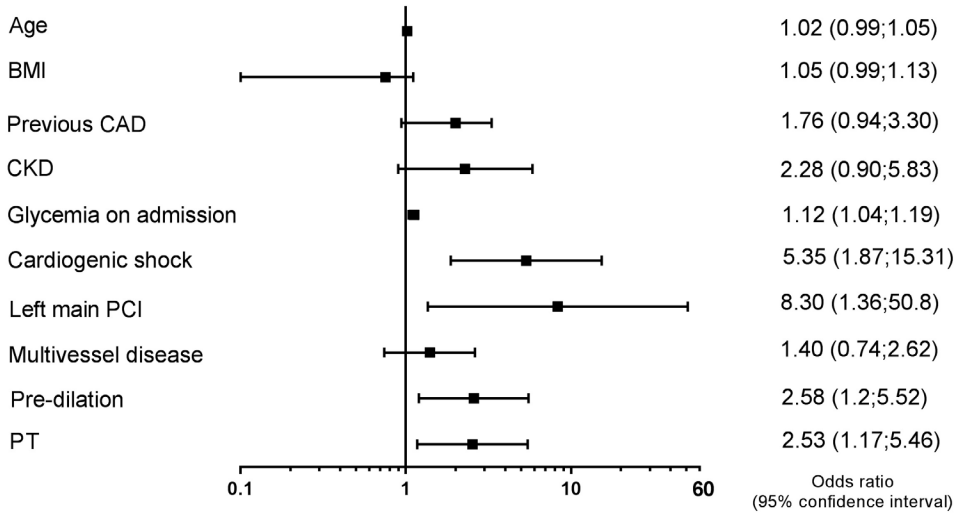

Fig. 1. *Multivariate logistic analysis for no-reflow.*

Discussion

Recent studies have confirmed that a pharmacoinvasive strategy in patients with STEMI may be comparable to PPCI. In a CAPTIM trial, pre-hospital fibrinolysis followed by early transfer to PCI led to 5-year survival rates similar to those of PPCI.[6] In the 5-year analysis of the French Registry of Acute Coronary Syndrome (FAST-MI-2005), survival with a pharmacoinvasive strategy was comparable to survival with PPCI.[7] In the registry from Belgium, the results were similar for in-hospital mortality and 1-year survival.[8] In the STREAM study, the 30-day primary composite outcome (death, shock, congestive heart failure, or recurrent MI) was similar in the PPCI and pharmacoinvasive groups.[4] A combined analysis of the CAPTIM and WEST trials shows the advantages of the pharmacoinvasive strategy for 1-year outcomes.[9] In analysis of the Mayo Clinic STEMI database, fibrinolysis followed by transfer for PCI was found to be a reasonable alternative when PPCI is not readily available.[10] But in all of these studies, cases with successful and failed thrombolysis were not analyzed separately. A pharmacoinvasive approach can be more effective than PPCI in cases when culprit artery patency is achieved. In this situation, pharmacoinvasive strategy can reduce the reperfusion time, which is especially important for patients presenting early after the onset of symptoms.[11,12] But the incidence of complete culprit artery patency (TIMI grade 3 flow) occurred in only 55% to 60% of patients treated with PT.[1] In our study, patients with a totally occluded culprit artery after PT had higher rates of recurrent MI and incidence of no-reflow during PCI. Potentially all of these patients could receive a primary PCI at an acceptable time. Thrombolysis in these cases increased the time to PCI.

According to our data, unsuccessful PT was an independent predictor of no-reflow during PCI. Negative effects of the no-reflow phenomenon on clinical outcomes in patients with STEMI were demonstrated in several studies. No-reflow was associated with higher mortality, poor healing of the infarct zone, and adverse remodeling with an increase in left ventricular failure.[13] The pathological mechanisms of no-reflow include injury related to ischemia, reperfusion, endothelial dysfunction, and distal thromboembolism.[14] Probably as a result of unsuccessful thrombolysis, components of the thrombus and atherosclerotic materials can produce small distal emboli, causing a further reduction in the coronary flow during PCI.

Several studies and meta-analyses have shown that early, routine, post-thrombolysis angiography with subsequent PCI can improve results, compared to a strategy of performing angiography and revascularization only in patients without ST-segment resolution or ongoing chest pain.[15-17] Our data confirm this statement. After PT, 8% of patients with persistent culprit artery occlusion had complete ST-segment resolution and 22% had partial ST-segment resolution. Moreover, most of the patients had no ongoing chest pain. An early routine post-thrombolysis strategy can be particularly effective in these cases.

Our study has several limitations. Firstly, this study was retrospective, nonrandomized and had the potential of selection bias. The number of patients in the PT-group was significantly lower than in the PPCI-group. Moreover, the number of events of recurrent MI and no-reflow were rather small. The multivariable models are not particularly strong with such a limited number of events. The study duration was over 10 years, and practice could change during this time. In addition, the long-term results were not assessed in this study.

In conclusion, PCI in patients with STEMI and completely occluded culprit artery after PT was associated with higher levels of no-reflow and recurrent MI. Completely occluded culprit artery after PT was an independent predictor of no-reflow during PCI.

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 20-25
http://dx.doi.org/10.21103/Article8(1)_OA2

ORIGINAL ARTICLE

Cardiology

INTERNATIONAL JOURNAL OF BIOMEDICINE

One-Year Results of the Use of Absorb Bioresorbable Vascular Scaffold in Patients with Different Forms of Coronary Artery Disease as Compared to a Drug-Eluting Stent

Nabijon P. Yuldashev, PhD[1*]; Robert J. Gil, PhD[2,3]; Ravshanbek D. Kurbanov, PhD, ScD[1]

[1]*Republican Specialized Center of Cardiology, Tashkent, Uzbekistan*
[2]*Central Clinical Hospital of the Internal Affairs and Administration Ministry, Warsaw, Poland*
[3]*Mossakowski Medical Research Centre, Polish Academy of Science, Warsaw, Poland*

Abstract

The aim of this study was to evaluate the immediate and long-term (12 months) clinical and angiographic efficacy of myocardial revascularization using Absorb GT1 Bioresorbable Vascular Scaffold (BVS) in comparison to second-generation drug-eluting stent (DES) in patients with various forms of coronary artery disease (CAD).

Material and Methods: The study included 152 patients with CAD. There were 131 men and 32 women with an average age of 54.6±10.4 years. Patients' data were evaluated retrospectively from the medical records.

Results:

• Implantation of BVS in patients with different forms of CAD did not cause any angiographic or clinical complications, either at the hospital or at 12-month observation stages, and the results were comparable to those of the DES group.

• The technique of implanting BVS and the reception of dual antiplatelet therapy are the key factors for achieving positive results in real clinical practice.

• The use of BVS-frameworks contributes to improving clinical, functional and laboratory indicators, while the observed positive dynamics are comparable to similar data of the DES group.

• Regardless of the type of implanted stents, the survival rate among CAD patients within 12 months after stenting was 100%, while none of the respondents during this time developed acute MI or recurrence of angina attacks. **(International Journal of Biomedicine. 2018;8(1):20-25.)**

Key Words: Absorb bioresorbable vascular scaffold • drug-eluting stent • coronary artery disease • dual antiplatelet therapy

Abbreviations

CAG, coronary angiography; **CAD**, coronary artery disease; **Ch**, total cholesterol; **DAT**, dual antiplatelet therapy; **DES**, drug-eluting stent; **FC**, functional class; **HDL**, high-density lipoprotein; **LDL**, low-density lipoprotein; **LVEDV**, left ventricular end-diastolic volume; **LVESV**, left ventricular end-systolic volume; **LVEF**, left ventricular **ejection fraction; MI**, myocardial infarction; **MACE**, major adverse cardiac events; **PCI**, percutaneous coronary interventions; **PTCA**, percutaneous transluminal coronary angioplasty; **TBA**, transluminal balloon angioplasty; **TG**, triglyceride; **VLDL**, very low-density lipoprotein.

Introduction

In recent years, in some developed countries, there has been a trend towards a reduction in mortality from CAD. This is due to the improvement of preventive measures for CAD, as well as the wide introduction of new effective methods for its diagnosis and treatment. In the USA, where about 4 million people suffer from CAD, despite an improvement in survival rates, more than 650,000 die each year. According to a prognosis by American scientists, by 2020 cardiovascular diseases will account for about 36% of deaths.[1] Thus, the fight against CAD is one of the primary tasks of the medicine in the twenty-first century.

In the last century, one of the most important achievements of cardiology was the development by Andreas Grüntzig

***Corresponding author:** Nabijon P. Yuldashev, PhD. Republican Specialized Center of Cardiology, Tashkent, Uzbekistan. E-mail: nabi1975@mail.ru*

of the technique of TBA,[2] which allows the elimination of stenotic lesions of the coronary artery with minimal surgical trauma during endovascular intervention. Practical interest in interventional cardiology has increased significantly with the advent of coronary prostheses—stents. The use of the stenting technique with unsatisfactory angiographic results of the TBA procedure (residual stenosis or coronary artery dissection) significantly reduced the incidence of complications from endovascular treatment of CAD.[3]

Currently, stenting of coronary arteries has a dominant position among other endovascular methods for CAD treatment. The introduction of the stenting procedure significantly improved not only the immediate but also the long-term (half-year) results of endovascular treatment of CAD in comparison with TBA.[4] At the beginning of the century, stents with an antiproliferative coating were developed and introduced into clinical practice. However, despite many achievements in this area, a number of problems remain: development of neointimal hyperplasia inside the stent, stent thrombosis, and neoatherosclerosis followed by the development of late and very late thrombosis.[5,6]

In this regard, the prospect of using a temporary vascular stent has become very important. However, on March 18, 2017, the FDA issued a letter to health care providers treating patients with Absorb GT1 Bioresorbable Vascular Scaffold (BVS) that there is an increased rate of MACE observed in patients receiving the BVS, when compared to patients treated with the approved metallic XIENCE DES.[7] On October 31, 2017, the FDA issued an update to the March 18, 2017 letter to health care providers to inform the health care community that interim study results through three years from the pivotal clinical trial (ABSORB III) continue to show an increased rate of MACE and BVS scaffold thrombosis in patients receiving the Absorb GT1 Bioresorbable Vascular Scaffold (BVS), when compared to patients treated with the approved metallic XIENCE drug-eluting stent.[8]

The above-mentioned data, as well as the experience of our clinic in the endovascular treatment of CAD, determined the purpose of this study: to evaluate the immediate and long-term (12 months) clinical and angiographic efficacy of myocardial revascularization using Absorb GT1 BVS in comparison to second-generation DES in patients with various forms of CAD.

Materials and Methods

In our clinic, 152 patients with CAD were examined. There were 131 men and 32 women with an average age of 54.6±10.4 years.

Patients' data were evaluated retrospectively from the medical records:

- General clinical and laboratory blood tests, including blood levels of Ch, TG, HDL, VLDL, and LDL
- ECG in 12 standard leads
- Echocardiography with determination of LVEDV, LVESV, and LVEF
- CAG was performed using Phillip Allura CV20 (Phillips Medical Systems, The Netherlands)
- Invasive intervention was performed by right transradial access according to L.Campeau.[9] Selective catheterization of the coronary arteries was performed with Judkins and Amplatz coronary catheters. The contrast agent "Unigexol-350" ("Unique", India) was used. All information was stored on the hard disk of the computer system with the subsequent export of information for processing and storage in the Xcellera system of the local computer network of the catheterization laboratory.
- In collegial analysis of CAG data, we determined the type of coronary blood supply and noted the number of affected coronary arteries, localization, and type of stenotic narrowing. To assess the risk of PCI, each SYNTAX score was calculated on an individual basis using an online calculator.
- The Quality of Life of patients was assessed using the SF-36 questionnaire.
- The condition of patients in the dynamics was assessed 3, 6, 12 and 24 months after CAG; some patients repeated CAG with a pharmacological test after 12 and 24 months.

Depending on the type of implants, two groups of patients were formed. Group 1 included 78 patients with implanted BVS; Group 2 included 74 patients with DES stents.

BVS implantation was performed according to the following set of rules:

1) Prepare the site of the lesion for pre-dilatation with a balloon in a ratio of 1:1 to the diameter of the vessel.
2) Correctly choose the diameter of the scaffold to avoid its overstretch in the vessel.
3) Take into account disclosure limits.
4) Perform post-dilation with a high-pressure balloon catheter.
5) Use DAT.

In this review, we present one-year results of the study.

The inclusion criteria:

- CAD determined by the results of the clinical and instrumental examination. Written informed consent obtained from each patient.
- Documented painless ischemia, stable and unstable angina, acute MI and MI history
- De novo native coronary artery lesions
- Coronary artery lesions, allowing a stent to be implanted
- Hemodynamically significant coronary artery stenosis (≥70%)

The exclusion criteria:

- Previous PCI with stenting or aorto-coronary artery bypass graft in history
- Severe concomitant pathology of the cardiovascular system (aortic aneurysm, valvular pathology requiring surgical correction, severe left ventricular systolic dysfunction (EFLV<35%), decompensated heart failure).
- Intolerance to anticoagulants / disaggregants.
- Bifurcation lesions or lesion of the coronary artery trunk
- Cardiogenic shock.
- Moderate and high risk PCI on SYNTAX score

The primary endpoints:

1) Sudden cardiac death
2) MI associated with the target artery
3) Revascularization of the target vessel according to clinical indications

4) Recurrent/progressive angina
5) Stent thrombosis (definite, probable, or possible)

The statistical analysis was performed using the statistical software «Statistica» (v6.0, StatSoft, USA). To determine whether the compared groups of patients share the same distribution of a single categorical variable, the "test of homogeneity" was applied. Baseline characteristics were summarized as frequencies and percentages for categorical variables and as mean±SD for continuous variables. Analysis of the distribution of values obtained was performed using the Kolmogorov-Smirnov test. Student's unpaired t-test was used to compare two groups for data with normal distribution. Differences of continuous variables departing from the normal distribution were tested by the Mann-Whitney U-test. Group comparisons with respect to categorical variables are performed using chi-square tests. Pearson's correlation coefficient (r) was used to determine the strength of the relationship between the two continuous variables. A probability value of $P<0.05$ was considered statistically significant.

Results

At the initial stage of the study, the groups were comparable in anthropometric, clinico-laboratory and functional parameters, as well as in the nosological structure. However, according to the manufacturer's recommendations for the installation of BVS-frames, mainly for young patients, our emphasis was placed on the age of the respondents. In this connection, the only difference between the groups was in the ages of the participants (Table 1, Figures 1 and 2).

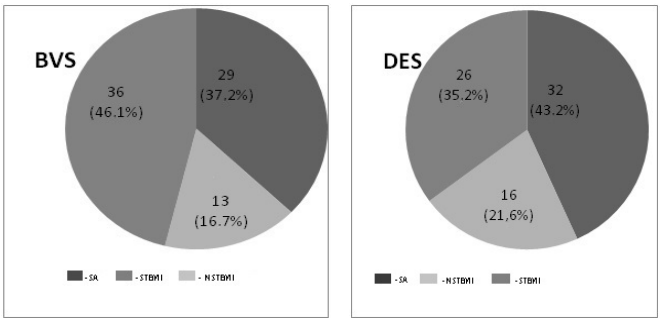

Fig.1. Nosological structure of the compared groups of patients. *SA - stable angina pectoris; (NSTEMI- non–ST-segment elevation MI; STEMI -ST-segment elevation MI.*

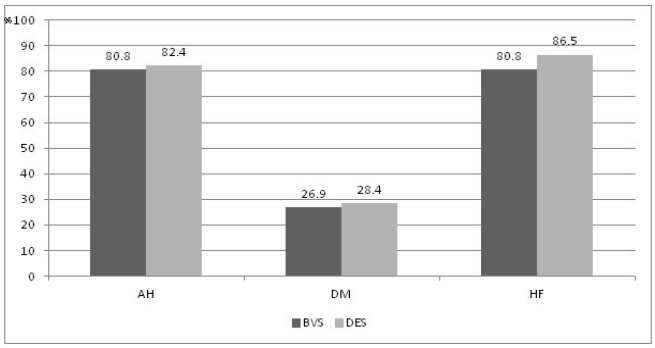

Fig. 2. *Comorbid conditions.*
AH - arterial hypertension; DM - diabetes mellitus type 2; HF - chronic heart failure. The data is presented as a percentage.

Table 1.

Anthropometric, clinico-laboratory and functional characteristics of the patients of the compared groups at the initial stage of the study

Variable		Group 1	Group 2
Age. years		51.83±10.59	57.74±9.33*
Gender	male	69 (88.5%)	62 (83.8%)
	female	9 (11.5%)	12 (16.2%)
Height, cm		1.71±0.07	1.70±0.07
Weight, kg		87.04±13.09	87.49±14.03
BMI, kg/m^2		29.76±3.84	30.33±4.43
Hb, g/l		128.2±14.0	129.3±17.4
Ch, mg/dl		192.6±59.0	180.7±45.1
TG, mg/dl		227.1±160.3	195.9±123.4
HDL, mg/dl		35.6±8.6	35.9±6.5
VLDL, mg/dl		46.5±33.3	39.2±24.7
LDL, mg/dl		110.1±46.8	105.6±35.9
Glucose, mmol/l		6.1±1.9	6.6±2.4
CRP, g/l		19.1±29.1	23.2±48.5
LVEDV, ml		150.7±33.9	150.1±39.7
LVESV, ml		64.7±27.4	65.8±26.8
IST, mm		10.4±1.4	11.3±2.4
LVPWT, mm		10.1±1.2	11.3±1.4
LVEF, %		58.2±10.2	57.3±7.7

**- P=0.000; BMI - body mass index; CRP - C-reactive protein; Hb – hemoglobin; IST- interventricular septum thickness; LVPWT – left ventricular posterior wall thickness.*

Angiography between the compared groups of patients also showed no statistically significant differences, except for the mean diameter of the artery, which was larger in the BVS group (Table 2).

Table 3 presents information in the form of quantitative characteristics for implantation techniques. In Group 1, the number of affected segments of the coronary bed was 110, in Group 2 - 105. In groups 1 and 2, the number of implants was 108 (1.38 per patient) and 98 (1.32 per patient), respectively. By the time of exposure of stents, a significant difference was found: average exposure was greater in Group 1. Immediate, good angiographic success was noted in 100% of cases in both groups of patients; in all patients we were able to recanalize the occluded coronary artery segment. There were no complications in the form of dissection, the phenomenon of "noreflow," acute thrombosis in the stent, coronary artery perforation, etc. The success rate of the procedure also amounted to 100% in both compared groups.

In Group 1, good clinical success was identified in 94.8% of cases, and 4 patients had angina attacks at the level of FC I. In Group 2, clinical success was noted in 95.9% of cases, and 3 patients had angina attacks at the level of FC I-II. There were no lethal outcomes in either group. Thus, direct clinical and angiographic results, regardless of the type of implanted stents, did not have complications such as MACE.

The next stage of the study was the assessment of clinical, laboratory and functional characteristics of patients 1 year after PCI.

Table 2.

Angiographic characteristics of the compared groups of patients

CAG		Group 1	Group 2
Mean SYNTAX score		9.77±4.64	10.36±4.92
The average number of vascular lesions		1.46±0.64	1.49±0.82
1-vascular lesion, n (%)		46 (59.0%)	43 (58.1%)
2-vascular lesions, n (%)		32 (41.0%)	31 (41.9%)
Total number of implants		108	98
The average number of implants per patient		1.38	1.32
Mean lesion length, mm		30.27±17.19	31.39±13.40
ADA	n (%)	68 (87.2%)	61 (82.4%)
	Mean% of stenosis	89.17±9.43	88.46±8.49
	Average diameter, mm	3.33±0.44	3.46±0.35
	Stenosis type A	5	4
	Stenosis type B	26	18
	Stenosis type C	39	43
	The defeat of the p/3	52	41
	The defeat of the m/3	22	30
	The defeat of the d/3	3	2
CA	n (%)	10 (12.8%)	10 (13.5%)
	Mean % of stenosis	82.50±15.17	84.40±14.86
	Average diameter, mm	3.35±0.54	3.15±0.49*
	Stenosis type A	2	2
	Stenosis type B	3	3
	Stenosis type C	5	5
	The defeat of the p/3	4	3
	The defeat of the m/3	6	6
	The defeat of the d/3	2	1
RCA	n (%)	28 (35.9%)	17 (22.9%)
	Mean% of stenosis	79.03±15.59	80.14±17.97
	Average diameter, mm	3.29±0.56	3.06±0.59^
	Stenosis type A	1	4
	Stenosis type B	14	8
	Stenosis type C	11	5
	The defeat of the p/3	10	6
	The defeat of the m/3	10	8
	The defeat of the d/3	6	5

**-P=0.018and^-P=0.015(differencesbetweengroups);ADA–Anterior descending artery; CA – Circumflex artery; RCA – Right coronary artery; p/3 – proximal third; m/3 – middle third; d/3 – distal third.*

The intra-group analysis showed highly valid progress in laboratory parameters. At the same time, according to echocardiography, there was a statistically significant increase in only LVEF in Group 2 (Table 4). Intergroup comparative analysis did not reveal statistically significant differences, which, in our opinion, is due to the same therapeutic effect of both BVS and DES. Figure 3 presents a clinical example of BVS implantation with a 12-month dynamic evaluation. Repeated CAG after 1 year was carried out in 5 respondents with implanted BVS, but no resorption of these devices was observed (Table 5).

Table 3.

Quantitative characteristics for implantation techniques (BVS/DES)

Variable	Group 1	Group 2
Total number of lesions	110	105
Defeats with L> 20mm	33 (42.6%)	37 (50.0%)
Implantation of more than one skeleton	32 (41.0%)	31 (41.9%)
Predilatation	70 (89.7%)	64 (86.5%)
Mean diameter of the ball for predilution	2.54±0.3	2.57±0.4
Mean pressure of the ball for predilution	12.8±1.6	13.1±1.7
Average exposure t sec.	50.8±1.7	48.4±1.3*
Postdilation	78 (100%)	74 (100%)
Total number of implanted scaffolds	108	98
Mean length of scaffold	24.4±10.3	26.5±8.2
Mean diameter of scaffold	3.2±0.3	3.3±0.5
Type of post-dilatation balloon		
High-pressure balloon	65 (83.3%)	62 (83.8%)
Low pressure balloon	13 (16.7%)	12 (16.2%)
The average diameter of the balloon for post-dilatation	3.17±0.33	3.29±0.53
The mean pressure of the balloon for post-dilatation	14.34±4.42	14.33±4.45

**-P<0.05 (differences between groups).*

Table 4.

The assessment of clinical, laboratory and functional characteristics of patients 1 year after PCI (intra-group analysis)

Variable	Initial data	After 12 months
BVS	n=78	n=54
Ch, mg/dl	192.6±59.0	188.8±55.2
TG, mg/dl	227.1±160.3	135.2±33.2**
HDL, mg/dl	35.6±8.6	37.8±4.8
VLDL, mg/dl	46.5±33.3	27.0±6.8**
LDL, mg/dl	110.1±46.8	124.0±54.6
Glucose, mmol/l	6.1±1.9	4.7±0.5**
CRP, g/l	19.1±29.1	4.6±1.3**
LVEDV, ml	150.7±33.9	129.1±28.6
LVESV, ml	64.7±27.4	56.6±19.5
IST, mm	10.4±1.4	11.1±1.1
LVPWT, mm	10.1±1.2	10.8±0.8
LVEF, %	58.2±10.2	60.3±5.9
DES	(n = 78)	(n=54)
Ch, mg/dl	180.7±45.1	144.5±36.8**
TG, mg/dl	195.9±123.4	139.7±45.5*
HDL, mg/dl	35.9±6.5	38.8±7.6*
VLDL, mg/dl	39.2±24.7	28.7±11.4*
LDL, mg/dl	105.6±35.9	77.7±25.4**
Glucose, mmol/l	6.6±2.4	5.3±1.8*
CRP, g/l	23.2±48.5	3.7±1.9*
LVEDV, ml	150.1±39.7	147.3±33.3
LVESV, ml	65.8±26.8	61.4±15.5
IST, mm	10.3±2.4	10.2±1.1
LVPWT, mm	10.3±1.4	10.7±1.1
LVEF, %	57.3±7.7	61.6±5.8*

**- P<0.05 and **- P<0.001 (differences between groups)*

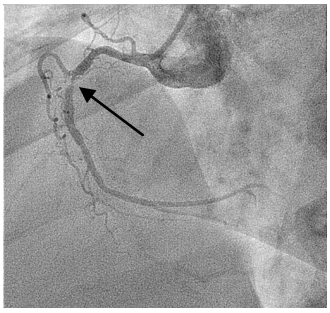

Fig. 3A. *SYNTAX Score: 2 points. Initial angiogram of RCA. (The arrow indicates stenotic lesion).*

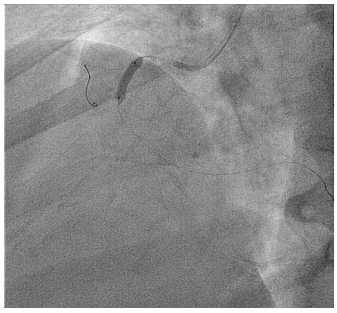

Fig.3B. *Inflating.*

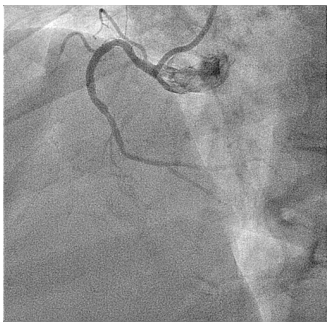

Fig. 3C. *Angiogram RKA after implantation of BVS (3.0 ×18mm).*

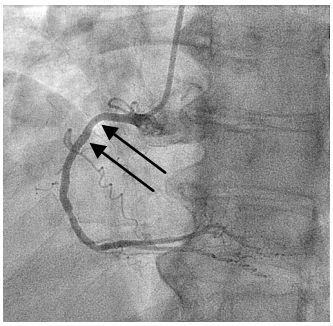

Fig. 3D. *Control CAG after 1 year. (The arrows mark the areas of visualization of the carcass marks).*

Table 5.

Long-term (12 months) clinical and angiographic results of myocardial revascularization

Variable	BVS (n=54)	DES (n=48)
Heart death	0	0
MI of the target vessel	0	0
Restenosis	0	0
Recurrent / progressive angina	0	0
Stent thrombosis	1 (after 3 months)	0
Bioresorption of BVS	0	

Discussion

The prospect of using a temporary vascular stent, or bio-soluble scaffold, has always been the goal of the intervention community. Such a device must have a radial stiffness to confront the acute collapse of the vessel after the intervention is finished, and must also completely resolve in a distant period, which would restore the biological and physiological properties of the vessel. The Absorb BVS manufactured by Abbott Vascular, Inc. is the first innovative device for the treatment of CAD. It gradually dissolves and is fully absorbed by the body over time. When installed, the BVS first increases the lumen of the artery, and then within 3 months secretes a drug that blocks the process of re-overgrowing the lumen of the artery. Two years later the scaffolds resolve themselves, breaking down into natural components for the body—water and carbon dioxide.

The first study of ABSORB showed impressive results. The main clinical endpoints of this study were the evaluation of the safety and efficacy of the device after 30 days, 6 months, and then every year for 5 years. The results of the first stage established that MACE frequency in 5 years was less than 3.4%. The results of the second stage (duration almost 3 years) also confirmed the efficacy and safety of Absorb in patients with CAD.[10-12] According to our data, MACE frequency (a 12-month evaluation) was 0%. In our opinion, this kind of success was due to strict adherence to the technique of implantation of this type of stent. A BVS has thick striae (beams), which makes it difficult to hold it in the place of constriction. This, in turn, requires not only special surgical skills, but also a certain anatomy of the vessels. In our study, the implantation of BVS-skeletons took more time, and they were installed in vessels with a larger diameter (Tables 2 and 3).

ABSORB-II was a prospective, randomized, controlled clinical trial evaluating the safety and efficacy of Absorb BVS as compared to the XIENCEPRIME DES. The study included 500 patients from the centers of Europe and New Zealand. After 1 year, the overall clinical results of the Absorb treatment were comparable to the XIENCE results at a lower revascularization frequency. The incidence of Absorb revascularization was 3.6% vs. 7.3% for XIENCE DES (P=0.08).[13] The results of our study also show comparability not only for angiographic, but also clinical and laboratory indicators between the BVS and DES groups.

However, the recently available 3-year follow-up data from the ABSORB III study continue to show an increased rate of MACE in BVS patients, when compared to patients implanted with the XIENCE stent. Specifically, there was a 13.4 percent rate of MACE (e.g., cardiac death, heart attack, or the need for an additional procedure to re-open the treated heart vessel) in patients treated with the BVS at 3 years, compared with 10.4 percent in patients treated with Abbott Vascular's approved metallic drug-eluting stent, the XIENCE stent (P=0.056). The ABSORB III study showed a 2.3% rate of thrombosis within the BVS scaffold versus 0.7% within the XIENCE stent at 3 years (P=0.01).[8]

It should be emphasized that these higher adverse cardiac event rates in BVS patients were more likely when the device was placed in small heart vessels. Among BVS-treated patients who developed device blood clots after 1 year, most had discontinued DAT. An additional preliminary analysis of ABSORB III data suggests improved clinical performance and a lower rate of complications associated with BVS implantation when health care providers follow the recommended implantation methods. The FDA-approved labeling for the BVS includes recommendations on selecting appropriately-sized heart arteries for BVS implantation (e.g., avoiding BVS use in small heart vessels) and methods to properly implant the device against the vessel wall.[7,8]

Stent thrombosis develops most often during the first month after stenting and, as a rule, ends with Q-wave MI or death of the patient. With gradual improvement of stent implantation technology and mandatory DAT (aspirin+thienopyridine) for 1 month, followed by continued use of aspirin without time limits, the incidence of stent thrombosis decreased to an acceptable 1%.[14]

According to the recommendations of AHA/ACC/SCAI,[15] aspirin is mandatory during the procedure; however, its dose and duration of administration depend on both the type of stent and the risk of bleeding in the patient. These 2 factors also determine how long the patient should take clopidogrel. At the same time, according to the CHARISMA study, a longer

DAT does not lead to a reduction in ischemic events among patients with atherothrombosis and persons with risk factors for its development. Such therapy was only accompanied by an increased risk of bleeding.[16] Thus at the present time, the question of the duration of DAT after PCI is open. Based on the data of our own experience, in particular on the above-mentioned single case of developing stent thrombosis, we tend to adhere to the tactics of long (at least 2 years) DAT reception. Our study has shown sufficient efficiency of the new scaffold Absorb. Based on the data we obtained, it can be argued that BVS can be used in everyday clinical practice, subject to strict adherence to the basic recommendations for implantation techniques. The reception of DAT should be conducted individually under the control of coagulogram indices.

In conclusion:

- Implantation of BVS in patients with different forms of CAD did not cause any angiographic or clinical complications, either at the hospital or at 12-month observation stages, and the results were comparable to those of the DES group.
- The technique of implanting BVS and the reception of DAT are the key factors for achieving positive results in real clinical practice.
- The use of BVS-frameworks contributes to improving clinical, functional and laboratory indicators, while the observed positive dynamics are comparable to similar data of the DES group.
- Regardless of the type of implanted stents, the survival rate among CAD patients within 12 months after stenting was 100%, while none of the respondents during this time developed acute MI or recurrence of angina attacks.

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 26-32
http://dx.doi.org/10.21103/Article8(1)_OA3

ORIGINAL ARTICLE

Neurology

INTERNATIONAL JOURNAL OF BIOMEDICINE

Process of Personalized Prescription of Valproic Acid as the Main Element of the Management of Epilepsy

Elena N. Bochanova, PhD; Natalia A. Shnayder, PhD, ScD*;
Diana D. Dmitrenko, PhD, ScD; Ivan P. Artyukhov, PhD, ScD; Sergey D. Gusev, PhD;
Elena A. Yurjeva, PhD; Olga S. Shilkina, MD

Krasnoyarsk State Medical University named after Prof. V.F. Voino-Yasenetsky
Krasnoyarsk, the Russian Federation

Abstract

The purpose of this study was to develop a sequential process of personalized valproic acid (VPA) prescription in patients with epilepsy.

***Materials and Methods*:** We randomly selected 167 patients with epilepsy receiving VPA, based on carriage of CYP2C9*2 and/or CYP2C9*3 and therapeutic drug monitoring. The patients' CYP2C9 status was determined by CYP2C9 genotyping before the beginning of anticonvulsant therapy.

Results: The sequence of personalized valproic acid prescription has been developed.

Conclusion: Using the sequential process of personalized VPA prescription will allow neurologists, psychiatrists and general practitioners to select starting and maintenance dosages of VPA with respect to the individual patient's pharmacogenetic profile and thereby, significantly improve the safety of pharmacotherapy in epilepsy patients.**(International Journal of Biomedicine. 2018;8(1):26-32.)**

Key Words: epilepsy • CYP2C9 • valproic acid • pharmacogenetics • pharmacokinetics

Abbreviations

ADRs, adverse drug reactions; **AEDs**, antiepileptic drugs; **CYP2C9**, cytochrome 2C9; **SNPs**, single-nucleotide polymorphisms; **TDM**, therapeutic drug monitoring; **VPA**, valproic acid.

Introduction

A personalized approach to the use of VPA, based on carriers of CYP2C9 gene polymorphisms and results of TDM of VPA, can significantly increase treatment safety by reducing the incidence of ADRs from AEDs.[1-3] Desired positive outcomes can only be achieved when a defined sequence of actions is adhered to. However, this sequence is often ambiguous and leads to errors. To avoid this, we utilized block diagrams based on conventional visual programming languages to capture the correct sequence of events in VPA prescriptions.[4,5]

Research into the carriage of polymorphisms of human cytochrome P450 enzymes involved in the metabolism of VPA (*CYP2C9, CYP2A6, CYP2B6, CYP1A1, CYP2D6, CYP2E1(1), CYP2E1(2)*) showed that the CYP2A6 was the most active in catalyzing VPA 3-hydroxylation whereas CYP2A6 and CYP2B6 were less active.[6]

The *CYP2C9* gene is highly polymorphic, with more than 50 known alleles. *CYP2C9*1* is the wild-type allele and is associated with normal enzyme activity.[7] In carriers of *CYP2C9*2* (c.430C>T; p.Arg144Cys) and *CYP2C9*3* (c.1075A>C; p.Ile359Leu), the *CYP2C9* activity is decreased.[8] The level of toxic metabolites of VPA in human liver microsomes is increased by 28%-31% in samples with one mutated *CYP2C9* allele (*CYP2C9*2* or *CYP2C9*3*) and by

***Corresponding author:** Prof. Natalia A. Shnayder, PhD, ScD; Head of the Neurological Center of Epileptology, Neurogenetics and Brain Research of the University Clinic of Krasnoyarsk State Medical University named after Prof. V.F. Voino-Yasenetsky, Krasnoyarsk, the Russian Federation. E-mail: nataliashnayder@gmail.com*

58%-73% in samples with two mutated *CYP2C9* alleles.[9] In carriers of *CYP2C9*2* and/or *CYP2C9*3*, in comparison with carriers of *CYP2C9*1*, there is an achievement of high blood concentrations of VPA, often exceeding the upper level of the therapeutic references, even when taking medium therapeutic doses of this drugs.[10,11] An accumulation of VPA in the blood up to a toxic level can cause the development of serious ADRs due to hyperammonemia, which results from a valproate-induced decrease in L-carnitine in serum. The decrease in L-carnitine is caused by an increase of L-carnitine excretion from urine (in the form of valproilcarnitine) and reduced renal reabsorption of free carnitine and acylcarnitine. L-carnitine deficiency in hepatocytes cases impaired urea synthesis and development of hyperammonemia.[12] Some authors [13,14] showed that the development of ADRs on the background of VPA use was associated with the development of hyperammonemia in patients with no liver diseases. L-carnitine is successfully used to eliminate the ADRs caused by hyperammonemia, including in the treatment of acute valproate overdose.[15] Thus, despite the large amount of information on VPA use in clinical practice, accumulated during more than half a century, pharmacogenetic studies of VPA open up new opportunities for improving the effectiveness and safety of therapy.[16]

The purpose of this study was to develop a sequential process of personalized VPA prescription in patients with epilepsy.

Materials and Methods

We randomly selected 167 patients with epilepsy receiving VPA, based on carriage of *CYP2C9*2* and/or *CYP2C9*3* and TDM status. The patients' CYP2C9 status was determined by CYP2C9 genotyping before the beginning of anticonvulsant therapy. We used a sorption method with a set of reagents for DNA isolation (DNA-Sorb-B*)* from the clinical material. Genotyping for the *CYP2C9*2* (Arg144Cys, C430T) and *CYP2C9*3* (Ile359Leu, A1075C) polymorphisms was performed according to previously published research protocols.[2] Block diagrams of processes were constructed using Misrosoft Visio 2013.

Results

The sequence of personalized VPA prescription was as follows (Figures 1-4). The first step is clinical diagnosis of epilepsy during the first visit to a neurologist, along with ruling out the following contraindications for VPA prescription: an increased sensitivity to any of the components of valproate; acute and chronic hepatitis; severe liver or pancreas dysfunction; hepatic porphyria; established mitochondrial diseases caused by mutations in the mitochondrial enzyme γ-polymerase (POLG) gene, such as Alpers-Huttenlocher syndrome; urea cycle disorders; patients on mefloquine or Hypericum perforatum preparations, and drugs that increase enzymatic liver activity or disturb intestine function (Fig.1).

After the diagnosis of epilepsy and the decision to start VPA, all patients are genotyped for the *CYP2C9*2* and *CYP2C9*3*.

Interpretation of results and VPA dosage are discussed and decided on at the next visit. A starting dose of VPA depends on the homo- or heterozygous carriers of the identified SNPs in *CYP2C9*. Normal metabolizers (genotype *CYP2C9*1/*1*) are prescribed a medium therapeutic dose of VPA. Intermediate metabolizers (heterozygous genotypes *CYP2C9*1/*2* and *CYP2C9*1/*3*) are prescribed a VPA dose 25%-30% lower than average. In poor metabolizers (homozygous genotypes *CYP2C9*2/*2* and *CYP2C9*3/*3* or compound heterozygotes of *CYP2C9*2/*3*), the VPA dose is reduced by 50%. The rate of titration of the daily dose of VPA also depends on the identified SNPs of *CYP2C9*: normal metabolizer – once every 3 days, intermediate metabolizer – once a week, poor metabolizer – once every 2 weeks. Patients are assigned a follow-up date with respect to their genotype: normal metabolizer after 6 months, intermediate metabolizer after 3 months, and poor metabolizer after 2 months. All patients are required to conduct VPA TDM prior to their appointment (Fig.1).

Further treatment depends on the results of TDM, clinical effectiveness and ADRs associated with VPA treatment over the last period.

The choice of drug for correction of ADRs of VPA is based on the pathogenetic mechanism of their development. Valproate-induced decrease in the synthesis of L-carnitine in hepatocytes' mitochondria is due to a complex formation between VPA and L-carnitine (valproilcarnitine), which is freely excreted in urine. In addition, reduced renal reabsorption of free carnitine and acylcarnitine leads to a deficiency of L-carnitine, and as a consequence, development of hyperammonemia.[8,9] It is also known that receiving VPA often leads to cholestasis development.[10] The course of hepatoprotective therapy is possible without VPA withdrawal.[11] Prescribing L-carnitine and ursodeoxycholic acid eliminates L-carnitine deficiency and provides a hepatoprotective effect.

For patients with a therapeutic level of VPA TDM (Fig. 2):

- In cases of a positive clinical and electroencephalographic dynamic during treatment, and absence of ADRs, treatment is continued at the same dose.
- In cases of a positive clinical and electroencephalographic dynamic during treatment with development of ADRs, L-carnitine and ursodeoxycholic acid are prescribed for 1 month. VPA dose is reduced by 25% in patients with ADRs such as aggravation of seizures, thrombocytopenia, hepatopathy, severe alopecia, hand tremor, and significant increase in body weight. In patients with such ADRs as onihodistrophy, non-coarse tremor of the hands and structurally unstable alopecia, and a slight increase in body weight, the therapy continues at the same dose, with the patient's consent.
- In cases of the absence of clinical and electroencephalographic effect and ADRs development, VPA is discontinued and another AED is prescribed. All patients in this group are prescribed L-carnitine and ursodeoxycholic acid for 1 month.
- In cases where the VPA effect and ADRs, are absent, treatment can be continued with dose escalation by 25%, with the patient's consent. The rate of dose titration is dependent on the individual patient's pharmacogenetic profile.

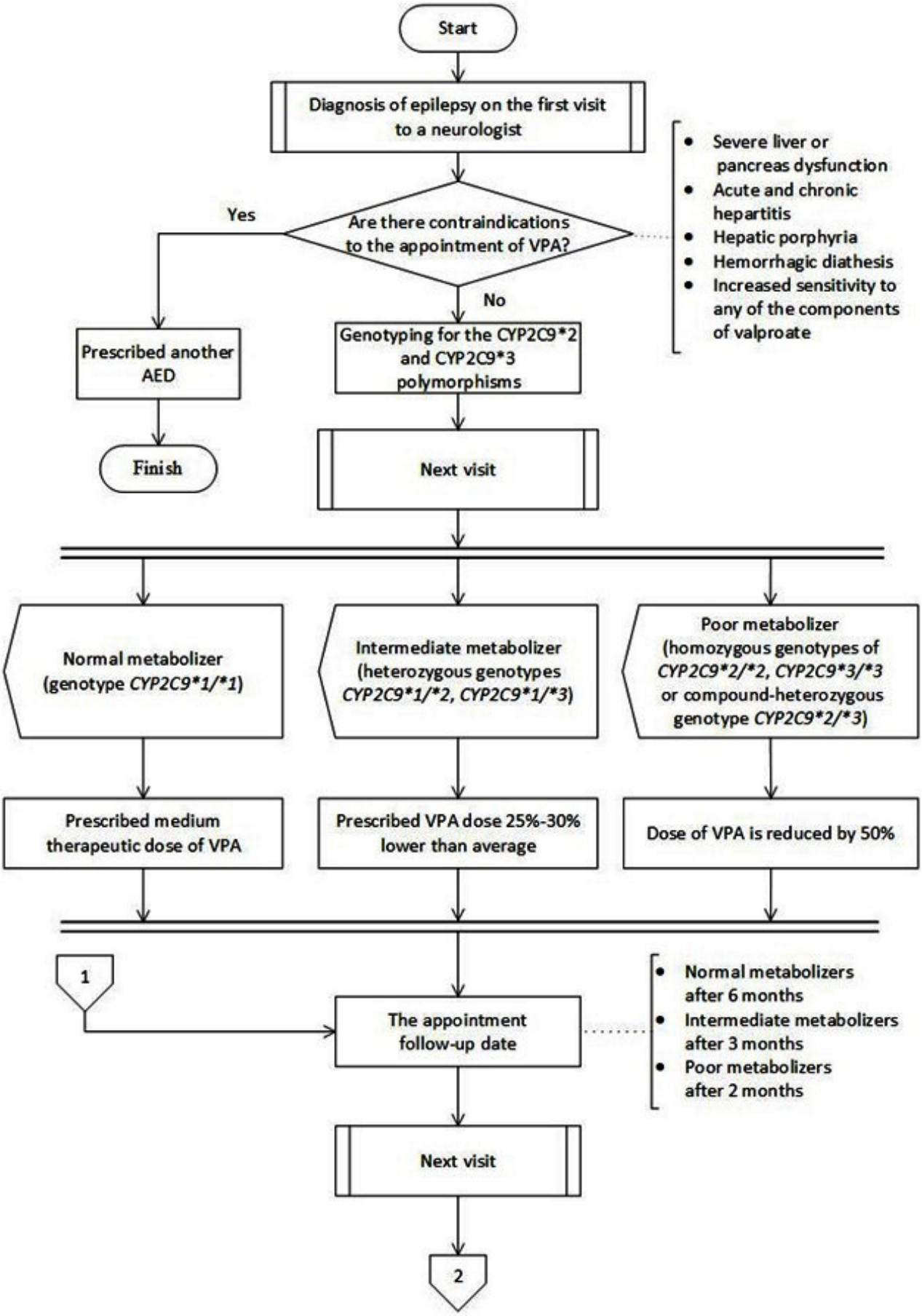

Fig. 1. *Sequence of personalized VPA prescription (the start of the process).*

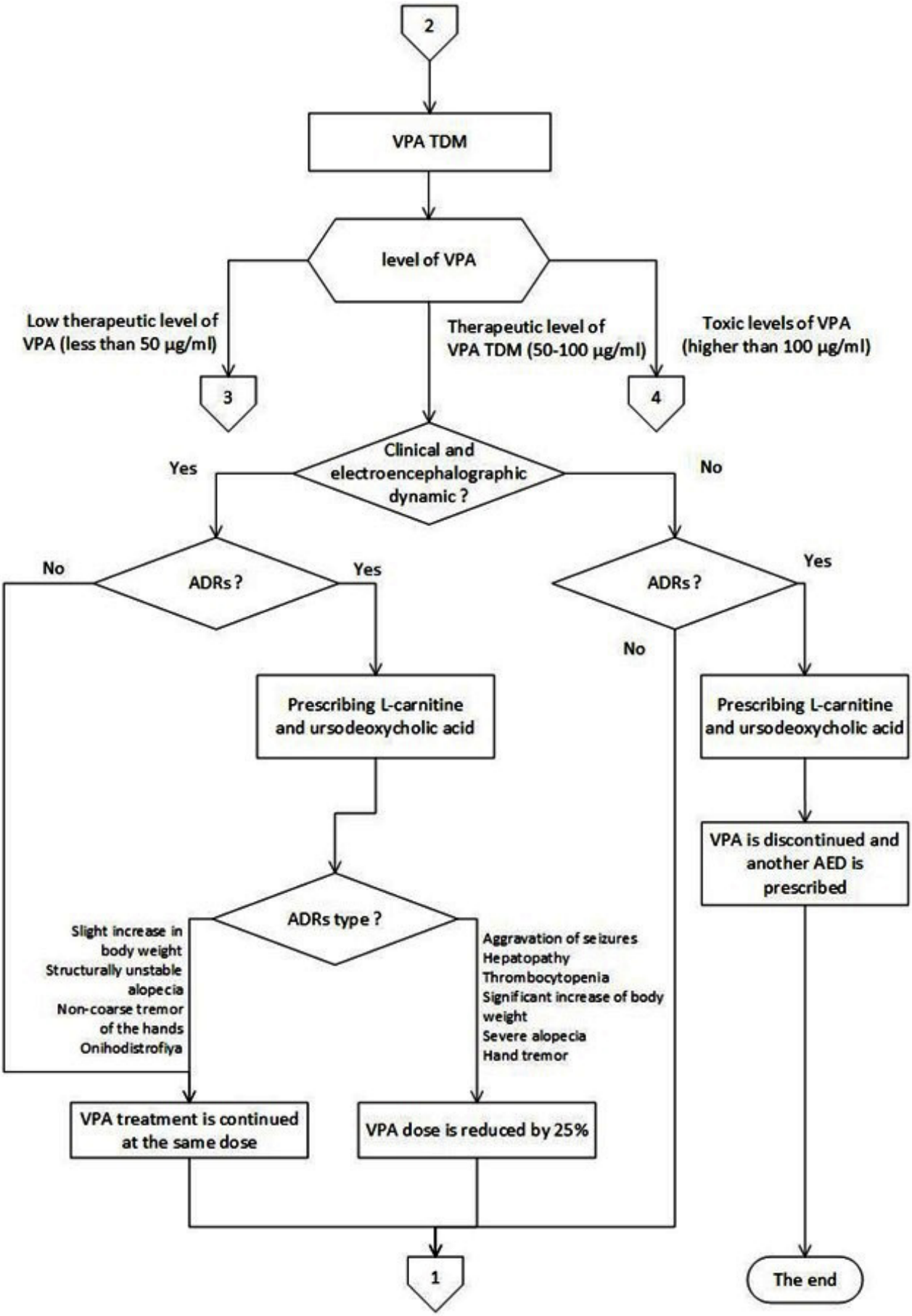

Fig. 2. *Process of personalized VPA prescription (continued).*

Patients with a low therapeutic level of VPA (< 50 µg/ml) are assessed for compliance with the dosing and scheduling regime of AED administration (Fig.3).

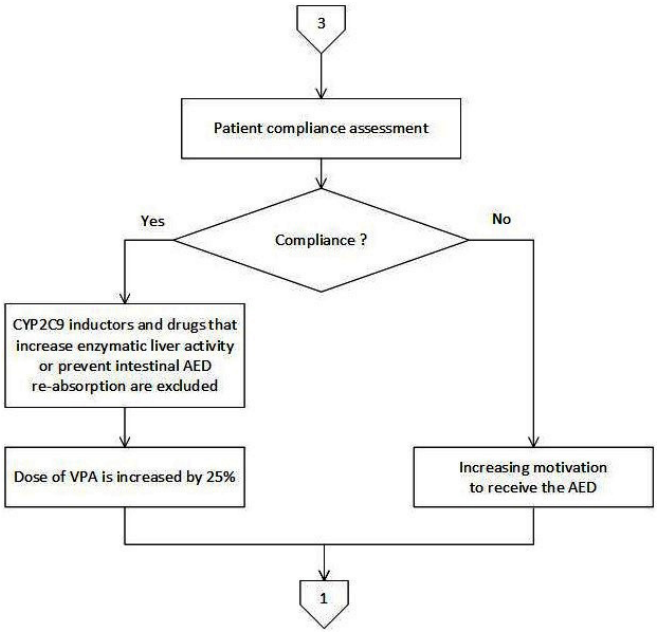

Fig. 3. *Process of personalized VPA prescription (continued).*

In cases of low patient compliance, motivation to receive the AED is increased and VPA treatment is continued at the same dose.

If a patient follows the recommendations for VPA administration, drugs that increase enzymatic liver activity or prevent intestinal AED re-absorption are excluded. Additional pharmacogenetic examination is advised to exclude profile "normal metabolizer." The dose of VPA is increased by 25% with the rate of dose titration depending on the individual pharmacogenetic profile of the patient.

Patients with toxic levels of VPA (>100µg/ml), as a result of TDM, are prescribed L-carnitine and ursodeoxycholic acid for 1 month (Fig.4).

In cases of positive clinical and electroencephalographic dynamic during treatment and the absence of ADRs, the dose of VPA is reduced by 25%.

In cases of a positive clinical and electroencephalographic dynamic during treatment and development of ADRs, the dose of VPA is reduced by 50% in patients with ADRs such as the aggravation of epileptic seizures, thrombocytopenia, hepatopathy, alopecia and coarse tremor of hands, and a significant increase in body weight. In patients with ADRs in the form of onychodystrophy, non-coarse tremor of the hands and structurally unstable alopecia, and a slight increase in body weight, the treatment dose is reduced by 25%.

In cases where the clinical and electroencephalographic effect of VPA treatment with development of ADRs is absent, VPA is discontinued and another AED is prescribed.

In cases of the absence of clinical and electroencephalographic effect of VA without development of ADRs, treatment may be resumed with a dose reduction by 25%, with patient's consent.

In conclusion, using this sequential process of personalized VPA prescription will allow neurologists, psychiatrists and general practitioners to select starting and maintenance dosages of VPA with respect to the individual patient's pharmacogenetic profile and thereby, significantly improve the safety of pharmacotherapy in epilepsy patients.

Competing interests

The authors declare that they have no competing interests.

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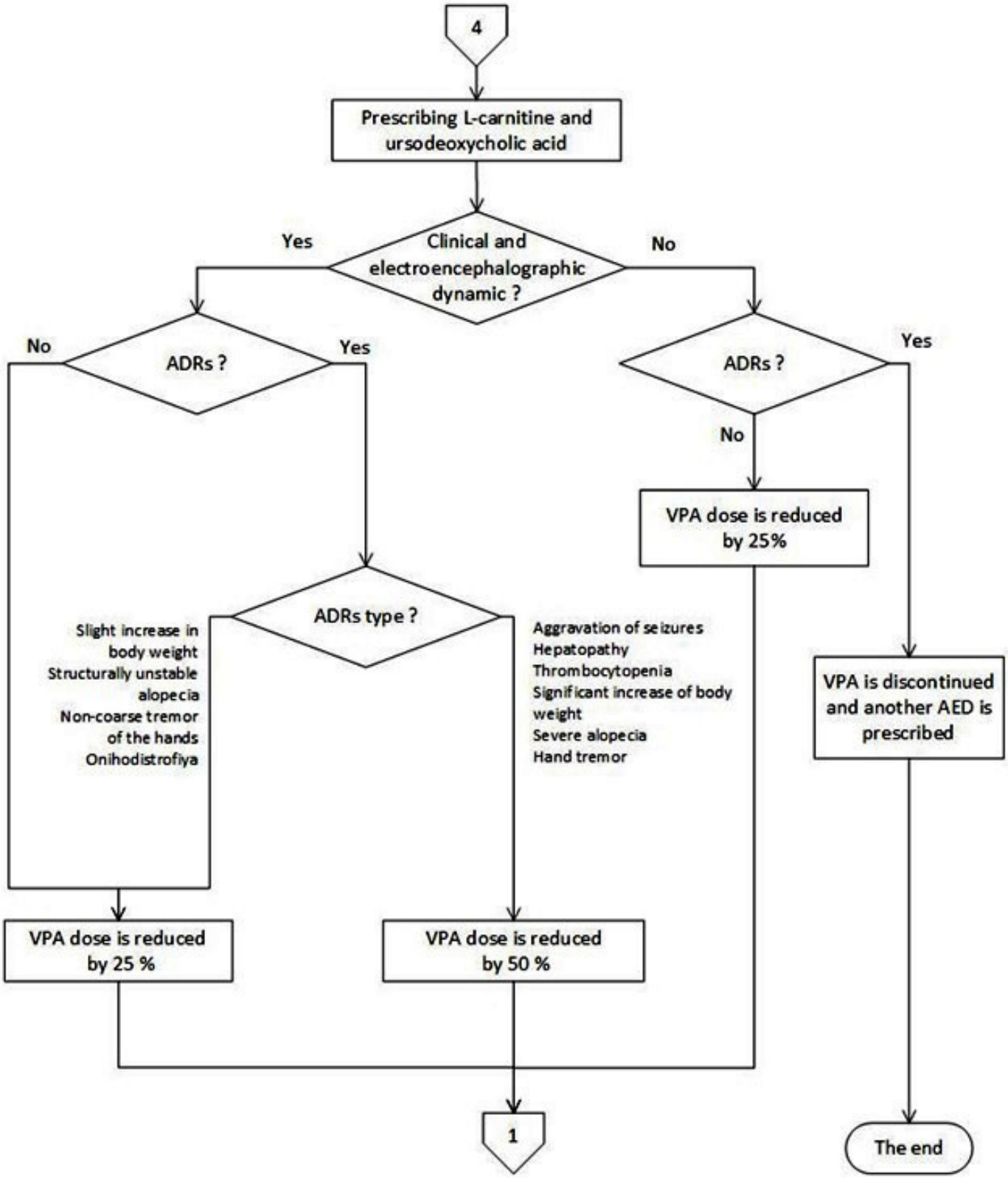

Fig. 4. Process of personalized VPA prescription (continued).

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International Journal of Biomedicine 8(1) (2018) 33-36
http://dx.doi.org/10.21103/Article8(1)_OA4

ORIGINAL ARTICLE

Pulmonology

INTERNATIONAL JOURNAL OF BIOMEDICINE

Impact of Metabolic Syndrome Components on Asthma Control and Life Quality of Patients

Andrey V. Budnevsky, PhD, ScD; Ludmila V. Tribuntceva, PhD; Svetlana A. Kozhevnikova, PhD; Evgeniy S. Ovsyannikov, PhD*

Voronezh State Medical University named after N.N. Burdenko
Voronezh, the Russian Federation

Abstract

The purpose of this study was to assess the characteristics of the clinical course of asthma in patients with metabolic syndrome (MetS) and to analyze the influence of the MetS components on the course of asthma, control of asthma symptoms, and the the quality of life (QL) of patients with asthma.

Materials and Methods: We examined 95 asthma patients aged from 18 to 60 years. The patients were divided into 2 groups. Group 1 included 35 patients without MetS (24/68.6% women and 11/31.4% men; mean age, 49.81±0.77 years), and Group 2 included 60 patients with MetS (45/75.0% women and 15/25.0% men; mean age, 49.82±0.77 years). The asthma diagnosis was based on the integral assessment of symptoms, medical history, health status, and spirometry values according to the Global Strategy for Asthma Management and Prevention (2017). MetS was diagnosed according to the IDF consensus criteria (2006).

Results: The MetS components, such as central obesity, high blood pressure, dyslipidemia, carbohydrate metabolism disorder, cause a more serious and unfavorable course of asthma, with frequent exacerbations, calls to emergency service and hospital admissions, severe asthma symptoms, uncontrolled asthma symptoms, low spirometry results, and low level of QL in the physical and psychological domains. **(International Journal of Biomedicine. 2018;8(1):33-36.)**

Key Words: asthma • metabolic syndrome • uncontrolled asthma symptoms • quality of life

Abbreviations

BP, bodily pain; **BMI**, body mass index; **CO**, central obesity; **FEV_1**, forced expiratory volume in 1 sec; **FP**G, fasting plasma glucose; **FVC,** forced vital capacity; **FEF**, forced expiratory flow; **GH**, general health; **HDL-C**, high-density lipoprotein cholesterol; **MetS**, metabolic syndrome; **MH**, mental health; **OGTT**, oral glucose tolerance test; **PEF**, peak expiratory flow; **PF**, physical functioning; **QL**, the quality of life**; RP**, role-physical functioning; **RE**, role emotional; **SF-36**, the 36-Item Short-Form Health Survey; **SF**, social functioning; **TG**, triglycerides; **VC**, vital capacity; **VT**, vitality; **VAS**, the 10-point Visual Analogue Scale; **WC**, waist circumference.

Introduction

Asthma and MetS have a considerable impact on public health, and their prevalence has increased in recent years.[1] Numerous studies have linked these disorders. The presence of complex, multiple pathogenetic links in these nosological forms is emphasized, which in most cases leads to the formation of the phenomenon of mutual burdening, limiting the achievement of a controlled course of asthma on the one hand, and increasing the risk of developing diabetes mellitus and cardiovascular diseases on the other.[2] WHO estimates that 235 million people currently suffer from asthma. Asthma is the most common noncommunicable disease among children. According to the latest WHO estimates, released in December 2016, there were 383 000 deaths due

***Corresponding author:** Evgeniy S. Ovsyannikov, PhD. Department of faculty therapy, Voronezh State Medical University named after N.N. Burdenko. Voronezh, the Russian Federation. E-mail: ovses@yandex.ru*

to asthma in 2015.[3] The number of severe and late-onset forms of asthma is increasing; the incidence of disability due to asthma is about 2.0% of cases.[4] The disease reduces life expectancy by an average of 7 years for men, 15 years for women.[5]

Worldwide, there has been a significant increase in the number of patients with MetS—up to 15%-25% of the adult population—which is associated with the high prevalence of obesity in the world.[6] The increase in MetS is reflected primarily in the increased prevalence of such diseases as diabetes, as well as diseases associated with dyslipidemia and atherosclerosis (ischemic heart disease, cerebrovascular disease, arterial hypertension). At the same time, the association between MetS and the index of total mortality from cardiovascular diseases is becoming more and more evident.[7]

In recent years, the question of the influence of MetS components on the course of asthma has been increasingly studied, and the features of the combined course of these nosological forms are also being studied.[8]

It has been found that improvement of living conditions (easy access to more food) contributes to the growth of both obesity and asthma. Obesity promotes the development of bronchial hyperreactivity caused by physical exertion in patients with asthma.[9]

Arterial hypertension is associated with asthma development. This association can be considered as a cardio-pulmonary problem because of close anatomical and functional relations between these two systems.[10]

Disorders of lipid metabolism and risk of asthma development are based on common risk factors, such as lack of adequate physical activity, sedentary life style, improper feeding, environmental influence, and psychoemotional stress.[11] Possible association between asthma and carbohydrate metabolism disorder can also be explained by lack of adequate physical activity and improper feeding.[12]

In the available literature, there are inconsistent, insufficient data on the relationship between the degree of severity of the MetS components and the features of clinical and instrumental indicators of asthma, the level of disease control, and QL of asthma patients.

The purpose of this study was to assess the characteristics of the clinical course of asthma in patients with MetS and to analyze the influence of the MetS components on the course of asthma, control of asthma symptoms, and the QL of patients with asthma.

Materials and Methods

We examined 95 asthma patients aged from 18 to 60 years. The patients were divided into 2 groups. Group 1 included 35 patients without MetS (24/68.6% women and 11/31.4% men; mean age, 49.81±0.77 years), and Group 2 included 60 patients with MetS (45/75.0% women and 15/25.0% men; mean age, 49.82±0.77 years). The asthma diagnosis was based on the integral assessment of symptoms, medical history, health status, and spirometry values according to the Global Strategy for Asthma Management and Prevention.[13] MetS was diagnosed according to the IDF consensus criteria. According to the new IDF definition (2006), the MetS is present when WC is increased (≥94 cm in males and ≥80cm in females (for Europids)) and at least two of the following factors are present: raised TG (≥1.7 mmol/l or specific treatment for this lipid abnormality); reduced HDL-C (< 1.03 mmol/l in males and < 1.29 mmol/l in females or specific treatment for this lipid abnormality); systolic blood pressure ≥ 130mmHg or diastolic blood pressure ≥ 85 mmHg or treatment of previously diagnosed hypertension; raised FPG (≥5.6 mmol/l) or previously diagnosed type 2 diabetes.[14]

The study was approved by the Ethics Committee of Voronezh State Medical University named after N.N. Burdenko. Written informed consent was obtained from each patient.

A comprehensive clinical examination and laboratory tests included the following procedures:

- Anthopometrical reference data: BMI was calculated using Quetelet's formula (in kg/cm^2). WC was measured using centimetric tape at the navel level on a horizontal line (in cm)
- Assessment of asthma severity included the number of exacerbations, calls to emergency service and hospital admissions for the past 12 months.
- Quantity assessment of asthma symptoms (dyspnea, chest tightness, cough, sputum) by VAS.
- Assessment of asthma symptoms control by Asthma Control Test™ (ACT)
- Functional tests: spirometry
- Assessment of QL by SF-36
- Assessment of BP by Korotkov's method.
- Assessment of FPG, OGTT, and blood levels of TG and HDL-C.

All data was evaluated with STATGRAPHICS Plus 5.1. Baseline characteristics were summarized as frequencies and percentages for categorical variables and as mean±SD for continuous variables. Student's unpaired t-test was used to compare two groups for data with normal distribution. A probability value of $P<0.05$ was considered statistically significant.

Results and Discussion

According to clinical examination results, 60/63.2% patients with asthma had MetS:

- CO plus high level of TG plus low HDL-C in 8/13.3% patients
- CO plus high BP plus high level of FPG in 29/48.3% patients
- CO plus high TG plus low HDL-C plus high FPG in 7/11.7% patients
- CO plus high BP plus high TG plus low HDL-C in 16/26.7% patients

A comprehensive clinical examination demonstrated statistically significant differences in assessment parameters between Groups 1 and 2. In Group 2, the number of asthma exacerbations was 1.6 times greater (3.50±0.11 vs. 2.20±0.13 per year, $P=0.000$), calls to emergency service - 1.4 times greater (2.83±0.08 vs. 2.03±0.12 per year, $P=0.000$), and hospital admissions - 1.3 times greater (1.82±0.08 and

1.37±0.10 per year, *P=0.0007*) compared to Group 1.

There were significant differences in subjective assessment of asthma symptoms between Group 1 and 2, according to the 10-point Visual Analogue Scale. In Group 2, patients' dyspnea intensity was 1.8 times greater (5.85±0.13 vs. 3.29±0.10, *P=0.000*), chest tightness intensity - 1.5 times greater (3.07±0.10 vs. 2.03±0.22, *P=0.000*), cough intensity - 1.6 times greater (4.30±0.13 vs. 2.68±0.16, *P=0.000*), and sputum intensity - 1.5 times greater (3.68±0,11 vs. 2.48±0,19, *P=0.000)* compared to Group 1.

All patients with asthma and MetS had uncontrolled asthma symptoms. There were significant ACT value differences between Group 1 and 2: 17.03±0.54 and 13.58±0.32 (*P=0.0000*).

Spirometry results were also significantly different between Group 1 and Group 2 (Table 1).

Table 1.

Spirometry parameters in two groups

Variable	Group 1	Group 2
FVC, % of predicted	70.07±2.48	60.55±0.58*
VC, % of predicted	71.13±0.42	63.01±0.36*
FEV_1, % of predicted	73.82±0.64	65.05±1.21*
FEV1/FVC, %	78.59±0.53	70.79±1.48*
PEF, % of predicted	54.23±1.21	46.81±0.77*
FEF_{75},% of predicted	49.47±1.82	41.11±0.94*
FEF_{50},% of predicted	51.79±0.58	43.73±1.71*
FEF_{25},% of predicted	65.75±0.67	56.09±1.39*

**- P < 0.05*

Patients in Group 2 demonstrated a low level of QL in the physical and psychological domains, according to SF-36, in comparison with Group 1 (Table 2).

Table 2.

SF-36 parameters in two groups

Variable	Group 1	Group 2
PF	75.74±1.77	59.67±1.61*
RP	56.54±1.98	43.50±1.59*
BP	63.66±1.86	47.97±1.53*
GH	58.28±1.99	45.35±1.41*
VT	67.46±2.05	51.80±1.64*
SF	65.69±2.21	50.08±1.28*
RE	60.80±2.17	53.02±1.35*
MH	73.29±2.01	63.70±1.47*
PF	75.74±1.77	59.67±1.61*

**- P<0.05*

The performed correlation analysis between the indicators characterizing the clinical course of asthma, the level of control over the disease, and the QL of asthma patients with MetS components revealed statistically significant values of correlation coefficients. The MetS components, such as central obesity, high blood pressure, dyslipidemia, carbohydrate metabolism disorder, cause a more serious and unfavorable course of asthma, with frequent exacerbations, calls to emergency service and hospital admissions, severe asthma symptoms (dyspnea, chest tightness, cough with sputum), uncontrolled asthma symptoms, low spirometry results (FEV_1 and Tiffeneau index), and low level of QL in the physical and psychological domains.

In conclusion, the patients with asthma and MetS have a more severe clinical course of asthma, with frequent exacerbations and uncontrolled asthma symptoms, low spirometry results and a low level of QL.

Competing interests

The authors declare that they have no competing interests.

Acknowledgments

This work was partially supported by the Council on Grants of the President of the Russian Federation for State Support of Young Scientists and Leading Scientific Schools (grant NSh 4994.2018.7).

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International Journal of Biomedicine 8(1) (2018) 37-41
http://dx.doi.org/10.21103/Article8(1)_OA5

ORIGINAL ARTICLE

Obesity & Metabolic Syndrome

INTERNATIONAL JOURNAL OF BIOMEDICINE

The Assessment of Oxidative Stress Intensity in Adolescents with Obesity by the Integral Index

Marina A. Darenskaya, PhD, ScD[1]; Oksana A. Gavrilova[1]; Lyubov V. Rychkova, PhD, ScD[1]; Olga V. Kravtsova, PhD[1]; Lyudmila A. Grebenkina, PhD, ScD[1]; Elena V. Osipova, PhD, ScD[1,2]; Sergey I. Kolesnikov, Academician of the RAS[1,3]; Lyubov I. Kolesnikova, Academician of the RAS[1]

[1]Scientific Centre for Family Health and Human Reproduction Problems, Irkutsk, the Russian Federation
[2]Irkutsk State University, Irkutsk, the Russian Federation
[3]M.V. Lomonosov Moscow State University, Moscow, the Russian Federation

Abstract

The aim of this research was to assess lipid peroxidation (LPO) and antioxidative defense (AOD) changes in adolescent boys with obesity using the integral index.

Materials and Methods: We examined 19 adolescent boys with obesity of the first degree (the study group). The control group included 23 healthy adolescent boys. The study included the collection of anamnestic data, physical examination, and anthropometric data analysis (body weight (BW), height, waist circumference (WC), hip circumference (HC), and body mass index (BMI)). Laboratory analysis included an assessment of the blood levels of total cholesterol, triglycerides, HDL, LDL, and glucose, as well as the intensity of LPO and AOD in blood plasma and primary and secondary products of LPO. To measure the intensity of OS, the oxidative stress index (OSi) was calculated (the ratio of the LPO-AOD system indicators in the study group to average indicators in the control group).

Results: We found a statistically significant increase in BW, BMI, SDS BMI, WC, and HC in the study group compared to the control group. The obese patients had higher values of blood glucose, total cholesterol, triglycerides and LDL compared to the control group. In the study group, we found a significant decrease in the concentration of diene conjugates and an increase in the level of ketodienes and conjugated trienes. The values of α-tocopherol and retinol, and SOD activity were significantly decreased in the study group compared to the control group. There were no statistically significant changes in total antioxidant activity and glutathione status components. According to the data received, the OSi level in the group of obese patients increased approximately 7 times, which confirms the results on the development of antioxidant insufficiency in this pathology.**(International Journal of Biomedicine. 2018;8(1):37-41.)**

Key Words: lipid peroxidation • antioxidative defense • oxidative stress index • adolescent boys • obesity

Abbreviations

AOD, antioxidative defense; **BMI,** body mass index; **BW**, body weight; **CDs,** conjugated dienes; **GSH,** reduced glutathione; **GSSG,** oxidized glutathione; **HC**, hip circumference; **HDL,** high-density lipoprotein; **KD-CT,** ketodienes and conjugated trienes; **LDL**, low-density lipoprotein; **LPO,** lipid peroxidation; **OS,** oxidative stress; **SDS BMI,** the standard deviation score of BMI; **SOD,** superoxide dismutase; **TAA,** total antioxidant activity; **TBARs**, thiobarbituric acid reactants; **WC**, waist circumference.

Introduction

Childhood obesity is a serious medical condition that affects children and adolescents. In the developed countries, 25% of adolescents have excessive body weight and 15% have obesity.[1-4] The incidence of obesity is 6.8% in boys 5 to 17 years old and 5.3% in girls.[5] Childhood obesity usually progresses and often leads to health problems in adulthood: hypertension, metabolic syndrome, diabetes, and cardiovascular and fatty liver diseases.

WHO experts connect the prevalence of obesity in childhood with economic and social changes of life in a

modern society, unhealthy diets, and low physical activity.[6,7] The study of molecular mechanisms of obesity formation in adolescence is very important.[8-10] One of the pathogenetic mechanisms of obesity development is OS activation and a decrease in AOD activity.[11-15] Endogenous aldehydes generated during OS can act as mediators of damage, which precedes the emergence of metabolic shifts.[14]

The aim of this research was to assess LPO and AOD changes in adolescent boys with obesity using the integral index.

Materials and Methods

We examined 19 adolescent boys (mean age of 4.41±0.45 years) with obesity of the first degree (the study group) according to the WHO classification.[16] The control group included 23 healthy boys (mean age of 15.12±0,32 years). The study included the collection of anamnestic data, physical examination, and anthropometric data analysis (BW, height, WC, HC, and BMI). BMI was calculated using Quetelet's formula:

$BMI = body\ weight(kg)/height(cm)^2$

Laboratory analysis included an assessment of the blood levels of total cholesterol, triglycerides, HDL, LDL, and glucose. The quality, quantity and regularity of food intake were evaluated. Hereditary anamnesis included the presence of relatives with type 2 diabetes, obesity, ischemic heart disease, and hypertension. All patients did not take vitamins during the blood sampling period.

Blood was taken from the ulnar vein in accordance with the existing requirements in the morning after an overnight fast. Blood samples were centrifuged for 5 min at 1.500g at 4°C; erythrocytes were rinsed three times with 0.9% NaCI. Aliquots of the separated EDTA plasma and washed erythrocytes were used immediately or kept frozen at -40°C (not longer than one month).

We estimated the intensity of LPO and AOD in blood plasma (TAA, SOD, GSH, GSSG, α-tocoferol, retinol) and primary and secondary products of LPO (CDs, ketodienes, KD-CT and TBARs). The concentrations of CDs and KD-CT were evaluated at 232 nm in plasma heptane extracts.[17] The coefficient of molar absorption (K=2.2 • 105 M-1 C-1) for conversion of absorption units to μmol/L was used. TBARs levels were detected by fluorometry[18] and estimated in μmol/L.

Plasma levels of GSH, GSSG, α-tocopherol and retinol, as well as SOD activity in hemolysate were detected by fluorometry.[19-21] Plasma TAA levels were detected photometrically.[22] The measurements were conducted by a spectrophotometer consisting of two blocks: a UV-1650PC spectrophotometer and an RF-1501 spectrofluorimeter. To measure the intensity of OS, the oxidative stress index (OSi) was calculated (the ratio of the LPO-AOD system indicators in the study group to average indicators in the control group).[23]

The study was conducted in accordance with ethical principles of the WMA Declaration of Helsinki (1964, ed. 2000) and approved by the Ethics Committee of Scientific Centre for Family Health and Human Reproduction Problems. Written informed consent was obtained from all participants.

Statistical analysis was performed using the statistical software «Statistica». (v6.1, StatSoft, USA).The mean (M) and standard deviation (SD) were calculated. For data with normal distribution, inter-group comparisons were performed using Student's t-test. Differences of continuous variables departing from the normal distribution, even after transformation, were tested by the Mann-Whitney *U*-test. A probability value of $P<0.05$ was considered statistically significant.

Results and Discussion

We found a statistically significant increase in BW (1.48 times, $P<0.001$), BMI (1.5 times, $P<0.0001$, SDS BMI (12.43 times, $P<0.0001$), WC (1.35 times, $P<0.0001$, and HC (1.21 times, $P<0.0001$) in the study group compared to the control group. In obese patients, we found an increase of 1.4 times in glucose level ($P=0.028$), which can be explained by impaired glucose tolerance.[24] The obese patients had higher values of total cholesterol, triglycerides and LDL compared to the control group (Table 1). According to a number of authors, changes in the blood lipids in children with obesity depend on the severity of the disease.[25]

Table 1.

Clinical characteristics and biochemical parameters in adolescents of the study and control groups

Variable	Control group	Study group
Age, years	15.69±1.49	14.58±1.98
Weight, kg	64.83±9.17	95.82±15.06*
Height, cm	174.20±9.17	173.40±8.93
BMI	21.25±1.01	31.79±3.61*
SDS BMI	0.23±0.30	2.86±0.51*
WC, cm	73.20±4.53	99.10±8.49*
HC, cm	88.00±4.00	106.2±6.65*
Total cholesterol, μmol/L	3.43±0.44	4.83±0.64*
Triglycerides, μmol/L	0.43±0.16	1.63±0.52*
HDL, μmol/L	1.10±0.24	1.18±0.38
LDL, μmol/L	2.75±0.39	2.83±1.06*
Glucose, μmol/L	4.02±1.66	5.64±1.29*

** - P<0.05 vs. the control group*

In the study group, we found a significant decrease in the concentration of diene conjugates (primary products of LPO) and an increase in the level of ketodienes and conjugated trienes (secondary products of LPO) on the background of the absence of statistically significant changes in the content of TBA-active products (Table 2). The obtained results partly agree with multiple research data showing that obesity stimulates the processes of free radical oxidation, and the resulting OS acts as one of the pathogenetic mechanisms of obesity. Over-expression of OS damages cellular structures together with under-production of anti-oxidant mechanisms,

leading to the development of obesity-related complications. [26] It was found that the increased content of ketodienes and conjugated trienes might provoke a multifaceted deleterious effect on biopolymers and cellular structures. In obesity, increased OS in plasma is due to increased ROS production from accumulated fat Shigetada Furukawa and colleagues suggested that obesity per se may induce systemic OS stress and that increased OS in accumulated fat is, at least in part, the underlying cause of dysregulation of adipocytokines and development of metabolic syndrome.[11] OS plays a crucial role in disorders related to obesity, such as dyslipidemia.[26] In addition to a pro-inflammatory process, ROS can also directly damage lipids, proteins or DNA and modulate intracellular signalling pathways, such as mitogen activated protein kinases and redox sensitive transcription factors, causing changes in protein/lipid expression and, therefore, irreversible oxidative damage.[27] Due to ROS-mediated changes in lipid expression, further oxidation-derived products, including oxidative LDL (Ox-LDL), can play a further critical role in CVD. Additionally, Ox-LDL alters the production of adipokines which can lead to further OS.[26,28] Increased Ox-LDL in obese patients with dyslipidemia may be due to loss of antioxidant capacity caused by low serum activity of the antioxidant enzyme (SOD).[29]

Table 2.

Indicators of LPO and AOD in blood plasma

Variable	Control group	Study group
CDs, μmol/L	2.32±0.74	1.67±0.71*
KD-CT, units	0.26±0.12	0.43±0.26*
TBA-active products, μmol/L	0.86±0.45	0.97±0.48
TAA, units	15.73±3.52	15.39±4.43
α-tocopherol, μmol/L	8.15±2.8	5.75±3.38*
retinol, μmol/L	0.68±0.21	0.45±0.24*
SOD activity, units	1.69±0.1	1.42±0.29*
GSH, mmol/L	2.29±0.22	2.12±0.45
GSSG, mmol/L	1.94±0.22	1.89±0.47

** - P<0.05 vs. the control group*

The AOD system involving special antioxidant enzymes (SOD, catalase, enzymes of the glutathione redox system, water- and fat-soluble vitamins) plays an important role in protecting the body from oxidative damage.[26,30] Dysfunction of the AOD system is characterized by the development of the LPO syndrome and can lead to a number of negative consequences for the cell, such as membrane damage, inactivation or transformation of enzymes, suppression of cell division, and accumulation of inert polymers.[11,26] We found a decrease in the values of α-tocopherol (1.42 times, P=0.0158) and retinol (1.51 times, P=0.0025), and SOD activity (1.19 times, P=0.0001) in the study group compared to the control group (Table 2). There were no statistically significant changes in TAA and glutathione status components.

It has been established that even a slight decrease in SOD activity is an important signal of a metabolic shift towards the prevalence of prooxidant processes. α-tocopherol and retinol are the strongest bioantioxidants and the necessary nutritive factors.[30] Moreover, α-tocopherol shows membrane-protective and antimutagenic activity and is the most important regulator of oxidative homeostasis of cells. The in vitro and in vivo evidence of the function of vitamin E as a peroxyl radical-scavenging antioxidant and inhibitor of lipid peroxidation is presented in study by E.Niki.[31] According to many studies, vitamin A in large doses increased the antioxygenic potential of the tissues, and it was suggested that retinol also might be considered as a potential antioxidant similar to tocopherol. [32-34]

It seems optimal to use OSi because multidirectional changes occur in the LPO-AOD system with the development of various pathological conditions. For that purpose, the formula for OSi calculation was developed in our previous study [23]:

$$OSi=(CDs_i/CDs_n)\times(KD\text{-}CT_i/KD\text{-}CT_n)\times(TBA\text{-}active\ products_i/TBA\text{-}active\text{-}products_n)/(SOD_i/SOD_n)\times(GSH_i/GSH_n)\times(\alpha\text{-}tocopherol_i/\alpha\text{-}tocopherol_n)\times(retinol_i/retinol_n),$$

where i - the indicators levels in obese patients, n - the indicators levels in the control group.

OSi >1 shows the presence of OS. This formula takes into account not only the accumulation of LPO products at various stages, but also the activity of various parts of the AOD system. According to the data received, the OSi level in the group of obese patients increased approximately 7 times, which confirms the results on the development of antioxidant insufficiency in this pathology (Fig.1).

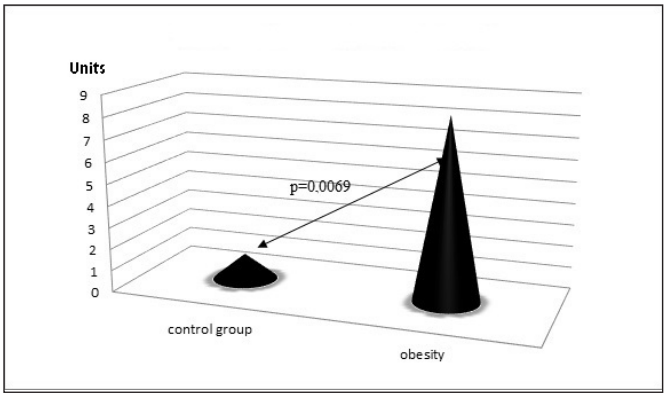

Fig.1. Oxidative stress index (OSi)

Conclusion

Thus, our study showed certain features of the changes in the LPO-AOD system in adolescent boys with obesity: a decrease in levels of primary LPO products and an increase in the level of secondary LPO products on the background of reducing the fat-soluble vitamins and SOD activity. The use of OSi confirmed the presence of antioxidant deficiency in adolescents with obesity.

Competing interests

The authors declare that they have no competing interests.

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***Corresponding author:** Marina A. Darenskaya, PhD, ScD. Scientific Centre for Family Health and Human Reproduction Problems, Irkutsk, the Russian Federation. E-mail: marina_darenskaya@inbox.ru*

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International Journal of Biomedicine 8(1) (2018) 42-46
http://dx.doi.org/10.21103/Article8(1)_OA6

ORIGINAL ARTICLE

Obstetrics & Gynecology

INTERNATIONAL JOURNAL OF BIOMEDICINE

Results of Fetal Ultrasound Imaging and Doppler Ultrasound Study Depending on the Factors of Perinatal Risk in Preterm Birth

Agamurad A. Orazmuradov, PhD, ScD[1]; Setonde Romeo D. Konnon, PhD[1]; Maya T. Khubetsova, PhD[1]; Irina V. Savenkova[1]; Anastasia V. Minaeva[1]; Dmitriy S. Novginov[1]; Vitaliy N. Kretsu[1]; Aleksey A. Lukaev, MD[2*]

[1]*RUDN University, Moscow, Russia*
[2]*Mytishchi City Clinical Hospital Mytishchi, Moscow Region, Russia*

Abstract

Premature birth (PB) is associated with placental insufficiency, which is confirmed in the morphological study of the placenta. Ultrasonic markers of placental abnormalities, often preceding PB, have been identified: changes in placental structure, premature ripening of the placenta, oligohydramnios, and fetal-placental circulation.**(International Journal of Biomedicine. 2018;8(1):42-46.)**

Key Words: oligohydramnios • intrauterine growth retardation • cervicometry • premature birth

Abbreviations

IUGR, intrauterine growth retardation; **PR**, perinatal risk; **PB**, premature birth.

Introduction

Premature birth (PB) is a multifactorial obstetrical complication, which should be predicted by taking into account the risk factors; the leading factors are infectious and inflammatory diseases of the genitourinary system, a burdened reproductive history of the mother (sexual debut before the age of 16, the early reproductive loss in the anamnesis) and tobacco smoking. The close interweaving of socio-biological factors and infectious and non-infectious factors, in combination with neuroendocrine disorders, create prerequisites for the placental insufficiency formation and disorders of uteroplacental and fetal-placental circulation. Currently, there is no clear evidence for the impact of perinatal risk (PR) on the outcome of PB. Numerical reflection of the degree of burden of the anamnesis and the course of pregnancy is the number of points of PR.

There is evidence of the leading role of placental insufficiency in the pathogenesis of premature termination of pregnancy.[1,2] The length of the cervix is the predictor of PB. Berghella and colleagues [3] indicated in their studies that the short cervix, measured with transvaginal ultrasound, is a risk for PB. Changes in the cervix are a more objective indicator than the evaluation of labor. If the length of the cervix is more than 3cm, the probability of labor in the next week is about 1%. Ultrasonic cervicometry is an objective, less invasive and more accurate method of assessing the patient's condition compared with a vaginal manual examination.[4]

The aim of this research was to assess the results of fetal ultrasound imaging and Doppler ultrasound study depending on the PR factors in PB.

Materials and Methods

The study was performed in Municipal clinical hospital #29 named after N.E. Bauman. We carried out a prospective analysis of 236 medical records of pregnant women with premature labor at 28 to 33 weeks (plus 6 days) of gestation (ICD-10 code O60).[5] According to premature labor classification, all pregnant women were divided into 2 cohorts: Cohort 1 (gestational age from 28 to 30 weeks plus 6 days) and Cohort 2 (gestational age from 31 to 33 weeks plus 6 days). Depending

[*]***Corresponding author:*** *Aleksey A. Lukaev, MD. Mytishchi City Clinical Hospital, Mytishchi, Moscow Region, Russia E-mail: aleksei_lukaev@mail.ru*

on the number of PR factors, each cohort was divided into 3 groups: a low PR, a moderate PR, and a high PR (Table 1).

Table 1.

Distribution of the examined women

Cohort 1 n=128			Cohort 2 n=108		
1st group Low PR	2nd group Moderate PR	3rd group High PR	1st group Low PR	2nd group Moderate PR	3rd group High PR
n=20	n=38	n=70	n=32	n=52	n=24

A specific group for PR was defined in accordance with the number of points derived from S. Knyazev's scale (2003): low risk (<15points), moderate risk (from 15 to 20 points), and high risk (≥25 points). Calculation of PR factors was performed twice: at admission and during labor. We identified the degree of PR based on the scale developed by O.G. Frolova and E.I. Nikolaeva (1981) and modified by V.E. Radzinsky et al.[6]

Statistical analysis was performed using the statistical software «Primer of Biostat 4.0» and «STATISTICA 7». Group comparisons with respect to categorical variables are performed using chi-square tests with Yates correction or, alternatively, Fisher's exact test when expected cell counts were less than 5. A probability value of $P<0.05$ was considered statistically significant.

Results and Discussion

Fetometry showed that IUGR was diagnosed in 71/30.1% patients. The degree 1 of IUGR was diagnosed in 49/20.8% pregnant women, and degrees 2 and 3 in 17/7.2% and 5/2.1% pregnant women, respectively. An analysis of the frequency of IUGR revealed significant ($P<0.05$) differences depending on the degree of PR. The main share of women with IUGR comprised those with a high PR in Cohort 1 and those with a high and moderate PR in Cohort 2 (Table 2).

Table 2.

Frequency of IUGR in the examined women

Cohort	Degree of PR	IUGR					
		I		II		III	
		n	%	n	%	n	%
I	Low (n=20)	2	10.0	1	5.0	0	0.0
	Moderate (n=38)	7	18.4	4	5.1	0	0.0
	High (n=70)	21	30.0*	5	7.1	1	1.4
II	Low (n=32)	3	9.3	0	0	0	0.0
	Moderate (n=52)	15	28.8*	2	3.8	0	0.0
	High (n=24)	1	4.2	5	20.8	4	16.7
Total (n=236)		49	20.8	17	7.2	5	2.1

**- P<0.05 in relation to the high PR group of Cohort 2*

Diagnosis of IUGR was confirmed in only 16/22.5% newborns. This situation is explained by the fact that the criteria for ultrasound diagnosis of IUGR differ from clinical diagnosis of IUGR, in accordance with centile scales: body weight, length, and circumference of the head and chest.

More reliably, IUGR was diagnosed in women with a high PR in Cohort 2—in every other newborn born to a mother with an antenatally diagnosed IUGR (Table 3).

Table 3.

Identified IUGR in premature infants at birth

Cohort	Degree of PR	n	IUGR	
			n	%
I	Low (n=20)	3	1	33.3
	Moderate (n=38)	8	2	25
	High (n=70)	12	4	33.3
II	Low (n=32)	17	2	11.8*
	Moderate (n=52)	23	3	13*
	High (n=24)	8	4	50
Total		71	16	22.5

**- P<0.05 in relation to the high PR group of Cohort 2*

The evaluation of the degree of maturity of the placenta according to Grannum et al.[7] obtained the following results: Grade 0 of maturity of the placenta was significantly more often diagnosed in women with a low and moderate PR in Cohort 1 (45% and 36.8%, respectively) and a low PR in Cohort 2 (34.4%). Ultrasonic signs of premature aging of the placenta (Grades II and III) were diagnosed in 87/36.9% of pregnant women ($P<0.05$). In this case, statistical significance was noted in Cohort 2 in all groups with Grade III of maturity of the placenta ($P<0.05$). Grade III of maturity of the placenta was more often diagnosed in women with a high PR in Cohort 2 (Table 4).

Table 4.

Grade of maturity of the placenta

Cohort	Degree of PR	Grade of maturity of the placenta							
		0		I		II		III	
		n	%	n	%	n	%	n	%
I	Low (n=20)	9	45.0*	7	35.0	4	20.0	0	0.0
	Moderate (n=38)	14	36.8*	9	23.7	12	31.6	3	7.9
	High (n=70)	4	5.7	38	54.3*	23	32.9	5	7.1
II	Low (n=32)	11	34.4*	15	46.9	5	15.6	1	3.1*
	Moderate (n=52)	8	15.4	26	50.0	11	21.2	7	13.5*
	High (n=24)	1	4.2	7	29.2	5	20.8	11	45.8
Total (n=236)		47	19.9	102	43.2	60	25.4	27	11.4

**- P<0.05 in relation to the high PR group in each Cohort*

The data obtained when measuring the thickness of the placenta ranged from 31.0 mm to 37.2 mm and averaged 32.4±0.3 mm.

Disorders in the uteroplacental and fetal-placental circulation were detected in 79 (33.5%) of pregnant women (2 and 3 degree in 16[20.2%] women). These disorders were most common in groups of women with a moderate PR (Table 5).

Table 5.

Disorders in the uteroplacental and fetal-placental circulation

Cohort	Degree of PR	n	IA		IB		II		III	
			n	%	n	%	n	%	n	%
I	Low (n=20)	7	4	57.1	3	42.9	0	0.0	0	0.0
	Moderate (n=38)	12	5	41.7	4	33.3	2	16.7	1	8.3
	High (n=70)	11	8	72.7	3	27.3	0	0.0	1	9.1
II	Low (n=32)	18	10	55.6	2	11.1	4	22.2	2	11.1
	Moderate (n=52)	20	13	65.0	7	35.0	0	0.0	0	0.0
	High (n=24)	11	4	36.4	1	9.1	2	18.2	4	36.4
Total (n=236)		79	44	55.7	20	25.3	8	10.1	8	10.1

The pathology of the amniotic fluid was determined in 137/58.1% pregnant women. In structure of the pathology of the amniotic fluid, oligohydramnios was predominant (45.3%). Oligohydramnios was diagnosed 2 to 3 times more often in women with high to moderate PR compared to women with a low PR. Polyhydramnios was found in 30(12.7%) women (Table 6). It should be noted that as the gestational age increased, the frequency of detection of oligohydramnios increased significantly (*P<0.05*).

Table 6.

The pathology of the amniotic fluid

Cohort	Degree of PR	Oligohydramnios		Polyhydramnios	
		n	%	n	%
I	Low (n=20)	4	20.0*	2	10.0
	Moderate (n=38)	14	36.8	7	18.4
	High (n=70)	34	48.6	11	15.7
II	Low (n=32)	7	21.9*/**	5	15.6**
	Moderate (n=52)	32	61.5	1	1.9*
	High (n=24)	16	66.7	4	16.7
Total (n=236)		107	45.3	30	12.7

**- P<0.05 in relation to the high PR group in each Cohort*
***- P<0.05 in relation to the moderate PR group in each Cohort*

Ultrasonic cervicometry, as a promising method for diagnosing and predicting the outcome of PL, was preceded by a determination of the degree of "maturity" of the cervix, according to E. Bishop.[8] The study showed that approximately half (114/51.6%) of the pregnant woman entered the hospital with a "maturing" cervix, of which every third (61/27.6%) woman was in Cohort 1 with a moderate PR. The "immature" cervix was identified in 33/15.0% women in Cohort 1 and 31/14.0% in Cohort 2 with a low PR. Pregnant women in Cohort 1 with the low and moderate PR had an "immature" cervix significantly more often, and, despite the ongoing medical and diagnostic measures, they developed labor activity. The smallest share was presented by women with the "mature" cervix, and of them, 14/40% were in Cohort I with moderate risk and had a degree of maturity of the uterine cervix from 9 to 13 points (Table 7).

Table 7.

Degree of maturity of the uterine cervix, according to E. Bishop

Cohort	Degree of PR	n	"immature" (0-5 points)		"ripening" (6-8 points)		"mature" (9-13 points)	
			n	%	n	%	n	%
I	Low	18	11	61.1*	3	16.7*	4	22.2
	Moderate	35	12	34.3*	9	25.7*	14	40.0*
	High	64	10	15.6	49	76.6	5	7.8
II	Low	28	10	35.7	13	46.4	5	17.9
	Moderate	52	13	25.0	31	59.6	8	15.4
	High	24	8	33.3	9	37.5	7	29.2
Total		221	64	29.0	114	51.6	43	19.5

**- P<0.05 in relation to the high PR group in each Cohort*

The average cervical length was 21.06±6.93 mm, while the maximum cervical length was 28.9 mm, and the minimum length - 13.2 mm. The results of ultrasound data on structural changes in the cervix at threat of PB are presented in Table 8.

Table 8.

Ultrasound data on structural changes in the cervix

Cohort	Degree of PR	n	cervical length > 2.5 cm		cervical length < 2.5 cm		cervical canal width ≥ 1 cm		V-shaped deformation of the internal throat cervix	
			n	%	n	%	n	%	n	%
I	Low	18	4	22.2*	14	77.8*	4	22.2	3	16.7*
	Moderate	35	8	22.9*	27	77.1*	9	25.7	1	2.9*
	High	64	41	64.1	23	35.9	12	18.8	39	60.9
II	Low	28	6	21.4	22	78.6	8	28.6	8	28.6**
	Moderate	52	19	36.5	33	63.5	17	32.7	44	84.6*
	High	24	8	33.3	16	66.7	5	20.8	3	12.5
Total		221	86	38.9	135	61.1	55	24.9	98	44.3

**- P<0.05 in relation to the high PR group in each Cohort*
*** - P<0.05 in relation to the moderate PR group in each Cohort*

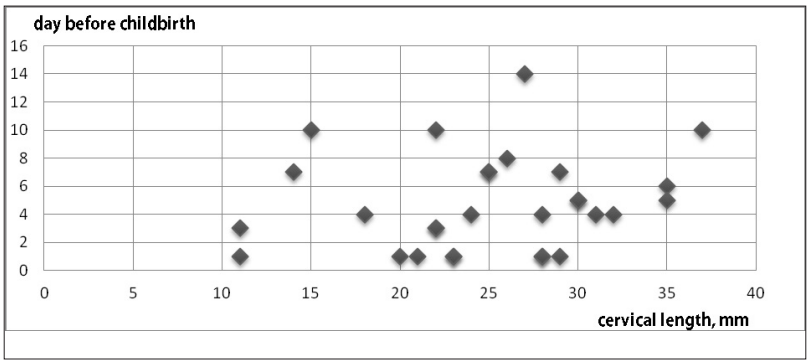

Fig. 1. The cervical length.

It should be noted that the short cervix was seen both for several weeks before delivery and for several days before delivery. Such data were noted in all groups (Figure 1).

Conclusion

The neonatal period carries higher morbidity and mortality than the rest of infancy and childhood combined. Two important measures associated with neonatal outcomes are gestational age and birthweight.[9,10] In accordance with ICD-10, newborns classified as "small for gestational age" are those who are smaller in size than normal for the gestational age, most commonly defined as a weight and length below the 10th percentile for the gestational age.[11,12] Currently, the analysis of publications shows an absence of generalized data on the features of the course of pregnancy and childbirth in cases of intrauterine growth retardation and premature birth. There are certain difficulties in the accurate and objective diagnosis of placental insufficiency, and there are no clear criteria for evaluating its various forms. This leads to the possibility of overdiagnosis of the decompensated form of placental insufficiency and hypodiagnosis of the compensated form of placental insufficiency.

Fetoplacental insufficiency averaged 39.8%. Intrauterine growth retardation was significantly ($P<0.05$) more often diagnosed in pregnant women with moderate and high perinatal risk regardless of the analyzed gestational periods—an average in 32.6% of women with premature birth. In pregnant women with a low perinatal risk, intrauterine growth retardation was diagnosed reliably less often—in 8% of cases. We found that intrauterine growth retardation was diagnosed in every third (30.1%) examined pregnant woman. However, the diagnosis of intrauterine growth retardation was confirmed in only 16(22.5%) newborns according to centile scales. A weak direct relationship between oligohydramnios and intrauterine growth retardation in Cohort 1 (r=0.364, $P<0.05$) and in Cohort 2 (r=0.39, $P<0.001$) was revealed.

Disorders in the uteroplacental and fetal-placental circulation were detected in 79(33.5%) of pregnant women (2 and 3 degree in 16[20.2%] women). These disorders were most common in groups of women with a moderate perinatal risk. The amniotic fluid pathology was determined in 137(58.1%) pregnant women. In structure of the amniotic fluid pathology, oligohydramnios was predominant (45.3%). Oligohydramnios was diagnosed 2 to 3 times more often in women with high to moderate perinatal risk compared to women with a low perinatal risk. It should be noted that as the gestational age increased, the frequency of detection of oligohydramnios increased significantly ($P<0.05$).

The results of assessing the state of the cervix clearly demonstrate the lack of effectiveness of tocolytic therapy in 51.6% of pregnant women (cervical ripening). In the remaining pregnant women with an "immature" cervix, cervicometry revealed istmiko-cervical insufficiency, that is, already at the time of 16-19 weeks of gestation the mechanism of anatomical transformation in the cervix was started, not diagnosed in time.

Competing interests

The authors declare that they have no competing interests.

Sources of Funding

The study was supported by the RUDN University Program 5-100.

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International Journal of Biomedicine 8(1) (2018) 47-50
http://dx.doi.org/10.21103/Article8(1)_OA7

ORIGINAL ARTICLE

Children's Health

INTERNATIONAL JOURNAL OF BIOMEDICINE

Evolution of the Structure of Children's Morbidity Rate in the Republic of Sakha (Yakutia)

Tatyana E. Burtseva, PhD, ScD[1,2*]; Sardana A. Evseeva[2]; Maya S. Savvina, PhD[2]; Svetlana Y.Yakovleva, PhD[2]; Vyacheslav G. Chasnyk, PhD, ScD[3]

[1]*M. K. Ammosov North-Eastern Federal University, Yakutsk, Russia*
[2]*Yakut Science Center of Complex Medical Problems, Yakutsk, Russia*
[3]*Saint Petersburg State Pediatric Medical University, St. Petersburg, Russia*

Abstract

Currently, the comparative studies of human health status definitely show that the health level of a population has a regional specificity. This paper analyzes the data of official medical statistics on morbidity among the child population in dynamics from 2000 to 2016 and presents the evolution of the children's incidence structure in the Republic of Sakha (Yakutia) (RS(Y)), according to ICD-10.**(International Journal of Biomedicine. 2018;8(1):47-50.)**

Key Words: children's incidence • children • adolescents • Yakutia

Introduction

In order to improve child protective measures, the years 2018-2027 in Russia were declared as the Decade of Childhood. At the same time, the regions of Russia differ greatly in their socioeconomic, climatogeographic, and medico-demographic parameters, and in their medical care structure, communication, and transport development, all of which definitely affects the costs and effectiveness of the health care system.

The impact of extreme climatogeographic and socioeconomic features in the North is reflected in the unfavorable dynamic changes in the health of the younger generation.[1-6] The indicators of the overall incidence among children in the regions of the Far North for some classes of diseases exceed the average federal level by approximately 1.5-2 times.[1,3]

The aim of this study was to analyze the data of official medical statistics on morbidity among the child population in dynamics from 2000 to 2016 and to present the evolution of the children's incidence structure in RS(Y), according to ICD-10.

***Corresponding author**: Tatiana E. Burtseva, PhD, ScD. M.K. Ammosov North-Eastern Federal University, Yakutsk, Russia. E-mail: bourtsevat@yandex.ru*

Materials and Methods

We analyzed the data of the official statistics of RS(Y) on the child incidence in dynamics from 2000 to 2016. We calculated the increase and decrease in morbidity (in percent) in order to present the evolution of children's incidence structure in the RS(Y) for 17 years.

Results

On 01/01/16, the population of RS(Y) numbered 959,600 people, including 258,200 children and adolescents. Thus, the share of children and adolescents was 27%. The demographic situation in RS(Y) at the end of 2016 was characterized by the stable rates of birth (16‰), total mortality (8.4%) and natural increase (7.6%). The infant mortality rate for the period 2000-2016 exceeded the all-Russian rate. However, it had decreased by 2.5 times in dynamics since 2000 and reached a historic low of 6.8 % in 2016. There were no maternal deaths for the first time in 2016.

The total and primary child incidence in RS(Y) remains at a high level, exceeding the average rate for the Russian Federation, and grows annually, which is associated with both a real deterioration in the health status of children and adolescents, and with an increase in the availability of medical services and the quality of medical examinations.

The total children's incidence in 2016 was 2774.4 per 1,000 children (0-14 years) compared to 2773.3 per 1,000 in 2015. Table 1 shows the significant changes in total children's incidence in SR(Y) in 2000-2017. In general, the morbidity rate had risen by 70.8%. The most significant increase was found in the following disease classes: neoplasm (229.5%), the musculoskeletal system diseases (127.8%), the digestive system diseases (123.3%), and congenital anomalies (122.1%). The reduction in this indicator was noted for such disease classes as: endocrine system diseases, infectious and parasitic diseases, mental and behavioural disorders.

Table 1.

The total children's incidence in SR(Y) (per 1,000 children of 0-14 years)

Disease classes ICD-10	2000	2016	Rate of increase/ decrease,%
Total	1623.6	2774.4	+70.8
Certain infectious and parasitic diseases	105.8	74.7	-29.4
Neoplasms	4.4	14.5	+229.5
Diseases of the blood and blood-forming organs and certain disorders involving the immune mechanism	19.3	21.1	+9.3
Endocrine. nutritional and metabolic diseases	43.5	25.6	-41.2
Mental and behavioural disorders	18.6	13.3	-28.5
Diseases of the nervous system	88.7	126.4	+42.5
Diseases of the eye and adnexa	79.5	127.8	+60.7
Diseases of the ear and mastoid process	29.0	51.3	+76.9
Diseases of the circulatory system	6.9	13.5	+95.6
Diseases of the respiratory system	897.7	1724.3	+92.0
Diseases of the digestive system	95.6	213.5	+123.3
Diseases of the skin and subcutaneous tissue	80.8	123.7	+53.1
Diseases of the musculoskeletal system and connective tissue	16.9	38.5	+127.8
Diseases of the genitourinary system	29.0	49.9	+72.0
Congenital malformations. deformations and chromosomal abnormalities	14.0	31.1	+122.1
Symptoms. signs and abnormal clinical and laboratory findings. not elsewhere classified	0	82.2	…
Injury, poisoning and certain other consequences of external causes	59.3	106.3	+79.2

The primary child morbidity rate increased in 2016 by 5.4%, amounting to 2338.3 per 1,000 children (vs. 2218.5 in 2015). During the study period, this rate increased by 73.3%. We noted the greatest increase for such disease classes as congenital malformations (276.0%), neoplasms (187.5%), respiratory diseases (98.8%), digestive system diseases (93.3%), genitourinary system diseases (75,8%), the musculoskeletal system diseases (61.7%), and nervous system diseases (61.7%) on the background of a decrease in such disease classes as endocrine system diseases, mental and behavioural disorders, infectious and parasitic diseases, and blood and blood-forming organ diseases.

Table 2.

The primary children's incidence in SR(Y) (per 1.000 children of 0-14 years)

Disease classes ICD-10	2000	2016	Rate of increase/ decrease. %
Total	1348.7	2338.3	+73.3
Certain infectious and parasitic diseases	86.7	67.0	-22.7
Neoplasms	2.4	6.9	+187.5
Diseases of the blood and blood-forming organs and certain disorders involving the immune mechanism	10.3	8.1	-21.3
Endocrine. nutritional and metabolic diseases	19.5	6.8	-65.1
Mental and behavioural disorders	4.8	2.1	-56.2
Diseases of the nervous system	24.8	40.1	+61.7
Diseases of the eye and adnexa	50.8	70.0	+37.8
Diseases of the ear and mastoid process	31.5	40.3	+27.9
Diseases of the circulatory system	4.3	6.2	+44.1
Diseases of the respiratory system	831.0	1653.0	+98.9
Diseases of the digestive system	77.9	150.6	+93.3
Diseases of the skin and subcutaneous tissue	71.3	96.3	+35.1
Diseases of the musculoskeletal system and connective tissue	16.2	26.2	+61.7
Diseases of the genitourinary system	17.4	30.6	+75.8
Congenital malformations. deformations and chromosomal abnormalities	2.5	9.4	+276.0
Symptoms. signs and abnormal clinical and laboratory findings. not elsewhere classified	…	0.2	…
Injury. poisoning and certain other consequences of external causes	67.4	106.2	+57.5

The total adolescent morbidity rate decreased in 2016 by 4.6% from 2390.8 to 2280.7 per 1,000 adolescents. The Table 3 shows that the total incidence rate in adolescents had no significant changes in 2000-2016. We noted the most significant increase for such disease classes as neoplasms (358.3%), the musculoskeletal system disease (169.7%), respiratory diseases

(164.4%), ear and mastoid disease (163%), on the background of a decrease in such disease classes as nervous system diseases, infectious and parasitic diseases, diseases of the endocrine system.

Table 3.

The total adolescent morbidity rate in RS(Y)c (per 1000 adolescents)

Disease classes ICD-10	2000	2016	Rate of increase/ decrease. %
Total	1089.8	2280.7	+109.2
Certain infectious and parasitic diseases	42.6	36.9	-13.3
Neoplasms	2.4	11.0	+358.3
Diseases of the blood and blood-forming organs and certain disorders involving the immune mechanism	…	27.6	…
Endocrine. nutritional and metabolic diseases	66.7	62.4	-6.4
Mental and behavioural disorders	34.7	33.5	-3.4
Diseases of the nervous system	222.7	168.4	-24.3
Diseases of the eye and adnexa	…	210.1	…
Diseases of the ear and mastoid process	14.6	38.4	+163.0
Diseases of the circulatory system	21.8	37.6	+72.4
Diseases of the respiratory system	346.1	915.3	+164.4
Diseases of the digestive system	103.7	242.0	+133.3
Diseases of the skin and subcutaneous tissue	58.9	133.8	+127.1
Diseases of the musculoskeletal system and connective tissue	27.8	75.0	+169.7
Diseases of the genitourinary system	58.4	70.32	+20.3
Congenital malformations. deformations and chromosomal abnormalities	13.1	17.3	+32.0
Symptoms. signs and abnormal clinical and laboratory findings. not elsewhere classified	…	0.1	…
Injury. poisoning and certain other consequences of external causes	76.3	190.5	+149.6

The primary incidence rate in adolescents in 2016 was reduced to 2%. The rate of primary incidence among adolescents increased in 2000-2016 by 94% (Table 4).

The reduction in the primary incidence was noted for such diseases as mental and behavioral disorders, congenital anomalies. We noted the most significant increase for such disease classes as the digestive system diseases (169.5%), injury and poisoning (141.5%), the respiratory system diseases (138.6%), the nervous system diseases (120.4%), the musculoskeletal system diseases (112.4%), diseases of the circulatory system (112.9%).

Table 4.

The primary incidence rate in adolescents in SR(Y) (per 1000 adolescents)

Disease classes ICD-10	2000	2016	Rate of increase/ decrease. %
Total	784.5	1522.1	+94.0
Certain infectious and parasitic diseases	35.4	29.2	+17.5
Neoplasms	2.0	3.3	+65.0
Diseases of the blood and blood-forming organs and certain disorders involving the immune mechanism	8.6	9.2	+6.9
Endocrine. nutritional and metabolic diseases	32.8	12.1	+63.1
Mental and behavioural disorders	20.2	6.0	-70.3
Diseases of the nervous system	25.4	56.0	+120.4
Diseases of the eye and adnexa	49.6	60.6	+22.1
Diseases of the ear and mastoid process	15.1	23.9	+58.2
Diseases of the circulatory system	7.7	16.4	+112.9
Diseases of the respiratory system	343.8	820.4	+138.6
Diseases of the digestive system	49.3	132.9	+169.5
Diseases of the skin and subcutaneous tissue	53.6	81.8	+52.6
Diseases of the musculoskeletal system and connective tissue	16.9	35.9	+112.4
Diseases of the genitourinary system	30.7	35.4	+15.3
Congenital malformations. deformations and chromosomal abnormalities	13.4	1.5	-88.8
Symptoms. signs and abnormal clinical and laboratory findings. not elsewhere classified	2.7	0.02	-99.2
Injury. poisoning and certain other consequences of external causes	78.7	190.1	+141.5

Conclusion

In recent years, there has been an annual reduction in child population in RS(Y) due to migration and a decrease in the birth rate. So, 301,800 children (from 0 to 17 years) lived in RS(Y) in 2000, but the number of children was reduced to 258,200 by 2016. It is obvious that the general and primary incidence in the child and adolescent population is increasing in RS(Y). The structure of the children and adolescent incidence has changed significantly. The primary incidence of infectious and parasitic diseases has clearly decreased due to widespread vaccination efforts against controlled infections. The increase in the primary incidence of congenital anomalies and neoplasms in child population is very worrying. The difficult climatic conditions of this northern territory cause a high primary incidence among children of respiratory diseases and genitourinary system diseases. In addition, changing the

traditional diet has led to an increase in digestive system diseases. All this dictates the necessity for a complex and interdepartmental approach to strengthening the health of the younger generation in RS(Y).

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 51-55
http://dx.doi.org/10.21103/Article8(1)_OA8

ORIGINAL ARTICLE

Children's Health

INTERNATIONAL JOURNAL OF BIOMEDICINE

Drug-Resistant Tuberculosis in Children and Adolescents in the Republic of Sakha (Yakutia)

Evdokiya F. Luginova, PhD*

Yakut NPZ "Ftiziatriia", Yakutsk, the Sakha Republic, Russia

Abstract

The aim of the study was to determine the prevalence of drug-resistant tuberculosis among children and adolescents in the Republic of Sakha (Yakutia) (RS(Y)) and the features of its clinical manifestation, in order to improve the prevention and treatment of the disease.

We monitored a total of 250 children and adolescents with tuberculosis (TB) who underwent inpatient treatment in the Yakut NPZ in 2012-2016: The patients included 34(13.6%) children in the 0-2 age group, 80(32%) in the 3-6 age group, 74(29.6%) in the 7-14 age group, and 62(24.8%) adolescents between 15 and 17 years. The timing of the development of the disease from the time of primary infection with *Mycobacterium tuberculosis* differed significantly depending on the age of children. In young children, compared with older children, signs of the disease appeared at an earlier time. In the structure of clinical forms of TB, the differences were observed depending on the age of the children:

- In children under 2 years, the lesion of intrathoracic lymph node tuberculosis (ITLN) was most common, as well as generalized TB
- For children aged between 3 and 6 years, the lesion of ITLN was characteristic
- For older children, the different clinical forms of TB were characteristic.

The expression of immunological tests depended on the age of the patients and the number of ITLN lesions. Drug resistance to anti-tuberculosis drugs was found in 21(36.8%) patients; *m*ultidrug-resistant TB was mainly registered in adolescents and multidrug resistance was mainly primary. Drug resistance to isoniazid, rifampicin, and streptomycin was the most frequent.

Thus, it has been established that the outcome of the disease and the timing of the onset of clinical involution of the disease depend on many factors, including the timeliness of disease detection, the age of the patient, the dissemination of TB process, and others. **(International Journal of Biomedicine. 2018;8(1):51-55.)**

Key Words: children • adolescents • drug-resistant tuberculosis • anti-tuberculosis drugs

Abbreviations

ATD, anti-tuberculosis drugs; **DR**, drug resistance; **DRT**, drug-resistance test; **DR-TB**, drug-resistant tuberculosis; **ITLN**, intrathoracic lymph nodes; **ITLNTB**, intrathoracic lymph node tuberculosis; **MTB**, *Mycobacterium tuberculosis*; **MDR**, multidrug resistance; **MDR-TB**, multidrug-resistant TB; **MT**, Mantoux test; **PTC**, primary tuberculosis complex; **RA**, recombinant allergen; **TB**, tuberculosis.

Introduction

Tuberculosis (TB) is one of the top 10 causes of death worldwide. In 2016, 10.4 million people fell ill with TB, and 1.7 million died from the disease (including 0.4 million among people with HIV).[1] Children represent about 10-11% of all TB cases.

At least 1 million children become ill with TB each year.[2] In 2016, 250 000 children died of TB.[1] The actual burden of TB in children is likely higher given the challenge in diagnosing childhood TB.

Multidrug-resistant TB (MDR-TB) remains a public health crisis and a health security threat. MDR-TB is TB that does not respond to at least isoniazid and rifampicin, the 2

***Corresponding author*: *Evdokiya F. Luginova, PhD.. Yakut NPZ "Ftiziatriia", Yakutsk, the Sakha Republic, Russia. E-mail: Luginovaef@mail.ru*

most powerful anti-TB drugs. In 2015, an estimated 480 000 people worldwide developed MDR-TB, and an additional 100 000 people with rifampicin-resistant TB were also newly eligible for MDR-TB treatment. India, China, and the Russian Federation accounted for 45% of the 580 000 cases.[3] In 2016, WHO estimates that there were 600,000 new cases with resistance to rifampicin - the most effective first-line drug, of which 490,000 had MDR-TB.[1]

Globally,[1] TB incidence is falling at about 2% per year. However, in Russia, the prevalence of MDR-TB of the respiratory organs continues to grow. Whereas in 1999 this indicator was 8.6, in 2015 it was 25.5 per 100 thousand of the population. In the Republic of Sakha (Yakutia) (RS(Y)), according to the official statistical reporting, there has been a clear growth trend in the proportion of patients with MDR-TB among newly diagnosed pulmonary TB patients with bacterial excretion: 23.0% in 2011, 25.8% in 2013, 30.0% in 2015, and 34.6% in 2016. M.K.Vimokurova et al.[4] have observed in RS(Y) a growth in the incidence of TB cases presenting with destructions in lung tissue and a bacillary-positive state, a persistent trend towards an increase in TB cases caused by MDR MTB, and a high proportion of deaths during the first year of outpatient follow-up for TB.

The effectiveness of treatment of patients with drug-resistant strains of MTB is much lower than in patients with drug-susceptible MTB. In this regard, in these patients there is a persistent bacteria discharge.[5-8] The increase in the number of patients with DR-TB leads to an increased risk of a further spread of infection and the emergence of new cases of disease with primary DR.[9] For children and adolescents, DR-TB represents the greatest danger.[10,11]

The aim of the study was to determine the prevalence of DR-TB among children and adolescents in RS(Y) and the features of its clinical manifestation, in order to improve the prevention and treatment of the disease.

Material and Methods

We monitored a total of 250 children and adolescents with TB who underwent inpatient treatment in the Yakut NPZ "Ftiziatriia" in 2012-2016: The patients included 34(13.6%) children in the 0-2 age group, 80(32%) in the 3-6 age group, 74(29.6%) in the 7-14 age group, and 62(24.8%) adolescents between 15 and 17 years. TB contacts were identified in 78.7% of cases (including 98% cases among children in the 0-2 age group, in 86.7% cases among children in the 3-6 age group, in 74.6% among children in the 7-14 age group, and 64.3% of cases among adolescents between 15 and 17 years).

All patients underwent clinical, laboratory, and radiation study methods according to the Federal Clinical Recommendations for Diagnosis and Treatment of Respiratory Tuberculosis in Children.[8]

In the diagnosis of TB, we used chest X-ray, sputum examination on MTB by fluorescence microscopy, culture in both liquid and solid media, and bronchial washing for AFB staining. Immunological tests included the 2 tuberculin unit MT and a skin test with MTB-RA. PCR was used for detecting drug resistance. Patients were treated according to standard regimens in accordance with the Federal Clinical Recommendations for Diagnosis and Treatment of Respiratory Tuberculosis in Children, taking into account the results of drug-susceptibility testing.

Results were statistically processed using Microsoft Office Excel 2007. Baseline characteristics were summarized as frequencies and percentages for categorical variables and as means and SDs for continuous variables. For data with normal distribution, inter-group comparisons were performed using Student's t-test. The Mann-Whitney U Test was used to compare the differences between the two independent groups (for nonparametric data). A probability value of $P<0.05$ was considered statistically significant.

Results

In the structure of clinical forms of TB in children under 2 years of age, ITLNTB was identified in 67.6% of cases, PTC in 26.4%, and generalized TB in 6.0% of cases. In children aged between 3 and 6 years, ITLNTB was detected most frequently (66.2%), and PTC in 33.7% of cases. In children of school age and adolescents, there were different clinical forms of TB. Thus, in children aged between 7 and 14 years, ITLNTB was detected in 48.6% of cases, PTC in 28.4%, infiltrative pulmonary TB in 10.8%, focal pulmonary TB in 8.1% of cases, disseminated TB in one case, and caseous pneumonia and generalized TB in one case. In adolescents (between 15 and 17 years), infiltrative (40.3%) and focal (33.9%) pulmonary TB were most often recorded, but we also diagnosed disseminated TB(11.3%), ITLNTB(6.5%), and PTC(3.2%). In addition, in this age group, one case of caseous pneumonia, one case of tuberculoma and one case of generalized TB were identified.

Thus, in the structure of clinical forms of TB, the differences were observed depending on the age of the children. Thus, in children under 2 years, the lesion of ITLN was most common, as well as generalized TB. For children aged between 3 and 6 years, the lesion of ITLN was characteristic; For older children, the different clinical forms of TB were characteristic.

The average time of onset of the disease manifestations after the appearance of signs of primary infection in children (for the first time a positive MT, not related to vaccination and revaccination of BCG) differed significantly depending on the age. The earliest terms for the appearance of signs of TB from the time of primary infection were observed in young children: 1.5±0.1 months for children aged between 0 and 2 years, and 4.1±0.7 months for children aged between 3 and 6 years. In older children, signs of the disease appeared in later terms: 11.4±1.6 months and 34.1±7.1 months for the 7-14 and 15-17 age groups, respectively.

We also found that in the 15-17 age group the timing of the onset of the disease from the first positive MT differed significantly depending on the clinical forms of TB. Thus, primary forms of TB developed after 17.5±1.7 months since the appearance of primary TB infection, and secondary forms after 38.8±8.4 months ($P<0.05$).

Thus, the timing of the development of the disease from the time of primary infection with MTB differed significantly depending on the age of children. In young children, compared

with older children, signs of the disease appeared at an earlier time.

We conducted a comparative analysis of the degree of involvement of ITLN by the number of groups of lymph nodes involved in the tuberculosis process in children of different ages. For example, ITLNTB was observed in 23(67.6%), 53(66.2%) and 36(48.6%) cases among children between 0-2 years, 3-6 years, and 7-14 years, respectively. Thus, TB of one group of ITLN was observed in 6(26.1%), 10(18.8%), and 7(19.4%) cases among children from 0-2 years, 3-6 years, and 7-14 years, respectively. TB of 2 groups of ITLN was observed in 6(26.1%), 18(34%), and 15(41.7%) cases among children from 0-2 years, 3-6 years, and 7-14 years, respectively. TB of 3 groups and more than 4 groups of ITLN was observed in 7(30.4%) and 4(17.4%), 18(34%) and 7(13.2%), 5(13.9%) and 9(25%) cases among children from 0-2 years, 3-6 years, and 7-14 years, respectively.

Thus, the above data indicate that in children at different ages with TB, 2-3 groups of ITLN are mainly affected. The lesion of more than 4 groups of ITLN was observed more often in children aged from 0-2 and 7-14 years: 17.4 and 25%, respectively.

The expression of immunological tests, depending on the number of ITLN lesions in different age groups, is presented in Table 1. In children aged between 0 and 2 years with a lesion of more than 4 groups of ITLN, there was a high immune response simultaneously in MT and MTB-RA. In children between 3 and 6 years, the immune response in MT and MTB-RA did not differ significantly and did not have a clear dependence on the severity of ITLNTB, but with the lesion of 4 or more groups of ITLNT, both immunological tests had a hyperergic character. In children aged between 7 and 14 years with ITLNTB of any degree, the test with MTB-RA had a hyperergic response, and the severity of reaction to MT was less pronounced with a lesion of one group of ITLN.

In the 15-17 age group, we examined the severity of immunological tests depending on the clinical forms of TB. It was found that in patients with primary forms of TB (ITLNTB and PTC), the average size of the papule in MT was 16.9±1.6 mm; in the skin test with MTB-RA it was 17.7±2.0 mm ($P>0.05$). In patients with secondary forms of TB (focal, infiltrative, and disseminated TB), the average size of the papule in MT was 13.0±0.8 mm; In the skin test with MTB-RA it was 16.9±0.7 mm ($P<0.05$). Data for immune response in MT and the skin test with MTB-RA, depending on the clinical forms of secondary TB in patients aged between 15 and 17 years, are presented in Table 2. The immune response in the test with MTB-RA was more pronounced than in MT. It should be noted that in the primary forms of TB, in contrast to secondary forms, a hyperergic reaction was simultaneously observed in MT and the test with MTB-RA. In the infiltrative and disseminated forms of TB, the degree of immune response in the test with MTB-RA was significantly higher than in MT. The average time of appearance of the first signs of the disease from the time of primary TB infection in patients aged between 15 and 17 years with primary forms of TB was 17.5±1.7 months, with secondary forms it was 38.8± 8.4 months ($P<0.05$).

Patients with MTB discharge accounted for 24.4%. Among children between 0 and 2 years, 3 and 6 years, 7 and 14 years, and 15 and 17 years, MTB discharge was determined in 2(3.3%), 1(1.6%), 12(19.7%), and 46(75.4%) cases, respectively.

DRT was performed in 57(93.4%) patients. It was found that drug sensitivity to all ATD was preserved in 36(63.2%) patients. DR to ATD was found in 21(36.8%) patients: 2(10%) in the 0-2 age group, 4(19%) in the 7-14 age group, and 15(71.4%) in the 15-17 age group. Monoresistance was noted only in 1(4.7%), MDR in 95.2%.

Table 1.

The expression of immunological tests depending on the number of ITLN lesions in different age groups

Immunological tests	Number of ITLN lesions			
	Children aged between 0 and 2 years (n=23)			
	1 group of ITLN (n=6)	2 groups of ITLN (n=6)	3 groups of ITLN (n=7)	4 or more groups of ITLN (n=4)
MT (mm)	11.9±1.7	13.0±0.9	14.4±1.3	10.5±1.2*
Skin test with MTB-RA (mm)	10.7±2.9	14.2±0.8	15.1±1.4	14.7±2.1
Children aged between 3 and 6 years (n=53)				
Immunological tests	1 group of ITLN (n=10)	2 groups of ITLN (n=18)	3 groups of ITLN (n=18)	4 or more groups of ITLN (n=7)
MT (mm)	13.9±1.1*	13.4±0.8	13.4±0.9	17.0±4.0
Skin test with MTB-RA (mm)	16.7±0.1	16.6±1.6	15.3±1.3	18.0±2.0
Children aged between 7 and 14 лет (n=36)				
Immunological tests	1 group of ITLN (n=7)	2 groups of ITLN (n=15)	3 groups of ITLN (n=5)	4 or more groups of ITLN (n=9)
MT (mm)	10.3±1.6*	16.6±1.6	16.8±1.7	14.7±1.0
Skin test with MTB-RA (mm)	17.5±1.8	17.5±1.5	20.8±1.1	17.3±3.6

*- $P<0.05$

Table 2

The expression of immunological tests depending on the clinical forms of TB in the 15-17 age group

Immunological tests	Focal TB (n=21)	Infiltrative TB (n=25)	Disseminated TB (n=7)	Primary forms of TB (n=6)
MT (mm)	14.1±1.2	13.1±1.3*	9.9±1.1*	16.9±1.6
Skin test with MTB-RA (mm)	16.7±1.0	18.0±1.2	15.3±1.2	17.6±2.0

**-P<0.05*

DR to isoniazid, rifampicin, and streptomycin was the most frequent: 95.2%, 81% and 76.2% of cases, respectively. DR to kanamycin, capreomycin and ethionamide was found in 2(10%), 1(4.7%), and 2(10%) cases, respectively. DR to the combination of isoniazid with streptomycin was found in 31.2% of cases, isoniazid with streptomycin and isoniazid with rifampicin in 8.3% of cases; other combinations of ATD were more rare. It should be noted that only 3(15%) out of 20 children with DR to isoniazid previously received this drug for preventive purposes. Other drugs have not been received by any child, which indicates that the children had primary DR to ATD, associated with drug-resistant strains of MTB.

MDR to ATD was observed in 17(27.8%) patients with MTB discharge, including 2(11.8%) children under the age of 2 years, 1(5.9%) in the 7-14 age group, and 14(82.3%) in the 15-17 age group. Thus, MDR-TB was mainly registered in adolescents and MDR was mainly primary.

It has been established that the outcome of the disease and the timing of the onset of clinical involution of the disease depend on many factors, including the timeliness of disease detection, the age of the patient, the dissemination of TB process, and others.

In patients with the multidrug-resistant strains of MTB, there were some features of the clinical course of the disease: the rapid onset of the disease (68%); the severity of symptoms of tuberculous intoxication(43%); a slow rate of decreasing the severity of the reaction to MT and MTB-RA; a decrease in the rate of resorption of the infiltrative changes, sputum negativization and elimination of destructive changes in the lungs; a high rate of pronounced residual changes (39%); a greater need for surgical treatment; and a high risk of recurrence of the disease (5.6%).

The duration of activity and the inadequacy of the healing process in MDR-TB were indicated by the presence of MTB growth in the biopsy material and the histological picture of the resected part of the lung in 5 postoperative patients. In 4 out of 5 postoperative patients, we noted the growth of drug-resistant strains of MTB. In all 5 patients, in a lung biopsy during surgery, MTB was found by fluorescence microscopy. In a histological examination of the resected regions of the lungs in 3 of the patients, we found the morphological picture of active TB: the presence of tuberculosis granulomas, large zones of productive tissue, and extensive areas of caseous necrosis. To reduce the risk of recurrence of the disease in the postoperative period, we performed a correction of chemotherapy, taking into account the drug resistance of MTB.

Conclusion

The timing of the development of the disease from the time of primary infection with MTB differed significantly depending on the age of children. In young children, compared with older children, signs of the disease appeared at an earlier time. In the structure of clinical forms of TB, the differences were observed depending on the age of the children:

- In children under 2 years, the lesion of ITLN was most common, as well as generalized TB
- For children aged between 3 and 6 years, the lesion of ITLN was characteristic
- For older children, the different clinical forms of TB were characteristic.

The expression of immunological tests depended on the age of the patients and the number of ITLN lesions. DR to ATD was found in 21(36.8%) patients; MDR-TB was mainly registered in adolescents and MDR was mainly primary. DR to isoniazid, rifampicin, and streptomycin was the most frequent.

Thus, it has been established that the outcome of the disease and the timing of the onset of clinical involution of the disease depend on many factors, including the timeliness of disease detection, the age of the patient, the dissemination of TB process, and others.

Worldwide, only 54% of MDR-TB patients are currently successfully treated. In 2016,[1] WHO approved the use of a short, standardized regimen for MDR-TB patients who do not have strains that are resistant to second-line TB medicines. Improving the prevention and treatment of tuberculosis in children and adolescents requires a joint effort by all stakeholders involved in TB control.

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 56-59
http://dx.doi.org/10.21103/Article8(1)_OA9

ORIGINAL ARTICLE

Environmental Health

INTERNATIONAL JOURNAL OF BIOMEDICINE

Assessment of the Space Weather Effect on Human Health in the Arctic Zone Using the Example of Tiksi Settlement

Alena A. Strekalovskaya, PhD[1*]; Palmira P. Petrova, PhD, ScD[1]; Kirill V. Komzin[1]; Larisa A. Malysheva, PhD[1]; Sergey N. Samsonov, PhD[2]; Svetlana S. Parshina, PhD, ScD[3]; Alla A. Andreeva[4]

[1]*North-Eastern Federal University named after MK Ammosov, Yakutsk, the Republic of Sakha (Yakutia), Russia*
[2]*Institute of Cosmophysic Researches and Aeronomy named after Yu.G. Shafer, Siberian Branch of the RAS, Yakutsk, the Republic of Sakha (Yakutia), Russia*
[3]*Medical University named after V.I. Razumovsky, Saratov, Russia*
[4]*Bulunsky Central District Hospital, Tiksi, the Republic of Sakha (Yakutia), Russia*

Abstract

In order to assess the space weather effect on the well-being and health of people with cardiovascular pathology in Arctic conditions, we carried out the processing and analysis of space weather parameters and the electronic database of patients with cardiovascular diseases at the Central District Hospital in Tiksi settlement (the Republic of Sakha (Yakutia) (RS(Y)). Patients visited the polyclinic or requested an ambulance because their health had deteriorated. As a result of our research, we found some conjunctions of trends in the change in geomagnetic disturbances (Kp-index) and the number of patients' visits to medical institutions for arterial hypertension (AH) in 2015, 2016 and 2017. It can therefore be concluded that geomagnetic disturbances have an impact on the cardiovascular system of a person living at high latitudes.**(International Journal of Biomedicine. 2018;8(1):56-59.)**

Key Words: cardiovascular diseases • arterial hypertension • space weather • geomagnetic activity • Arctic zone

Introduction

The effect of the environment on human health is one of the key issues at the present time. Apart from the obvious effects of certain environmental factors, such as meteorological parameters and social factors, it has been found that electromagnetic radiation, produced by electric and magnetic fields, which are not observed by human senses, can also have an effect on people's health. At the end of the last century, after the first flight of a spacecraft that measured the parameters of particle flows, electromagnetic fields, and radiation, there appeared an understanding of the interplanetary environment and how the fields, particles and radiation can reach a particular area of the interplanetary environment and have an effect on life on Earth. Moreover, at the beginning of our century the term "space weather" appeared. It refers to the interplanetary environmental conditions. The sun has the main impact on the interplanetary environment. Therefore, the identification of link mechanisms between solar activity and the functioning of the various objects of the biosphere, including humans, is one of the fundamental problems of modern science.

There are many published works about the effects of space weather on human health.[1-19] However, there are still many open questions about the specific parameters of space weather affecting the human body, and about the mechanisms of such influence.

At present, the pathogenesis of AH is being actively studied by the world's leading institutions. The Arctic zone is located in the territory most affected by the heliogeophysical factors of space weather because of its geophysical and atmospheric features. Within the framework of this research, it was planned to determine the presence or absence of a link between episodes of increasing blood pressure in AH patients and the periods of helio-geomagnetic agitations.

Materials and Methods

***Corresponding author:** Alena A. Strekalovskaya, PhD. M. K. Ammosov North-Eastern Federal University. Yakutsk, the Republic of Sakha (Yakutia), Russia. E-mail: a_strekalovskaya@mail.ru*

To research the dynamics of health deterioration among people having cardiovascular diseases living in high latitudes, we studied the electronic database of patients' visits to the medical institutions of Tiksi (the Bulunsky district of RS(Y)) between 2015 and 2017.

To define the reasons influencing the deterioration of health among patients with heart and vessel failures, we compared the visits for AH with the Kp-index (planetary index of geomagnetic disturbance), which is a measure of the level of geomagnetic storminess. For this purpose, we used the Kp-index and the average monthly number of visits for AH to the medical institutions of Tiksi. The Kp-index reflects the average planet geomagnetic field deviation. It is measured in conditional units from 1 to 9. The index rises as the intensity rises. For the statistical processing and plotting of data, we used the computer program Origin.

Results and Discussion

To analyze the effect of heliogeophysical factors on the health of people living in Tiksi settlement, the data from the ambulance and polyclinic service of the Bulunsky Central District Hospital were selected. For comparison, we chose the cases of deterioration in the well-being of people due to an increase in blood pressure in 2015, 2016 and the first 6 months of 2017. We then analyzed patients' visits and compared them with the Kp-index for the same period of time (2015, 2016 and the first 6 months of 2017). In general, during these years we can observe a reduction in the 11-year cycle of solar activity and a general decrease in geomagnetic disturbance.

The analysis of data on the number of cases of AH and the comparison with the Kp-index showed the following. In 2015, there were 2,109 visits from patients diagnosed with AH, 1,416 of them were women, and 693 men. Out of the total number of visits, 1,116/53% people were registered as dispensary for hypertension. In most cases (65%), AH was observed in the elderly, 60 and older, which is quite logical, since the pathogenetic factors having an effect on the development of hypertensic crisis in the elderly are high: blood-circulating angiotensin II, damage to the vascular endothelium and a decrease in the allocation of vasodilating substances. Patients of middle age (<60 years) constituted 740/35% of visits. Myocardial infarction was registered in 14 patients, stroke - in 29.

In 2015, there was a conjunction of the main changes in the Kp-index and the number of visits of patients diagnosed with hypertension, which indicates a connection between these two parameters (Fig.1).

Figure 1 shows a graph of the dynamics of changes as well as the conjunctions of two main maximums—the spring and autumn months (March and September-October) of 2015. Thus, for example, the maximum conjunction of AH patients (n=215) and the Kp-index was observed in March 2015, where the Kp-index of geomagnetic disturbance was 612.33. In September, the number of visits increased to 234, compared to the summer months of the year, and the Kp-index was 578.35.

In 2016, there were 2,425 cases of patients with hypertension, of which 1,606 were women and 819 were men. As in 2015, the most cases of AH (62.8%) were observed in the elderly from 60 years and older; middle-aged patients constituted 37.2% - 900 people. Myocardial infarction was registered in 14 patients, stroke - in 30.

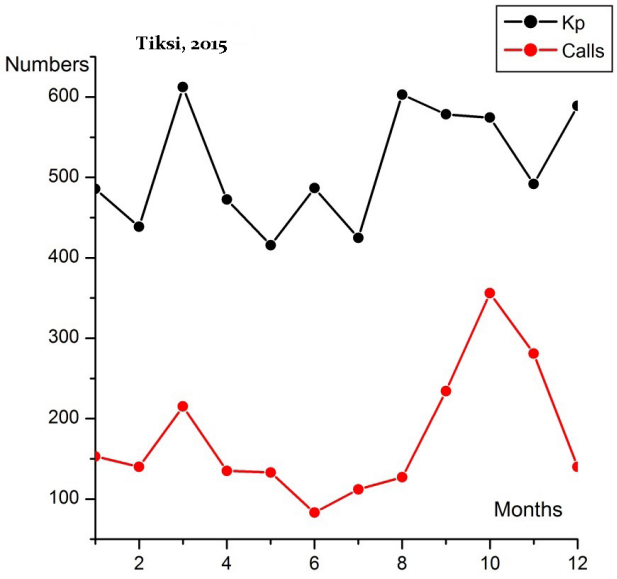

Fig. 1. *Annual dynamics of the Kp-index and the number of visits of patients diagnosed with AH in 2015.*

Figure 2 shows the annual dynamics of the geomagnetic disturbance index and the number of patients with hypertension in 2016: Early in the year, there was a conjunction in March, when the number of visits was 276, and the Kp-index was 502.0. Further, the trends of changes in the components of 2016 began to differ, which may happen because of the imposition of interferences and errors and, accordingly, it requires further detailed consideration and further research.

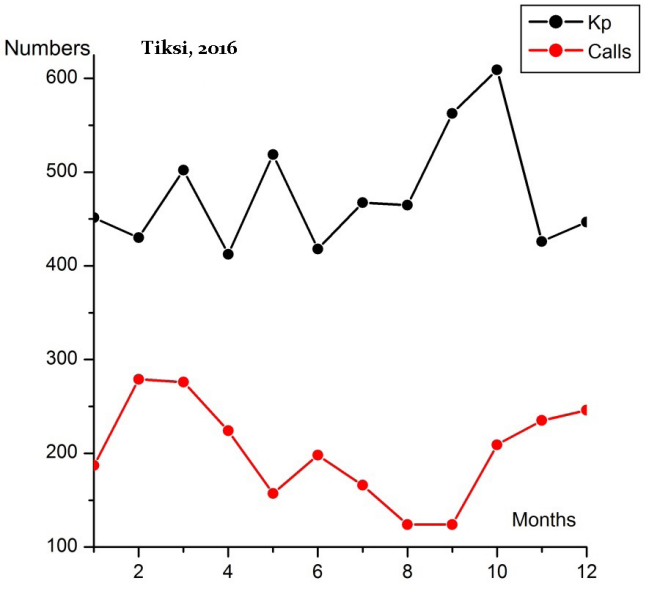

Fig. 2. *Annual dynamics of the Kp-index and the number of visits of patients diagnosed with hypertension in 2016.*

For 6 months (January-June) in 2017, 998 patients were diagnosed with AH, of whom 659 were women and 339 were men. This is slightly less than the number of AH cases during the same period of 6 months in 2015 (n=859), but significantly less than the 6 months period in 2016 (n=1321). Myocardial infarction was detected in 4 patients, stroke - in 15.

As we have the medical data for only 6 months in 2017, the comparison with the Kp-index showed a conjunction of the trend of changes in both parameters. Figure 3 shows that in March 2017, there was a conjunction of the maximum number (n=238) of AH patients with the maximum of the Kp-index of geomagnetic disturbance, which was 567.67. Thereafter, from March to June, there was a conjunction of the dynamics of the Kp-index and the number of complaints about AH: The complaints were gradually reduced to a minimum in June 2017.

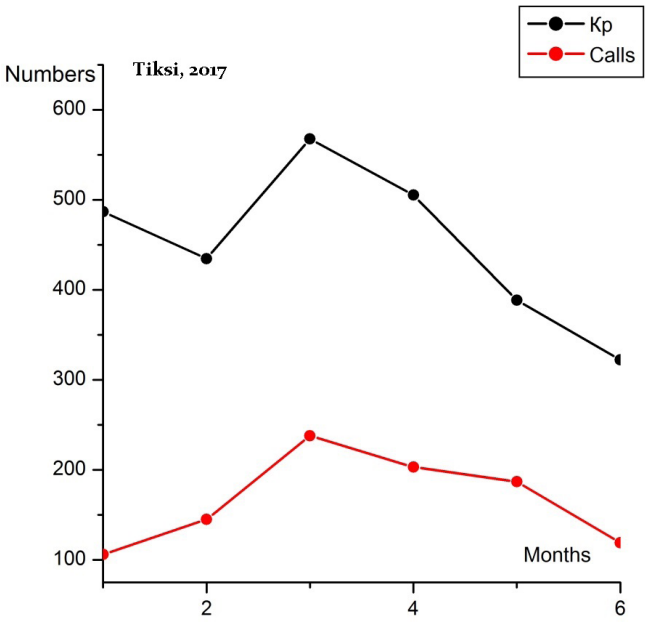

Fig. 3. *Annual dynamics of the Kp-index and the number of visits of patients diagnosed with hypertension for 6 months in 2017.*

Figure 4 shows daily data in 2015, when the changes in the Kp-index and the number of patients' visits for hypertension were taken into account, in comparison with the sex and age of patients. In general, the figure shows an increase and decrease in geomagnetic activity per day during a whole year. The conjunction of the Kp-index correlates with the increase in the number of applications from the older people (65%) and from female patients (n=1416).

Figure 5 represents in more detail the data showing an increase in the Kp-index and the number of medical assistance visits for AH in February 2015. From the graph, we can see a conjunction of the medical data dynamics and geomagnetic disturbance data within one month. At the same time, the sex and age of patients seeking treatment in the Bulunsky Central District Hospital were also taken into account. Figure 5 shows that almost every change in geomagnetic disturbance affects people as an increase in blood pressure. The conjunction of the medical data dynamics and geophysical parameters indicates the effect of geomagnetic disturbance on the human cardiovascular system.

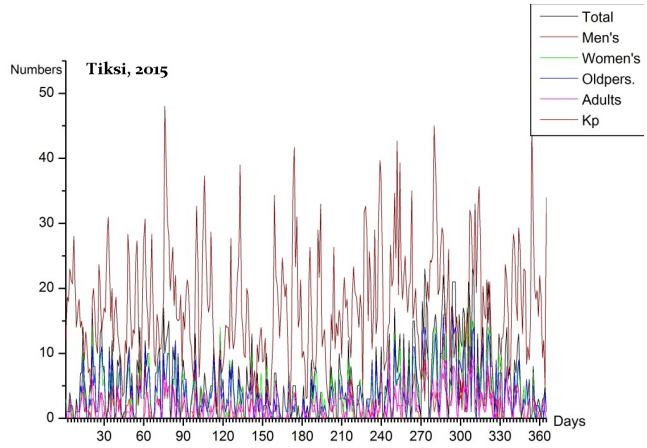

Fig. 4. *Daily data of the Kp-index and the number of visits of patients diagnosed with AH in 2015. Note: the "adults" group corresponds to the average age according to the WHO classification (2017).*

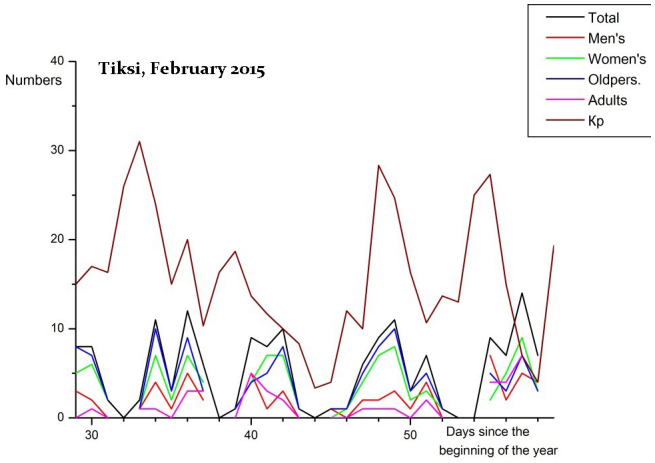

Fig. 5*. Daily data of the Kp-index and the number of visits of patients diagnosed with AH in February 2015. Note: the "adults" group corresponds to the average age according to the WHO classification (2017).*

In conclusion:

1. The conjunction of the maxima of the temporal change in the geomagnetic disturbance (Kp-index) and the number of patients' visits to medical institutions for AH in 2015 and 2017, as well as a partial conjunction in 2016, was determined. On this basis, we conclude that geomagnetic disturbances affect the cardiovascular system of a person living at high latitudes (the Arctic zone of the RS(Y)).

2. The increase in the number of visits for medical care for AH in different periods of a year correlates with the intensity of the Kp-index: The most active periods were the winter-spring in 2015, 2016 and 2017.

The article was prepared based on the results of the project "Assessment of the main trends in change of the natural and socio-economic status, human development of the Arctic

Economic Zone of Yakutia" of the Program of Integrated Research in RS(Y) aimed at developing its productive forces and social sphere in the years 2016-2020.

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 60-64
http://dx.doi.org/10.21103/Article8(1)_OA10

ORIGINAL ARTICLE

Biotechnology

INTERNATIONAL JOURNAL OF BIOMEDICINE

Swelling and Degradation of Calcium-Pectic Gel Particles Made of Pectins of *Silene vulgaris* and *Lemna minor* Callus Cultures at Different Concentrations of Pectinase in an Artificial Colon Environment

Anatoly A. Shubakov, PhD[1]; Elena A. Mikhailova[1,2*];
Valentina I. Prosheva, PhD[1]; Vladimir A. Belyy, PhD[3]

[1]*Institute of Physiology, Komi Science Centre, Ural Branch of the RAS*
[2]*Komi Republic Research Institute of Agriculture, Komi Science Centre, Ural Branch of the RAS*
[3]*Institute of Chemistry, Komi Science Centre, Ural Branch of the RAS*
Syktyvkar, Komi Republic, the Russian Federation

Abstract

Spherical calcium-pectic gel particles (CaPGPs) were obtained from pectins of callus cultures of campion *Silene vulgaris* (silenan) and duckweed *Lemna minor* (lemnan), as well as from commercial apple and citrus pectins by the method of ionotropic gelation. We studied the morphological characteristics of the obtained gel particles and found that the largest gel particles were formed from citrus pectin, and the densest gel particles were formed from lemnan pectin. The swelling and degradation of CaPGPs were comparatively evaluated by incubation in a simulated gastrointestinal environment. The swelling and degradation of CaPGPs formed from pectins of the silenan and lemnan callus cultures were studied at different concentrations of pectinase (0.4, 0.8 and 1.7 mg/ml) in the simulated fluid of the colon. It has been established that the CaPGPs obtained from lemnan are more resistant to degradation in the simulated colon fluid than CaPGP obtained from silenan. It was shown that the concentration of pectinase in the simulated fluid of the colon and the type of pectin affect the degradability of CaPGPs formed on the basis of pectins of callus cultures.**(International Journal of Biomedicine. 2018;8(1):60-64.)**

Key Words: pectins • silenan • lemnan • callus culture • gel particles • gastrointestinal tract • pectinase

Abbreviations

GIT, gastrointestinal tract; **CaPGPs**, calcium-pectic gel particles; **DDS**, drug delivery systems; **CP**, citrus pectin; **LP**, lemnan pectin; **SP**, silenan pectin; **LMEPs**, low methyl-esterified pectins.

Introduction

Enzymes secreted by the body, as well as the enzymes of symbiont microflora, participate in the process of digestion.[1] The major components of dietary fibers are cellulose, non-cellulose polysaccharides, such as hemicelluloses and pectic substances, and lignin, the non-carbohydrate component.[2]

The complex nature of the pectic macromolecule leads to the existence of a variety of structures of pectins with specific properties. Their carbohydrate chains have an irregular block structure and contain various macromolecular fragments that determine the differences in physico-chemical properties and physiological activity of pectin. Pectins are widely used in the pharmaceutical and food industries due to their nontoxicity and biodegradability, and to their high physiological activity (immunomodulating, antiulcer, antitoxic, antitumor) and gel-forming ability.[3,4]

Pectins with the main carbohydrate chain formed by 1,4-linked α-D-galactopyranosyluronic acid residues are

***Corresponding author:** Elena A. Mikhailova. Department of Molecular Immunology and Biotechnology, Institute of Physiology, Komi Science Centre, Ural Branch of the RAS. Syktyvkar, the Russian Federation. E-mail: elena_elkina@mail.ru*

divided into two types: high methyl-esterified (the degree of esterification greater than 50%) and low methyl-esterified (the degree of esterification less than 50%).[5] Low methyl-esterified pectins (LMEPs) form gels in the presence of calcium ions due to the cross-linking of pectin molecules by calcium ions. The study of pectin gels is of particular interest in connection with the problem of creating new functional materials.[6,7]

In human nutrition, pectin is one of the most important sources of dietary fiber. Like other types of dietary fiber, pectin is practically invulnerable to depolymerization by endogenous enzymes of gastrointestinal tract (GIT) when passing through the stomach and small intestine. The physicochemical conditions of the stomach and small intestine can lead just to partial degradation of pectin. In the colon of healthy people and animals, pectin undergoes more or less complete fermentation by pectinolytic enzymes produced by symbiotic microflora.[8] In this case, LMEPs are more preferable substrates for the pectin-depolymerizing enzymes of human and animal microflora.[9]

Currently, the use of pectins in the form of spherical particles for controlled drug delivery systems (DDS) in the body is being actively studied. CaPGPs delay the release of drugs in the upper GIT and release the drugs after the degradation of the particles by pectinolytic enzymes in the colon.[10,11]

However, the scientific literature lacks information on the relationship between the degradability of pectins from different plant sources and the concentration and activity of pectinases in the colon of humans.

The aim of this work was to study the swelling and degradation of CaPGPs in an artificial gastrointestinal medium at different concentrations of pectinase in the artificial medium of the colon.

Materials and Methods

Objects of study and reagents

In this work, we used LMEPs (10-12%) from callus cultures of campion *Silene vulgaris* (M.) G. (SVC) and duckweed *Lemna minor* L. (LMC) with molecular masses of >300kDa,[7] isolated and chemically characterized in the Department of Molecular Immunology and Biotechnology, LMEPs of apple AU-701 (AP, Herbstreith & Fox KG, Germany) and citrus (CP, MP Biomedicals, Inc., Germany), pectinase from *Aspergillus niger* (P, Sigma, 1.18 U/mg of enzyme sample, USA), and calcium chloride ($CaCl_2$, Sigma, USA).

Formation of dry calcium-pectic gel particles and study of morphological properties

CaPGPs were obtained from pectins of callus cultures (SVC, LMC) and from commercial pectins (AP, CP) in the presence of calcium ions by the method of ionotropic gelling, which was described earlier.[10, 11] The pectins (30 or 50 mg) were dissolved in distilled water (1 ml) by slow stirring with a magnetic stirrer MM-5 (Russia) by 2-5 hours at room temperature until complete dissolution.

Gel particles of spherical shape were prepared by drop-by-drop injection of the pectin solution (3% or 5%) from a syringe through a needle with a hole diameter of 0.63 mm on the distance of 4-5 cm in the slowly stirred solution of calcium chloride (0.34 M) and further stirring for 20 minutes at room temperature. The resulting gel particles were then washed three times in distilled water with stirring for 5 minutes and dried for 10-14 h at 37°C.

Further, the diameter, surface area, volume and density of the calcium-pectic gel particles were determined using an optical microscope (Altami, Russia) with a camera and an image analysis program (ImageJ 1.46r program, National Institutes of Health, USA). For calibration, a linear scale was used, one pixel corresponded to 0.024 mm.

Study of swelling and degradation of calcium-pectic gel particles in simulated gastrointestinal media

The swelling and degradation of CaPGPs were studied the under conditions simulating the gastrointestinal environment. (Fig.1) For these purposes, the simulated gastric fluid (SGF solution, pH 1.25), simulated intestinal fluid (SIF solution, pH 7.0) and simulated colonic fluid (SCF solution, pH 7.0). The SGF medium was prepared with NaCl (2.0 g/l), KCl (1.12 g/l), KH_2PO_4 (0.4 g/l) and $CaCl_2$ (0.11 g/l).

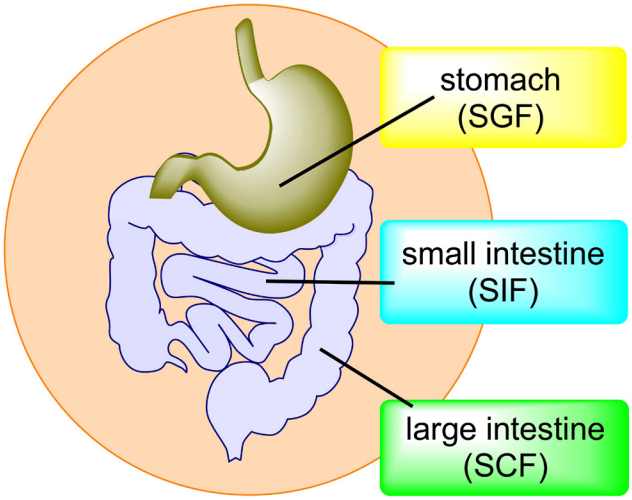

Fig. 1. *Modeling of gastrointestinal tract parts of human by the artificial model environments.*

The pH of the solution was adjusted to 1.25 by addition of 0.1N HCl solution. The SIF medium was prepared by addition of 1N $NaHCO_3$ solution to the SGF solution to the pH value of 7.0. The SCF medium was prepared by addition of pectinase (Sigma, 1.18 U/mg, USA) to the SIF solution (Sigma, USA).[12] Three variants of the SCF medium with different concentrations of pectinase were used: 0.4 mg/ml (0.5 U/ml), 0.8 mg/ml (1.0 U/ml) and 1.7 mg/ml (2.0 U/ml).

10 mg of gel particles of each pectin sample were placed in Petri dishes (diameter 3.5 cm) and subsequently incubated in 3 ml of the SGF (2 h), SIF (4 h) and SCF (18 h) solutions with shaking on a shaker (Titramax 1000, Heidolph, Germany) at 100 rpm and 37°C. The diameter, surface area, volume and density of 100 randomly selected gel particles of each

pectin type were measured as described above after certain time intervals. The experiments were performed in triplicate. The degree of gel swelling (SD, %) was determined by the formula[13]: $SD = (D_1-D_0)/D_0 \times 100\%$, where D_1 – diameter of the particles (mm) after a certain incubation time in the medium, D_0 – initial diameter of the particles (mm).

The statistical analysis was performed using the statistical software BioStat (version 4.03) and Microsoft Office Excel 2007. The mean (M) and standard deviation (SD) were calculated. Multiple comparisons were performed with one-way ANOVA and Tukey's HSD test. A probability value of $P<0.05$ was considered statistically significant.

Results and Discussion

The gel-forming properties of pectins depend on the degree of methyl esterification of carboxyl groups of galacturonic acid residues, on the structure of pectin side chains, pectin concentration, pH, calcium ion concentration, ionic strength of solution, and temperature.[14,15] Spherical CaPGPs are formed as a result of the gelation, in which intermolecular cross-links arise between divalent calcium ions and negatively charged carboxyl groups of pectin macromolecules.[16,17]

The 3% aqueous solutions of silenan, lemnan and apple pectins, and 5% aqueous solutions of CP in the presence of calcium ions yield the spherical CaPGPs, which were subjected to determine their morphological characteristics (diameter, surface area, volume, density). The 3% aqueous solution of CP did not form spherical CaPGPs; it yielded only plate-like particles, which were excluded from further experiments.

The diameters of the dry CaPGPs obtained from the lemnan, silenan, apple and citrus pectins were 0.91±0.03 mm, 1.07±0.07 mm, 1.21±0.05mm and 1.73±0.20 mm, respectively (Table 1).

Thus, the larger particles were formed from CP. However, the densest gel particles were formed from LP (0.93±0.08 mg/mm^3); the density of the gel particles from silenan and apple pectins was lower (0.64±0.08 mg/mm^3 and 0.51±0.05 mg/mm^3, respectively). The gel particles formed by CP were characterized by the lowest density (0.14±0.05 mg/mm^3). Morphological characteristics of CaPGPs obtained from pectins of callus cultures of silenan and lemnan and from commercial apple and citrus pectins have also been studied by other authors.[7,18]

In the study by E.Gűnter,[7] CaPGPs had dimensions and density comparable to our particles. The diameter of the dry gel particles was 1.13±0.08 mm (silenan), 1.14±0.08 mm (lemnan) and 1.35±0.08 mm (apple pectin), with the density of the gel particles of 0.62±0.13 mg/mm^3 (silenan), 0.63±0.14 mg/mm^3 (lemnan), 0.45±0.09 mg/mm^3 (apple pectin).

Thus, stable spherical CaPGPs can be formed from pectin polysaccharides according to the described ionotropic gelling method. Based on the data from various authors, these particles have similar morphological characteristics (sizes and density).[7,16,18]

The influence of pectinase concentration (activity) in the simulated colonic fluid (SCF) on the degradability of CaPGPs was studied during their sequential incubation under the simulated GIT conditions: the simulated gastric fluid (SGF), the simulated intestinal fluid (SIF), and the simulated colonic fluid (SCF) with different concentrations of pectinase.

Three concentrations of pectinase (Sigma, USA) were used in the SCF medium: 0.4mg/ml (0.5U/ml), 0.8mg/ml (1.0U/ml) and 1.7mg/ml (2.0U/ml). The concentration of pectinase of 1.7mg/ml (2.0U/ml), also was used in the SCF medium by other researchers.[7,18] The enzyme activity of 2.0U/ml at the concentration of pectinase of 1.7mg/ml was obviously higher than the activity of pectinase, which we determined in the large intestine of mice (unpublished data), and higher than the activity of pectinase produced by fecal human bacteria.[19,20] At pectinase concentrations of 0.8mg/ml and 0.4mg/ml, the activity of the enzyme was lower than the previous one by 2 and 4 times, respectively. At the concentration of 0.4mg/ml, the activity of the enzyme of 0.5U/ml was comparable to the activity of the enzyme of 0.3U/ml that we revealed in the colon of mice (unpublished data).

Nutrition components undergo complete degradation and are metabolized in different parts of GIT. We have established that CaPGPs formed from commercial pectins of apples and citrus fruits are completely degraded in the artificial medium of the small intestine (Fig.2).

The incubation of the gel particles from 3% apple pectin AU-701 and 5% CP in the simulated fluid of GIT led to their complete degradation in the simulated fluid of the small intestine SIF after 3 hours of incubation in it. Other authors have also showed that CaPGPs formed from apple or citrus pectins are degraded and dissolved in SIF medium.[7,18]

Table 1.

Morphological characteristics of dry calcium-pectic gel particles

Gel particles	Diameter, mm	Area surface, mm^2	Volume, mm^3	Density, mg/mm^3
Ca-CP (1)	1.73±0.20	9.46±1.65	2.82±0.88	0.14±0.05
Ca-AP (2)	1.21±0.05	4.34±0.28	0.96±0.10	0.51±0.05
Ca-SVC (3)	1.07±0.07	3.67±0.47	0.64±0.11	0.64±0.12
Ca-LMC (4)	0.91±0.03	2.67±0.17	0.42±0.04	0.93±0.08
Statistics	F=1044.5825 P=0.0000 P_{1-2}=0.0000 P_{1-3}=0.0000 P_{1-4}=0.0000 P_{2-3}=0.0000 P_{2-4}=0.0000 P_{3-4}=0.0000	F=1202.7884 P=0.0000 P_{1-2}=0.0000 P_{1-3}=0.0000 P_{1-4}=0.0000 P_{2-3}=0.0000 P_{2-4}=0.0000 P_{3-4}=0.0000	F=602.0298 P=-0.0000 P_{1-2}=0.0000 P_{1-3}=0.0000 P_{1-4}=0.0000 P_{2-3}=0.0000 P_{2-4}=0.0000 P_{3-4}=0.0030	F=1664.5995 P=0.0000 P_{1-2}=0.0000 P_{1-3}=0.0000 P_{1-4}=0.0000 P_{2-3}=0.0000 P_{2-4}=0.0000 P_{3-4}=0.0000

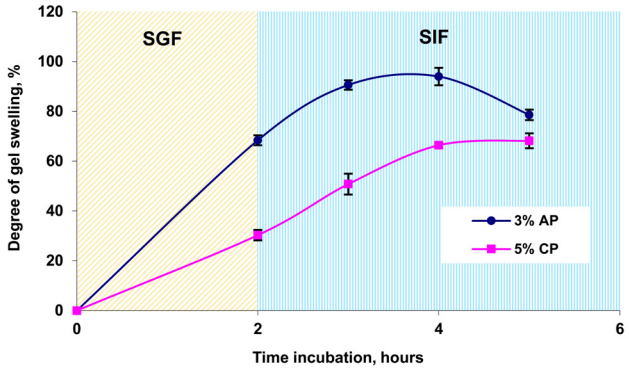

Fig. 2. *Swelling and degradation of CaPGPs of apple and citrus pectins under the conditions of simulated fluid of GIT.*

Morphological changes and swelling and degradation of CaPGPs obtained from pectins of callus cultures of silenan and lemnan, were studied in the SCF medium containing different concentrations of pectinase. The silenan particles were degraded within 2 hours of incubation at the pectinase concentration of 1.7mg/ml, within 2-4 hours of incubation at the concentration of 0.8mg/ml, and within 4-6 hours of incubation in SCF at the concentration of 0.4 mg/ml (Fig.3).

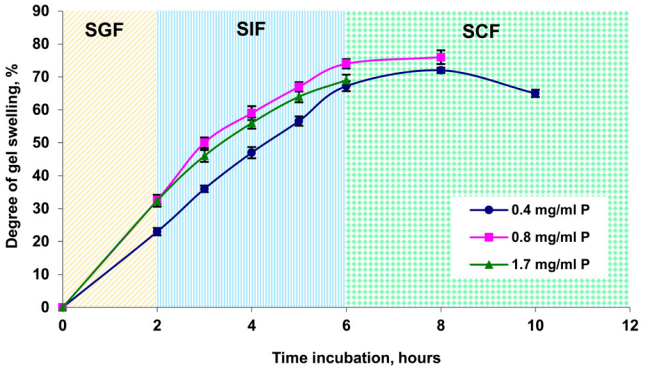

Fig. 3. *Swelling and degradation of CaPGPs of silenan under the conditions of simulated fluid of GIT.*

The lemnan gel particles are more resistant to degradation in the SCF medium than the particles from silenan, which was degraded during a longer incubation period in this medium (Fig.4).

The lemnan gel particles were degraded in the SCF medium at the pectinase concentrations of 1.7mg/ml, 0.8mg/ml or 0.4mg/ml after 16, 18 and 20 hours, respectively. Therefore, reducing pectinase concentration in the SCF medium by 2 or 4 times reduces the degradability of CaPGPs obtained from SP, and the period of complete degradation and dissolution of the gel particles increases by 2 or 4 hours, respectively. It should be noted that in the absence of pectinase in the SCF medium, the gel particles obtained from silenan were resistant to degradation during the 18 hours of the incubation period. It is considered that the lower degradability of CaPGPs based on pectins of callus cultures in conditions of an artificial gastrointestinal environment, compared to gel particles from commercial pectins, can be explained by their higher gel density and lower degree of methoxylation.[14]

Earlier, we and other authors noted that the differences in stability of gel particles formed from different pectins may be due to differences in molecular sizes and in the fine structure of pectic macromolecules.[4,5,16]

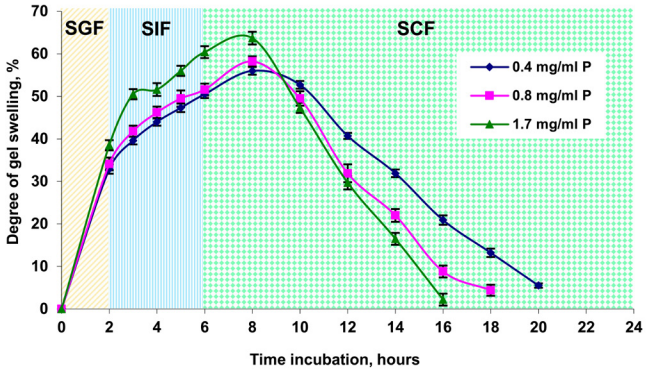

Fig. 4. *Swelling and degradation of CaPGPs of lemnan under the conditions of simulated fluid of GIT.*

Thus, the degradability of CaPGPs in the conditions of a gastrointestinal fluid depends on the plant source of pectin, on the density of gel particles and on the concentration of pectinase in the simulated fluid of the colon.

Competing interests

The authors declare that they have no competing interests.

Sources of Funding

The study was supported by the Federal Agency for Scientific Organisations (Project No. AAAA-A17-117012310147-8).

Acknowledgments

We thank Elena Günter, Oksana Popeyko and Anatoly Melekhin, who kindly provided samples of pectins from callus cultures for the study.

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International Journal of Biomedicine 8(1) (2018) 65-68
http://dx.doi.org/10.21103/Article8(1)_CR1

INTERNATIONAL JOURNAL OF BIOMEDICINE

CASE REPORT

Arrhythmogenic Convulsive Syncope in Neurological Practice: A Case Report

Diana V. Dmitrenko, PhD, ScD[1*]; Irina G. Strotskaya, MD[1]; Ivan P. Artyukhov, PhD, ScD[1]; Olga V. Alekseeva, MD[1]; Natalia A. Shnayder, PhD, ScD[1]; Gennady V. Matyushin, PhD, ScD[1]; Vladimir V. Shprakh, PhD, ScD[2]; Svetlana B. Sayutina, PhD[2]; Oksana A. Gavrilyuk, PhD[1]

[1]Krasnoyarsk State Medical University named after Prof. V.F.Voino-Yasenetsky, Krasnoyarsk
[2]Irkutsk State Medical Academy of Postgraduate Education, Irkutsk
the Russian Federation

Abstract

This case report presents a 24-year-old man with a long history of arrhythmogenic convulsive syncope, which was managed as a cryptogenic generalized pharmacoresistant epilepsy (PRE). During complex examination of the patient within the framework of preoperative screening for the purpose of neurosurgical treatment of PRE, an idiopathic SSS was diagnosed. The clinical diagnosis was changed. The patient underwent emergency surgery at the center for cardiac surgery. ECP was implanted, the seizures stopped, resulting in dramatically improved quality of life.**(International Journal of Biomedicine. 2018;8(1):65-68.)**

Key Words: epilepsy • seizure, syncope • sick sinus syndrome • cardiac rhythm disorders • management

Abbreviations

ECG, electrocardiography; **ECP**, electric cardiac pacemaker; **EEG**, electro-encephalography; **HR**, heart rate; **MRI**, magnetic resonance imaging; **PRE**, pharmacoresistant epilepsy; **SSS**, sick sinus syndrome.

Introduction

Differential diagnosis of arrhythmogenic convulsive syncope and generalized tonic-clonic seizures represents a complicated interdisciplinary problem of clinical medicine,[(1)] since convulsive syncope is often diagnosed as epileptic seizures: In cases of temporary loss of consciousness with convulsions, a major epilepsy diagnosis does not admit of doubt among the majority of primary care physicians.[(2)] All this leads to a long-term and ineffective prescription of anti-epileptic drugs, misdiagnosis of PRE, and patients' referral to neurosurgical treatment.[(3)]

A syncopal condition (syncopal attack, syncope) represents a temporary loss of consciousness due to general cerebral hypoperfusion, characterized by a rapid development, short duration and spontaneous remission.[(4)] The problems of managing major convulsive syncope include refusal of or technical impossibility of conducting long-term video-electroencephalographic monitoring with parallel implementation of ECG monitoring, lack of an interdisciplinary approach to following up and differential diagnostics, and refusal to conduct additional patient-specific, load-adaptive testing that models a trigger situation for the development of an arrhythmogenic convulsive syncope. Our clinical observation of a young male patient who was followed up, together with that of our colleagues is indicative of the abovementioned.

Case Presentation

In June 2016, a 26-year-old male diagnosed with cryptogenic generalized PRE was admitted to the Neurological Center of Epileptology, Neurogenetics and Brain Research of the

***Corresponding author**: Diana V. Dmitrenko, PhD, ScD. Neurological Center for Epileptology, Neurogenetics and Brain Research. Krasnoyarsk State Medical University n.a. Prof. V.F. Voino-Yasenetsky, Krasnoyarsk the Russian Federation. E-mail: mart2802@yandex.ru*

University Clinic of Prof. V.F. Voino-Yasenetsky Krasnoyarsk State Medical University for a pre-surgical examination and adjudication in the matter of further neurosurgical treatment in the Federal Center.

At the initial consultation, the patient presented problems with seizures during the medical manipulations or discussion of medical procedures and events. The seizures started from dizziness, shortness of breath and fluctuations in the level of consciousness for periods up to 2 minutes with subsequent loss of consciousness, skin pallor and single-time clonic convulsions in the upper limbs with periodic urinary incontinence. The convulsion duration did not exceed one minute and no tongue biting was present. There had been no control over either blood pressure level or HR and blood sugar level during seizures. The patient had received long-lasting antiepileptic treatment with lamotrigine (50 mg per day) for one year previously with no clinical effect.

The patient's medical history showed that perinatal anamnesis was not burdened; early psychomotor and speech development conformed to the patient's age. At the age of 3 to 5 months, affective respiratory attacks were registered. At the age of 3, an episode with loss of consciousness and a fall without convulsions was registered for the first time while the patient was viewing an abdominal surgery on television. Syncope was mainly registered during undertaking of medical procedures and/or discussion of the revealed results as well as during discussions about any medical topics and issues, including complaints of the patients' relatives about the state of his health. Seizures with loss of consciousness and convulsions were registered at the ages of 9, 12, 13-14, 17-18, 23, and 25 years. In 2016, 4 events were registered, including seizures with involuntary urination, and generalised myoclonic seizures in the upper and lower limbs, which occurred when the patient saw blood or was present during discussions about results of examinations of his health or health of his relatives. The patient denied having a hereditary history of epilepsy or cardiac rhythm disorders. Electrophysiological examinations of first- and second-degree relatives was not conducted. A cranial-cerebral injury at the age of 6 was present in his medical history.

Objective data: the patient had a satisfactory state, clear consciousness, and preserved intellectual function; he was oriented, critical and emotionally labile. Levels of the state and trait anxiety were elevated. The patient was of normosthenic constitution. Skin cover was smooth, normal color and with no peripheral oedema. No acute distress was observed in his somatic status. In the neurological status, during examination, no symptoms of brain lesions were revealed.

According to data obtained during repeated routine EEG, epileptiform activity was not registered; the ECG channel was turned off during the EEG procedure. The level of lamotrigine in the blood was subtherapeutic (2.44 ug/ml). In order to specify the diagnosis, the patient underwent complete electrophysiological, neurophysiological and neuroradiological examination. An MRI of the brain (1.5 Tesla) according to the epilepsy protocol revealed a single locus of leukoaraiosis (0.2 cm) on the left frontal-subcortical parts of the insula, mild asymmetry of hippocampi D>S (10%) without visible structural alterations.

Considering the provoked character of his seizures, the patient was given long-term video-EEG monitoring with parallel Holter-ECG registration with amplified exercise testing, including partial sleep deprivation with a forced wake-up test. Ictal and interictal epileptiform activity were not registered in passive wakefulness or during sleeping and standard physical exercise tests. In this connection, low-trauma invasive manipulation was performed (the patient lying on his back, venous blood sampling from the cubital vein) for the purpose of modelling the clinical situation that was a trigger to a convulsive seizure. During this time, tachycardia with HR of up to 120 bpm for 10 seconds was registered with subsequent lowering of HR to 45 bpm during 15 seconds and asystole for 15 seconds. During parallel video-EEG monitoring, the patient had a seizure of consciousness impairment with mainly left-sided myoclonic seizures in the lower limbs lasting for up to 2 seconds. During the seizure, verbal contact with the patient was unavailable. Upon analysis of the channel in II standard lead of the ECG before loss of consciousness, an RR pause of 15540 ms was registered with subsequent recovery of the sinoatrial rate (Fig.1).

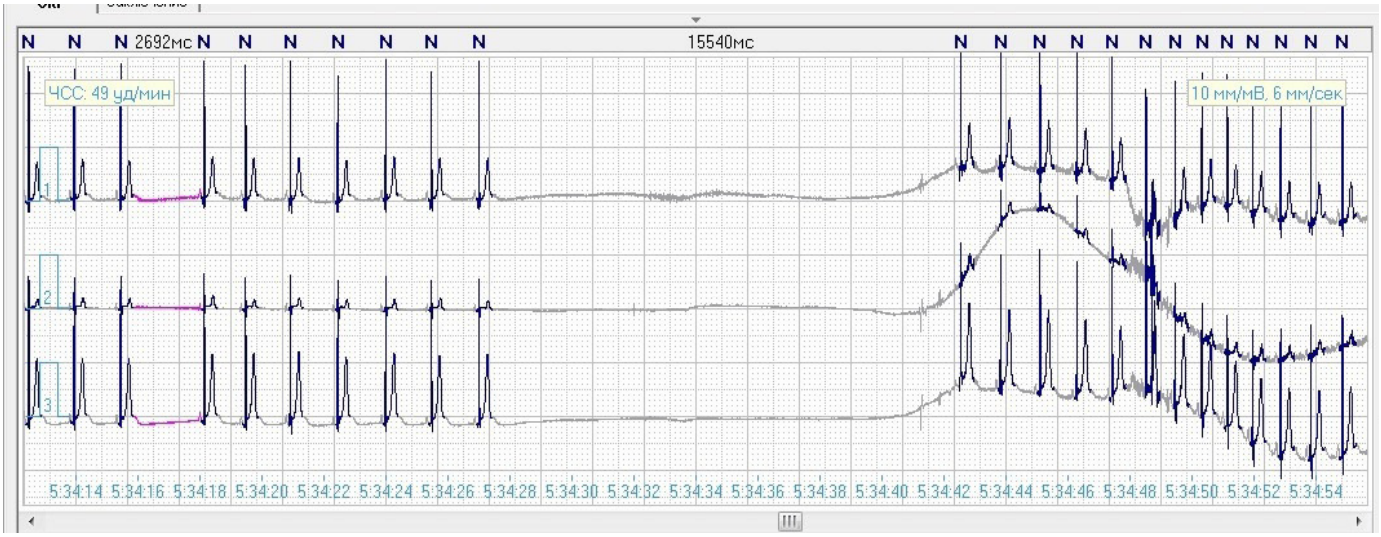

Fig. 1. *Fragment of the ECG with RR pause of 15540 ms.*

According to EEG data, diffuse delta waves (as a marker of a decrease in cortical neuron functional activity) were registered for 4 seconds during consciousness impairment and sinus arrest. Thereafter, defuse polyspike-wave activity with an amplitude of 70 uV was registered for one second (the pattern of a myoclonic seizure), as well as motor artifacts with a duration of 5 seconds with subsequent polymorphous low-amplitude bioelectric activity (Fig.2). Total duration of the paroxysm was 30 seconds. Blood pressure after the paroxysm was 135/70 mmHg.

Taking data of the pre-surgical examination into account, clinical diagnosis was re-established as cardiogenic (arrhythmogenic) generalized myoclonic seizures. SSS? Sinoatrial node arrest.

The patient was consulted by a cardiologist-arrhythmologist during the first 24 hours after development of the above-described seizure. A diagnosis was established for the first time: Idiopathic SSS and transient sinus arrest. The patient underwent emergency hospitalization in the Krasnoyarsk Federal Centre of Cardiovascular Surgery where he was implanted with a bicameral ECP. The patient's state was satisfactory; antiepileptic therapy was cancelled against this background.

According to data obtained from 3 hours of video-EEG-monitoring after sleep deprivation for 24 hours with parallel Holter-ECG monitoring after 3 months following ECP implantation, ictal and interictal epileptiform activity was not registered in the state of passive wakefulness, during exercise tests, and in the slow wave sleep phase. During an exercise test with intravenous injection imitation, HR increased up to 90 bpm (while sitting). Low-amplitude activity in the alpha-two frequency range was registered by means of EEG monitoring; beta-activity showed no alteration in the consciousness level or behaviour of the patient. However, a short episode of consciousness impairment with slackening and without seizures, tongue biting or involuntary urination, and with registration of second-degree AV block with RR pause up to 2004 ms (ECP detection function compromise was not improbable) was registered in the patient during Holter-ECG monitoring (after 24 hours of sleep deprivation).

Catamnesis after 12 months: the patient's state is satisfactory, no repeated bouts, life quality and emotional state of the patient have improved significantly. The patient has received repeated consultations by a cardiologist-arrhythmologist; no seizures were registered within the past 12 months.

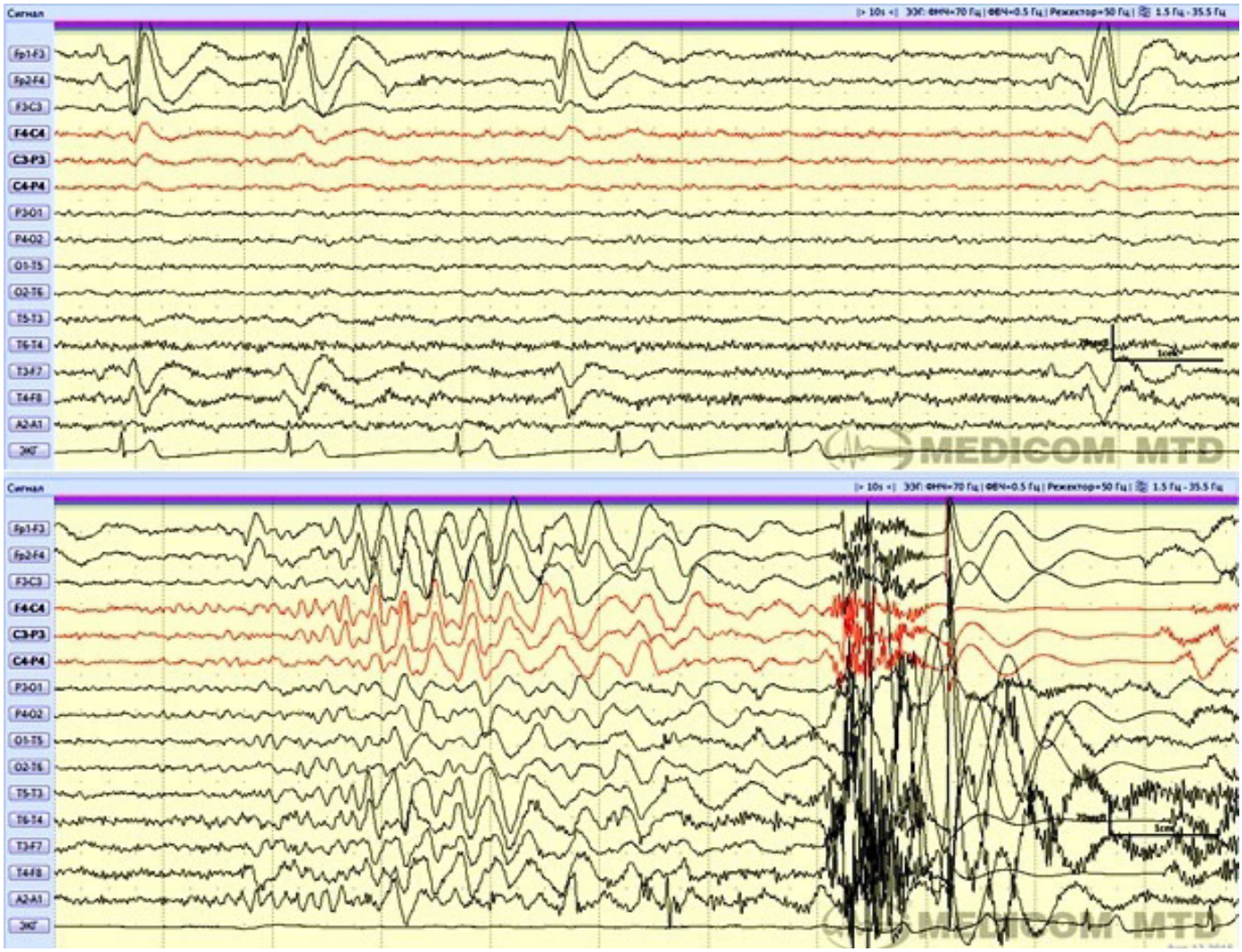

Fig. 2. *Fragment of video EEG monitoring: lowering of HR to 45 bpm during 15 seconds and asystole for 15 seconds. Fragment of EEG: high-amplitude polyspike-wave activity with an amplitude of 70 uV for one second, as well as motor artifacts with a duration of 5 seconds with subsequent polymorphous low-amplitude bioelectric activity.*

Discussion

Differential diagnosis of seizures leading to loss of consciousness is very extensive. In general, an examination is carried out to exclude epilepsy, metabolic disorders, transient ischemic attacks in the posterior circulation system (or drop attacks), narcolepsy and psychogenic pseudosyncopes.[2] Usually, differential diagnosis is not difficult in such cases, although sometimes it can be complicated by insufficient anamnesis, atypical symptoms or actual complexity in syncope identification.[5]

Clinical manifestations of the generalized epileptic seizure can be similar to arrhythmogenic convulsive syncope and include fainting/precollaptoid state, visual and audial disorders, convulsions, tongue biting and involuntary urination. Other possible syncope symptoms comprise epigastric aura with abdominal discomfort and/or unusual foul smells, diffuse hyperhidrosis and skin pallor.[6] Involuntary motions in limbs, urination and tongue biting might be present in both epilepsy and syncope. Modern video-analysis (video-EEG monitoring) makes it possible to state that 90% of patients have myoclonic convulsions, oral automatism symptoms and oculogyric deviation during syncope. Therefore, such patients have clinical manifestations which can be misdiagnosed as epilepsy.[7] Generally, post-seizure disorientation in time and space is absent after a syncope episode. However, retrograde amnesia might occur more often in such cases than had been suggested before, especially in aged patients. Sometimes the postsyncopal period includes manifestations of general weakness and somnolence.[8]

Cardiac rhythm disorders are the most frequent reasons for convulsive syncope as they induce hemodynamic impairments leading to critical lowering of cardiac output and cerebral blood flow. Nevertheless, syncope may be caused by such multiple accessory factors as HR, arrhythmia type (supraventricular or ventricular), myocardial function of the left ventricle, body position and adequacy of vascular compensatory reactions. The latter include baro-receptor nervous reflexes as well as reflex response to eye opening induced by arrhythmia. In cases of SSS, there is damage to the sinoatrial node due to impairment of its automatism or sinoatrial conduction. In this situation, syncope is conditioned by long pauses resulting from sinus node arrest or sinoatrial block and insufficiency of second-order replacing centers. Similar pauses are most often developed after a sudden stop of atrial tachyarrhythmia paroxysm (tachy-brady syndrome).[4]

Arrhythmogenic syncope is an independent factor in risk of sudden death syndrome.[9] The difficulty of diagnosing idiopathic heart rhythm disorders is conditioned by their oligosymptomatic or asymptomatic progression and frequent total absence of representative data from a physical examination. Among high-risk factors, the presence of which requires immediate hospitalisation or intensive treatment, there are clinical and ECG syncope symptoms of arrhythmogenic aetiology, including a syncope seizure during tension or in a supine position, and an increase in HR during syncope.[4]

Most critical elements in the differential diagnosis of epilepsy and convulsive syncope are medical case history, triggers, presence and/or absence of other types of epileptic seizures, and epileptiform activity in EEG scans. At the same time, the absence of epileptiform activity in EEG scans, especially against the background of a single-step examination, still does not exclude the possibility of the seizure having an epileptic nature.[6] Differential diagnostics requires participation of a cardiologist-arrhythmologist in the patient's examination. A tilt-test (or tilt-table test), Holter-ECG monitoring, and event-monitor implantation, as well as distant-recording thermometry, can be used on therapeutic grounds.[3,4,10]

Conclusion

Thus, differential diagnostics of convulsive syncope and epileptic seizures deserves the focused attention of clinicians and requires an interdisciplinary approach to ensure early diagnosis and appropriate treatment, improve the quality of life and reduce the risk of life-threatening conditions, particularly sudden death syndrome.

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 69-74
http://dx.doi.org/10.21103/Article8(1)_CR2

CASE REPORT

INTERNATIONAL JOURNAL OF BIOMEDICINE

Duct Adenoma of the Breast as Background Process for Cholesterol Granuloma: Case Report and Literature Review

Nina A. Oleynikova, PhD[1*]; Olga A. Kharlova[1]; Pavel G. Malkov, PhD, ScD[1]; Natalia V. Danilova, PhD[1]; Nikita S. Kharlov[2]; Sara Y. Davydova[1,3]

[1]Lomonosov Moscow State University, Moscow, Russia
[2]First Moscow State Medical University, Moscow, Russia
[3]Tel-Aviv Souransky Medical Center, Tel-Aviv, Israel

Abstract

Cholesterol granuloma is an extremely rare benign condition that should be considered in the differential diagnosis of breast lesions suspected of being malignant. We present a case of a 55-year-old woman who presented with a nodule mass in the upper external quadrant of the right breast on a routine mammography examination. The case presented here is the first one where the mammary duct adenoma represents a background process for cholesterol granuloma. The described case is noticeable by the combination of multiple cholesterol granulomas in the lungs with breast presentation, which was associated ductal adenoma. **(International Journal of Biomedicine. 2018;8(1):69-74.)**

Key Words: cholesterol granuloma • breast • lung • duct adenoma of the breast

Introduction

Cholesterol granuloma is an extremely rare benign condition that should be considered in the differential diagnosis of breast lesions suspected of being malignant. There is no difficulty in distinguishing cholesterol granuloma from a breast carcinoma on histological examination; however, the clinical findings and radiographic images of cholesterol granuloma are similar to those of cancer, leading to possible confusion between these two diseases.[1] The pathogenesis of breast cholesterol granuloma is still poorly understood and remains controversial. Since 1974 only 20 cases of cholesterol granulomahave been described as a background process for mammary duct ectasia.[2-6] However, in 2004, C. Furuhira et colleagues reported a case of breast cholesterol granuloma accompanied by cancer.[7]

Case Presentation

We present a case of a 55-year-old woman who presented with a nodule mass in the upper external quadrant of the right breast on a routine mammography examination. Physical examination revealed a palpable nodule (d=2 cm) in the upper external quadrant of the right breast that was elastic, firm, and mobile, with ill-defined margins and without associated skin symptoms or regional lymph node involvement. Contralateral breast examination has not revealed any pathological findings.

Blood test: Hb – 143g/L, RBC – 4.87x10^{12}/L, WBC – 6.6×10^{9}/L, lymphocytes – 41.2%, monocytes – 4.8%, eosinophils – 2.0%, basophils – 0.5%, platelets – 179×10^{9}/L, ESR – 5 mm/h.

Blood chemistry: total cholesterol – 6.58 mmol/L; triglycerides – 3.36 mmol/L; urea – 463 µmol/L; total bilirubin – 21.2 µmol/L.

Chest CT: In S3 of the right lung and in the C4 of the left lung, the dense foci with clear contours are visualized.

An ultrasonogram of the right breast disclosed an irregularly shaped mass 35×18mm with multiple, floating, hyperechoic specks within the cyst and ill-defined margins (Fig. 1e-f).

An MSCT of the chest showed a solid nodule with indistinct tuberous margins 23×15×13mm without enlargement of the axillar nodules or bone distraction (Fig. 1a-d).

Cytological examination of a specimen obtained by fine needle aspiration biopsy presented as a small piece of breast tissue with solitary ducts and lobules. More detailed

***Corresponding author:** Nina A. Oleynikova, PhD. Staff Scientist, Department of Pathology, Lomonosov Moscow State University, the Russian Federation. E-mail: ale_x_05@mail.ru*

examination of lobules revealed epithelial proliferation and nuclei enlargement, which was considered as a carcinoma in situ with tentative diagnosis of ductal carcinoma (Fig. 2e-f). Based on that, a lumpectomy of the right breast was performed.

Gross examination of a surgical specimen measuring 10.5×7×2.5cm showed an irregular shaped mass with ill-defined margins 1.8×1×2.3cm at 0.8cm from the fascial margin of resection. An oyster white mass with brown patterns was revealed on section. Microscopic examination showed ducts and lobules with epithelial hyperplasia and apocrine metaplasia, which had formed intraductal papillary structures (Fig. 2a-d). A great mass of fibroadipose tissue was found in breast stroma containing cholesterol granuloma composed predominantly of cholesterol clefts, some of which were surrounded by multinucleated giant cells.

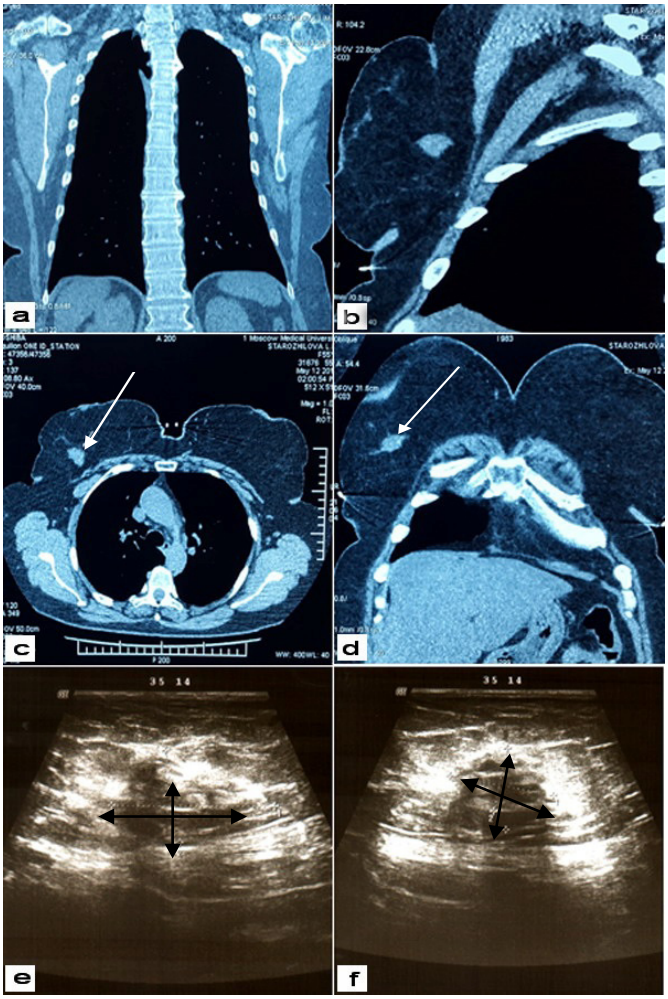

Fig. 1.

The data of CT scan (a-d) and ultrasonography of the right breast (e-f). Note a large, solid mass (black arrow).

The periphery showed haemosiderin-laden macrophages and moderate chronic inflammatory infiltrate (Fig. 3a-e). Malignant tumor growth was absent in resected material. The final histological diagnosis: duct adenoma of right breast with duct ectasia and cholesterol granulomas.

Discussion

Cholesterol granuloma of the breast is composed of fibrous granulation tissue containing a large number of cholesterol crystals surrounded by foreign-body giant cells. This pathological condition is more common in the middle ear and mastoid process, frequent in parotid gland, lymph nodes, liver, and spleen. However, peritoneum and mammary glands are rarely affected.[1]

The pathogenesis of breast cholesterol granuloma is still unclear, but two theories have been reported. According to the first one, mammary duct ectasia is a disease affecting middle-aged women.[8] The primary disorder is dilatation and stasis of the large duct, with accumulation of fatty detritus in the lumen. If the luminal material escapes from the duct, a florid inflammatory reaction rich in macrophages and plasma cells may ensue. Calcification (often as microcalcification) is common, producing tubular, annular, and linear shadows on a mammogram.[1] The second theory is based on periductal inflammation as a primary lesion, with duct dilatation resulting from damage to the elastic lamina of the duct wall.[8]

On one hand, cholesterol granuloma could be caused by periductal inflammation, and cholesterol crystals probably result from lipid-rich material normally found in ectasic ducts escaping from the duct. On the other hand, it can also be the consequence of a rupture of ectasic ducts into the periductal parenchyma due to previous trauma or biopsy (our patient had no history of mammary trauma or biopsy).[5,9,10]

The case presented here is the first one where the mammary duct adenoma represents a background process for cholesterol granuloma. Our finding is in accord with both theories of pathogenesis, due to the presence of ductal ectasia with epithelial hyperplasia and periductal inflammation.

The most important aspect is the process of differentiating between cholesterol granuloma and breast carcinoma, due to the lack of clinical and mammographic features of cholesterol granuloma.[4,5,10-12] For instance, clinical examination of cholesterol granuloma, as well as ductal carcinoma, revealed a firm, mobile mass not associated with skin findings, no abnormal nipple discharge, and an absence of axillary or supraclavicular lymphadenopathy. Mammography revealed a high-density shadow with a partly ill-defined margin and calcifications. Although fine needle aspiration biopsy and smears are less expansive and well tolerated by the patient, these methods do not suffice to render a diagnosis because of the scarcity of the material, or they contain scanty inflammatory cells mixed with very few epithelial cells, as in cholesterol granuloma. Therefore, a definite diagnosis cannot be made. It might be more useful to perform a core needle biopsy that provides a tissue core for histologic diagnosis. Moreover, the cytological examination is not sufficient to exclude a carcinomatous lesion.

Macroscopically, the cut surface of granuloma revealed a nodule that was cystic and firm, with yellow concentric ring-fingers, whereas a ductal carcinoma of the breast shows irregular, stellate or nodular configuration. However, some authors have reported cholesterol granulomas more than 2cm in diameter with papillary structures.[6]

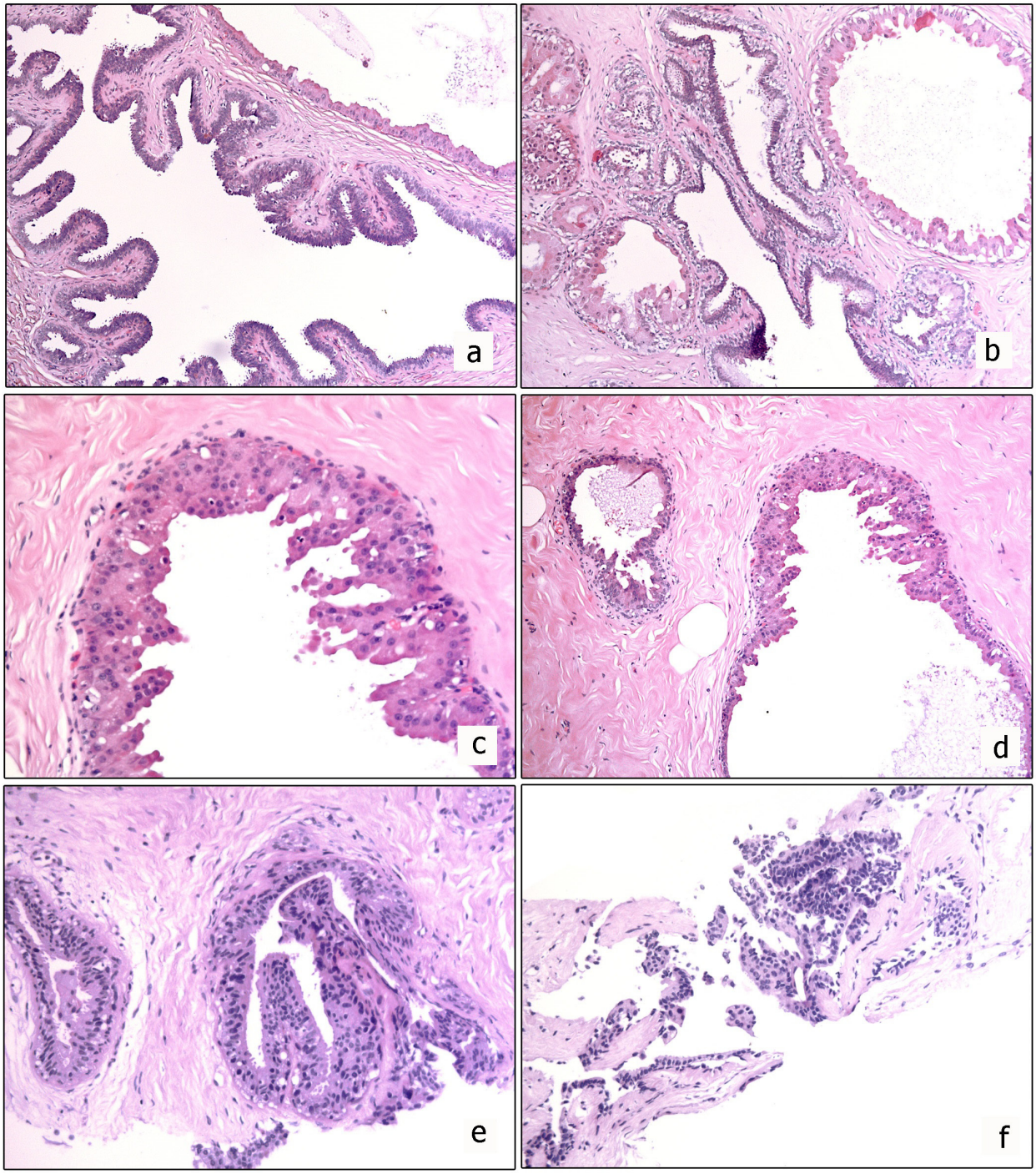

Fig. 2. *Ductal adenoma of the breast.*

a-d – surgical material; e-f – biopsy. a – duct with papillary configuration (H&E; magnification:×10); b – ducts with epithelial proliferation and apocrine change (H&E; magnification:×10); c – apocrine changes of the ductal epithelium (H&E, magnification:×20); d – the elaboration of the apocrine metaplasia (H&E; magnification: ×10); e – gland with epithelial proliferation and cribriform structure; nucleus are in the different levels (H&E, magnification:×20); f – noncohesive cells of similar type, the structure of the gland is not defined, perhaps the focus of infiltrative growth (H&E; magnification:×20).

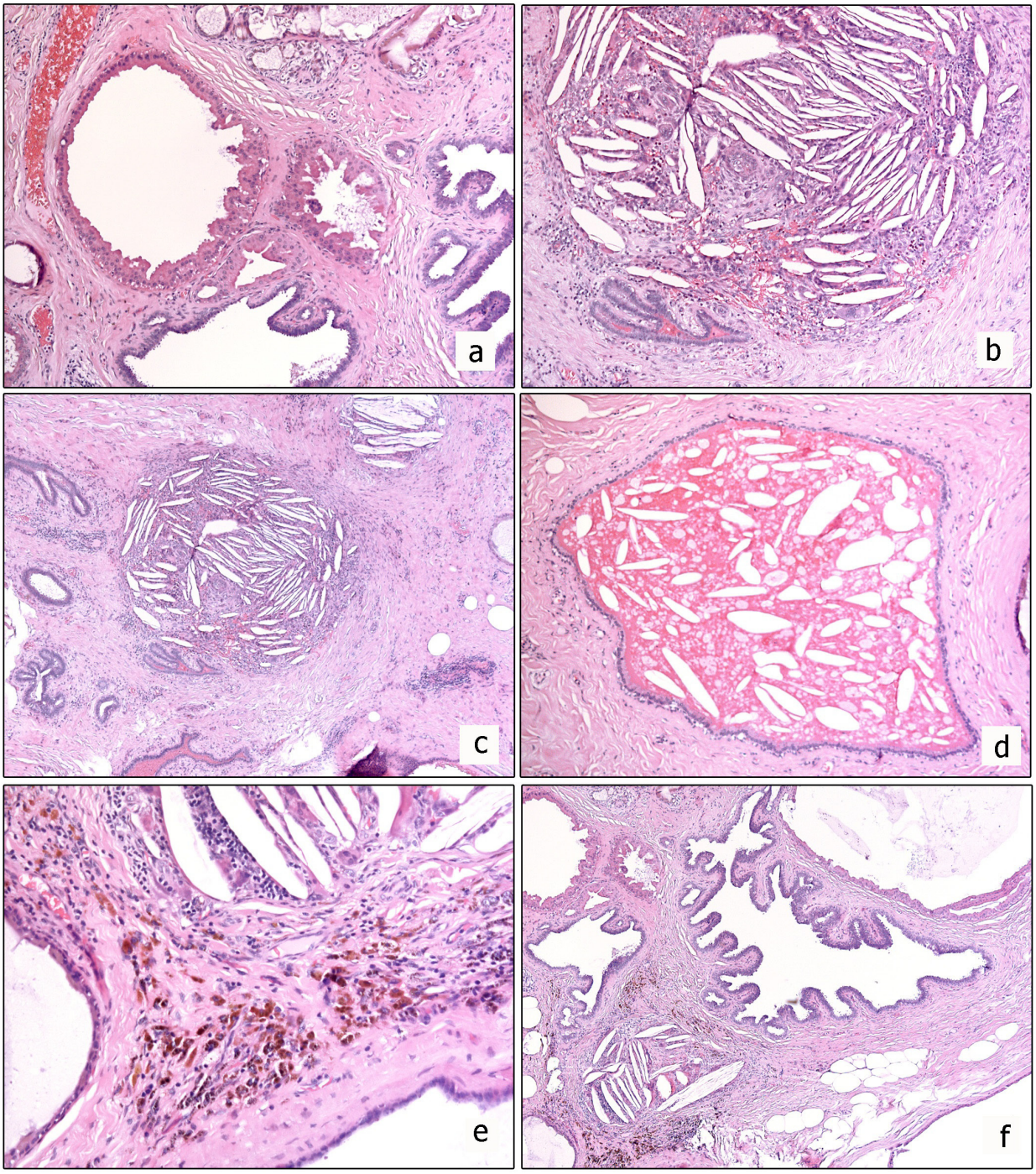

Fig. 3. *The breast cholesterol granuloma.*

a – the cholesterol granuloma between the breast ducts (H&E; magnification:×5); b – cholesterol clefts in fibrotic interstitium with surrounding chronic inflammatory infiltrate and foreign body type multinucleated giant cells (H&E; magnification: ×5); c – cholesterol clefts with surrounding chronic inflammatory infiltrate and foreign body type multinucleated giant cells (H&E; magnification: ×10); d – dilated duct lumen contains many needlelike cholesterol crystals (H&E; magnification: ×10); e – in the periphery of granuloma - deposites of hemosiderine (H&E; magnification: ×20); f – duct ectasia with apocrine metaplasia, papillary structure and periductal inflammation (H&E; magnification: ×10).

Differential diagnosis from papillary neoplasms is based on ultrasonographic features, such as the shape and vascularity of the intraductal lesion, and associated findings such as ductal dilatation. The latter appears as an intracystic solid lesion with peripheral fronting in the dilated duct, with secondary ductal dilatation proximal to the mass. Moreover, the intracystic solid lesion showed increased vascularity.[13] Blood chemistry reveals high cholesterol and triglyceride levels. Although this sign is not specific, especially in patients with hypercholesterolemia, atherosclerosis or other disorders of lipid metabolism.

Histologically, it is not difficult to distinguish between cholesterol granuloma and breast carcinoma. Granuloma of the breast has a typical aspect of needle-like crystals arranged in parallel or radial arrays, sometimes with microcalcifications and metaplastic unusual bone surrounded by histiocytes and giant cells. However, the background process for cholesterol granuloma, such as epithelial hyperplasia and apocrine metaplasia with intraductal papillary structures, could be mistaken for a malignant process.[1,14] Although cholesterol granuloma is not claimed as a risk factor associated with cancer, these conditions progress parallel to each other. Therefore, good histological analysis is required because cancer can be associated with cholesterol granuloma of the breast.

We conclude that breast cholesterol granuloma is a rare lesion that might be accompanied by osseous metaplasia. It must be suspected in woman that carry no risk factors for breast cancer, and an accurate, preoperative study must also be conducted using core needle biopsy, which is more sensitive than fine needle biopsy. Even if surgery is necessary, it must be conservative, and the pathologist must perform a good macroscopic and histological analysis to recognize, eventually, an unknown associated cancer.

There is a tendency to believe that cholesterol granulomas in the lungs have mutual origins with cholesterol granulomas in the breast. These are extremely rare conditions unrelated to endogenous lipoid pneumonia, pulmonary alveolar proteinosis, or cholesterol pneumonia, which could be found during pneumectomy or autopsy. On the one hand, pulmonary parenchymal cholesterol granulomas have been reported in patients with known pulmonary hypertension with the conclusion that a severe increase in pulmonary blood pressure was most important in the pathogenesis.[15,16] On the other hand, cholesterol granulomas could be secondary to or independent of pulmonary hypertension.[17]

Histopathologic examination of 36 patients with plexogenic pulmonary arteriopathy revealed cholesterol granulomas in 9 cases.[18] In another series of radiographic and histopathologic examinations of 20 patients with pulmonary hypertension, cholesterol granulomas were found in 5 cases, but no association with plexiform lesions was suggested.[19]

Another patient who developed cholesterol granulomas after busulfan treatment for chronic myeloid leukemia, was postulated to be secondary to pneumocyte desquamation and independent of pulmonary hypertension.[20] Although microangiopathic hemolytic anemia and thrombocytopenia have been associated with primary pulmonary hypertension, they have not been linked to the development of granulomatous lesions with cleft-like cholesterol deposits.

One of the possible pathogenetic factors is the lysis of red blood cells with release of their membrane lipids.[16] Microangiopathic hemolytic anemia and thrombocytopenia are in association with primary pulmonary hypertension, and the consumption of erythrocytes and platelets was attributed to their destruction in plexiform lesions.[21] In this case, plexiform lesions most likely developed secondary to long-standing pulmonary hypertension, causing hemolysis and thrombocytopenia. Destruction of red blood cells and platelets in plexiform lesions could release membrane lipids and provide them for uptake by multinucleated phagocytes, which could later form granulomas. The presence of hemosiderin granules within those cells supports this hypothesis. Therefore, chronic hemolytic anemia and thrombocytopenia might be contributing factors to the development of cholesterol granuloma. There are reliable data about increased (18)F-fluorodeoxyglucose uptake with cholesterol granuloma during PET-CT examination.[22] Therefore, our patient's reported history of thrombocytopenia, arterial hypertension and severe hypercholesterolemia, could increase the possibility of revealing cholesterol granulomas in the lungs.

Mammary cholesterol granuloma is an extremely rare benign condition affecting middle-aged women with duct ectasia as a background process. There are no data about duct adenoma of the breast as a background process for cholesterol granuloma. This condition is not claimed as a risk factor associated with cancer, although on clinical examination these diseases are difficult to differentiate. There are several cases with cholesterol granuloma and carcinoma that progressed parallel to each other. The described case is noticeable by the combination of multiple cholesterol granulomas in the lungs (with arterial hypertension and thrombocytopenia as a background) with breast presentation, which was associated ductal adenoma.

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 75-78
http://dx.doi.org/10.21103/Article8(1)_CR3

CASE REPORT

INTERNATIONAL JOURNAL OF BIOMEDICINE

Fever as a Presentation of Tumoral Calcinosis: A Case Report

Nga V. Nguyen, MD*; Edgardo J. Guzman MD;
Christopher J. Mesa, MD; Shante A. Hinson, MD

Lincoln Medical and Mental Health Center
Bronx, NY, USA

Abstract

Tumoral calcinosis (TC) is a rare condition in which there is periarticular calcium deposition in the soft tissue forming a mass. The most common locations of TC are the larger joints such as the hip, shoulder, and knee, as well as the hands and wrists. Patients will often present with localized swelling, pain, and reduced joint mobility. We will discuss a 48-year-old male on hemodialysis who presented with a fever of unknown source and diffuse joint pain. He was found to have progressive, multiple tumor-like swellings on his shoulders, hands, and knees. He was diagnosed with TC and managed with a high dose phosphate binder with resolution of his fever and improvement in his pain.**(International Journal of Biomedicine. 2018;8(1):75-78.)**

Key Words: tumoral calcinosis • seronegative spondyloarthritis • hemodialysis • renal insufficiency

Introduction

Tumoral calcinosis (TC) is a rare condition and a complication of hemodialysis therapy.[1] The lesions formed are described as a subcutaneous, periarticular, densely calcified mass confined to the soft tissue, generally at the extensor surface of the joint in the anatomic distribution of a bursa. The formation of the calcifications has been attributed to high calcium and phosphorus levels in the blood. The most common locations of TC are the larger joints such as the hip, shoulder, and knee, however, hands, wrists, and feet have been involved.[2] Plain radiographs reveal periarticular calcifications, sparing the underlying joint.[2] If resected, the lesions have been described as cystic with a white to pale yellow chalky material, identified as calcium hydroxyapatite crystals with amorphous calcium carbonate and calcium phosphate. We report here a case of severe TC in a febrile patient.

Case Presentation

A 48-year-old male with a past medical history of human immunodeficiency virus on treatment, end stage renal disease on hemodialysis via permacath, hypertension, and chronic reactive arthritis/seronegative spondyloarthritis presented to the emergency department (ED) with complaint of fever, chills, malaise, and joint pain. The patient stated he had multiple joint pains, including his hands, shoulders and knees with his right knee being more prominent. In addition, he noticed "lumps" in several parts of his body growing in the past several months, causing discomfort and interfering with his activities of daily living. On review of systems, he denied any rashes, myalgia, sore throat, coughing, nausea, diarrhea, or genital discharge. Medications were consistent with his medical conditions. On examination, he was febrile at 101.5°F, tachycardic with HR of 110bpm, blood pressure was 100/60mmHg, and pain was 10/10 in severity. He was in mild distress with examination notable for tenderness and swelling on multiple joints including the knees and shoulders with the right knee being more prominent. His shoulders and knees had limited range of motion in all directions. In the ED, there was concern for septic arthritis, for which the patient had an arthrocentesis of his right knee. He was started empirically on cefepime and vancomycin. Of note, the patient had been admitted to an outside hospital two weeks prior for similar complaints and was treated with vancomycin at that time. Patient was advised to discontinue his Humira and prednisone. He was admitted to the hospital for further management of sepsis, likely secondary to septic arthritis.

During admission, blood tests: CD4 count of 277cells/mm^3, WBC - 12.4×10^9/L, Hb - 8.7g/dL, ESR - 143mm/hr, C-reactive protein - 28.89mg/dL, blood urea nitrogen - 62mg/dL, creatinine - 10.3mg/dL, calcium - 9.2mg/dL, phosphate

***Corresponding author:** Nga V. Nguyen, MD. Department of Internal Medicine, Lincoln Medical and Mental Health Center, Bronx, NY, USA. E-mail: ngavnguyen@gmail.com*

- 8.4mg/dL, parathyroid hormone - 389.5pg/mL, vitamin D - 31.5. Arthrocentesis of the right knee joint had fluid with a red/bloody appearance; decreased viscosity; WBC-0.9K/uL, 97% segmented neutrophils; RBC/WBC clumps; negative for crystals. A right knee X-ray showed calcification on the medial and lateral collateral ligaments with no bony erosions (Fig.1).

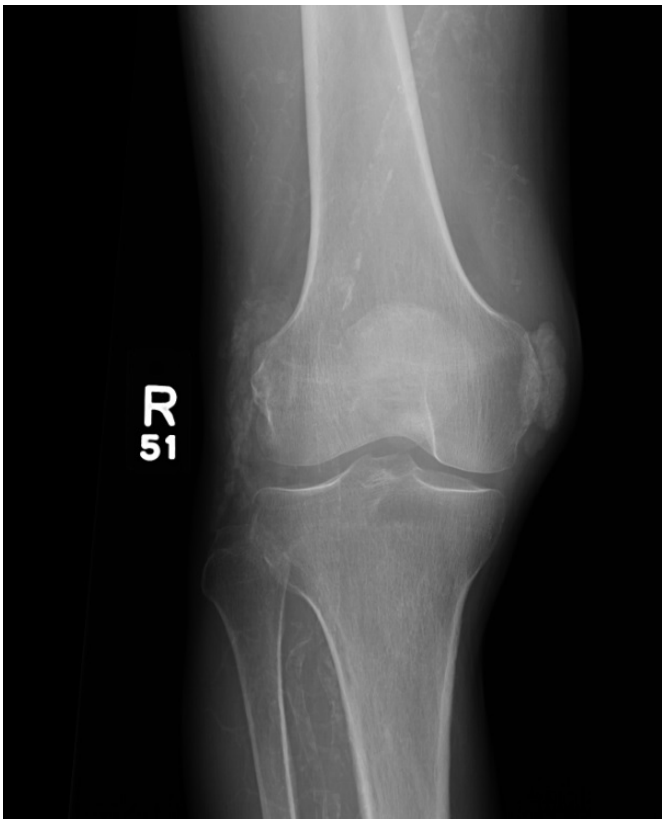

Fig. 1. *Right knee X-ray on admission.*

Given his persistent fever, there was an extensive investigation. Three blood cultures were negative along with a negative blood culture drawn from the permacath; fungal blood culture, urine culture, influenza swab, and chlamydia/gonorrhea urethral culture were also negative. During that time, he complained of a left shoulder mass which was causing discomfort. The impression was that the fever may be related to ongoing inflammation from joint pain. Recommendations were to obtain an echocardiogram to rule out culture-negative endocarditis and imaging of left shoulder, and to continue antibiotics as fever was trending downward, until a complete work-up was obtained. Rheumatology was consulted for a complaint of bilateral knee pain and bilateral hand pain, more on the right than the left. On examination at that time, the right hand had diffuse soft tissue swelling and tenderness over the second and third fingers, nodules on the left forearm, and a palpable mobile soft tissue mass on the left shoulder. There was also tenderness of the medial and lateral aspect of the right knee and tenderness of the lateral aspect of the left knee. The impression was TC, possibly due to secondary hyperparathyroidism in the setting of a hemodialysis patient. The recommendation was to obtain imaging of the left knee, left shoulder, and bilateral hands. It was also recommended to have a discussion with renal services to determine if the patient would benefit from a low calcium dialysate. Echocardiogram did not show vegetations, ruling out endocarditis. The left knee X-ray showed soft tissue calcifications over the lateral femoral condyle and in the anterolateral aspect of the knee joint. Hand X-rays showed multifocal calcifications in the hands and wrists (Fig. 2).

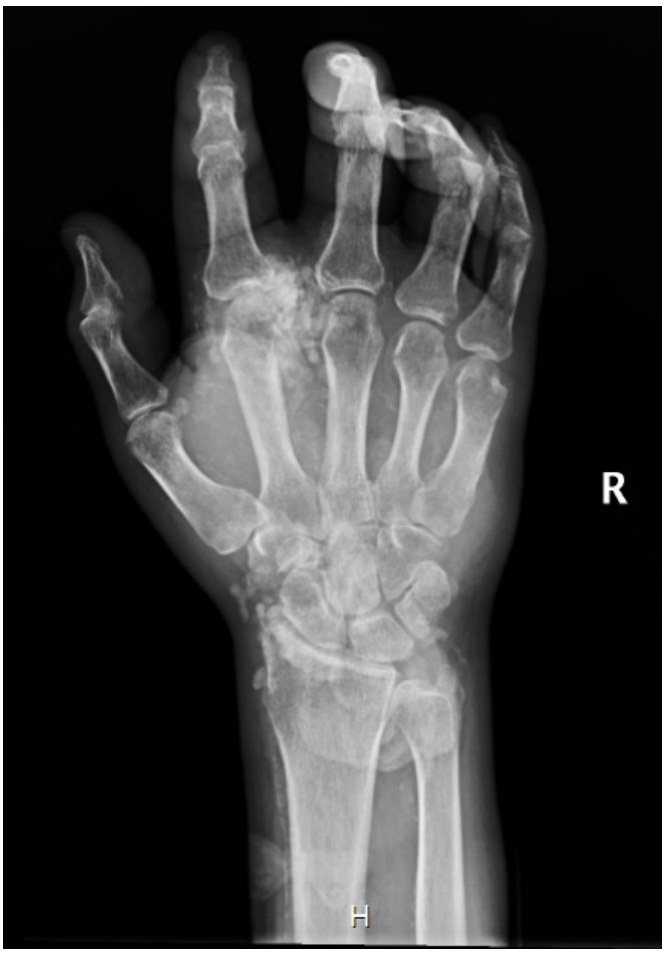

Fig. 2. *Right hand X-ray during admission.*

Shoulder X-rays showed calcification in the left shoulder joint greater than the right with no marginal erosions in the humeral heads (Fig. 3-4). After a multidisciplinary discussion, the patient was started on a phosphate binder, sevelamer 2400mg three times a day, a phosphate restricted diet, and low calcium dialysate during hemodialysis. He completed two weeks of cefepime and one week of vancomycin. As the patient was feeling well and fevers resolved, he was discharged to a short-term skilled nursing facility.

One month after his discharge from the hospital, the patient followed up in the rheumatology clinic. His symptoms improved; however, he was found to have an elevated calcium level of 12.5mg/dL with improving phosphate levels. Given case reports of resolved calcium deposition with parathyroidectomy, he was referred to the endocrinology clinic, where it was discovered the patient was receiving calcitriol from his dialysis center. After calcitriol was discontinued, his calcium levels normalized. Three months after his discharge from the hospital, repeat X-ray imaging of his shoulders showed improving calcifications (Fig. 5-6).

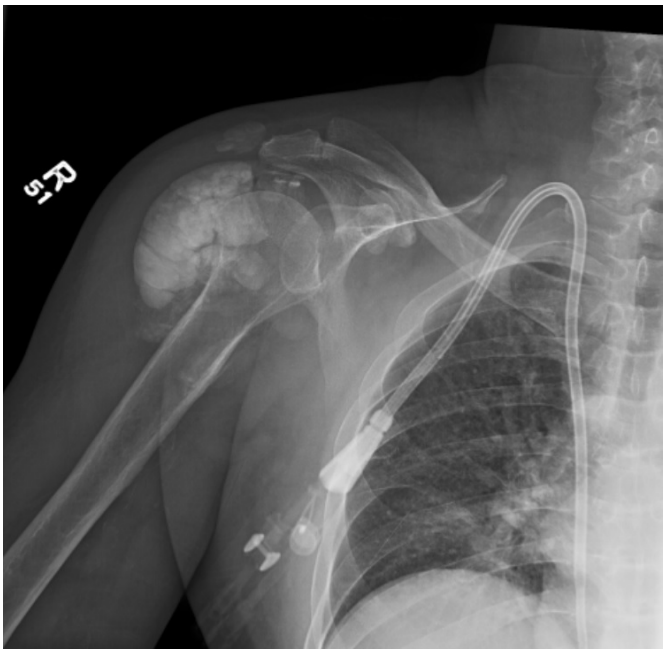

Fig. 3. *Right shoulder X-ray during admission.*

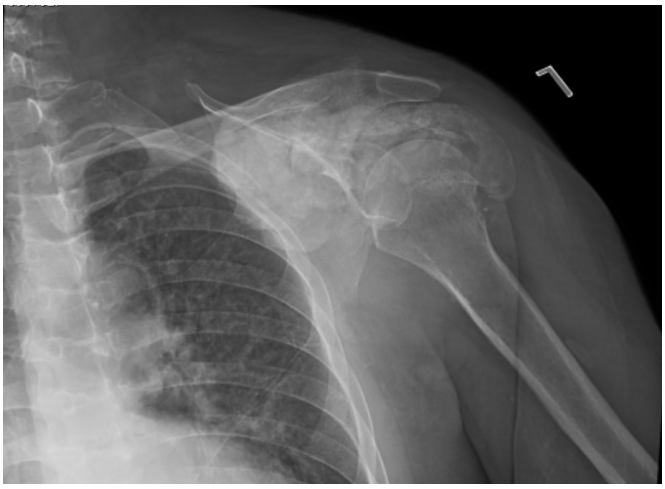

Fig. 4. *Left shoulder X-ray during admission.*

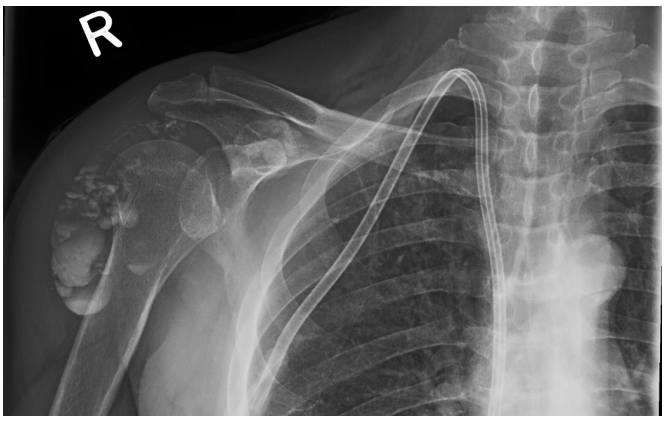

Fig. 5*. Right shoulder X-ray 4 months after admission.*

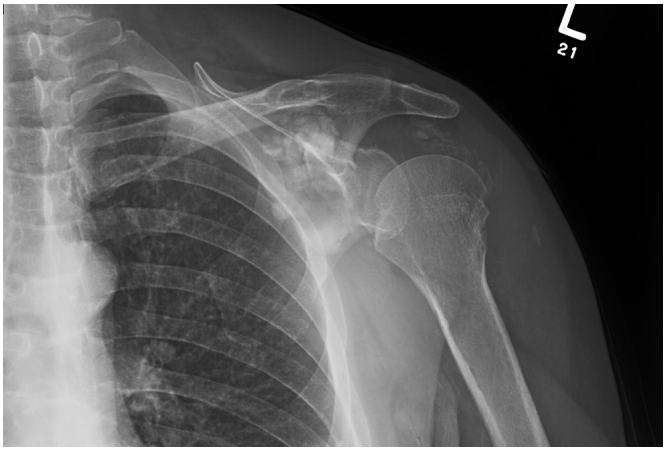

Fig. 6. *Left shoulder X-ray 4 months after admission.*

Discussion

TC is a rare condition in which a calcified lesion is deposited in the periarticular soft tissue, sparing the joint capsule. It can be found as a complication in those with end stage renal disease on hemodialysis due to high serum calcium or phosphate. In 1899, the original condition was first described by Duret in siblings who had multiple calcifications in the hips and elbows.[2] In 1943, the term TC was first described by Inclan, who reported three separate cases with no familial relation.[3] Initially, the condition was thought to be familial/hereditary as it was seen in young, healthy individuals. However, it can also be described as a secondary entity from chronic renal insufficiency, hyperparathyroidism, and hypervitaminosis.[4] The most common sites of tumoral calcinosis are the hip, shoulder, elbow, and knee. The TC deposition leads to reduced mobility, arthralgia, nerve compression, and pain. There are few reports of TC with presenting signs of systemic inflammation, such as fever and constitutional symptoms mimicking infections, as in this case.[5] Such a presentation could lead to a delay in management, inappropriate antibiotic use and extensive investigations. A multidisciplinary approach is often needed for the correct diagnosis and effective management.

Management of TC is often difficult and can be challenging. It involves dietary restriction of phosphate, non-calcemic phosphate binders, dialysis treatment with low calcium dialysate, parathyroidectomy in patients with high parathyroid hormone levels (due to tertiary hyperparathyroidism), surgical resection of the mass, or renal transplantation.[1] In this case, dietary restriction of phosphate, a non-calcemic phosphate binder, and low calcium dialysate was used to manage the extensive TC. A few cases have provided immunohistochemical and microscopic findings from the resected soft tissue calcifications, indicating involvement of histiocytes and osteoclast-like giant cells of histiocyte origin.[6] The fever and systemic inflammatory response from TC could be associated with the release of cytokines and anti-inflammatory markers from these cells. A few reports have described bisphosphate use to prevent a systemic inflammatory response due to cytokine release from osteoclastic activity.[5] Ultimately, the selection

of treatment depends on the severity of the presentation and a change in treatment if there is minimal or no response.

Conclusion

In our case, the patient presented with fever and pain from the diffuse periarticular soft tissue calcifications. Initially, it was presumed he had septic arthritis; however, the joint aspiration was not convincing. He continued to have fever despite being on antibiotics. After a multidisciplinary approach and extensive investigations, he was diagnosed with TC and started on the appropriate treatment with resolution of his fever and improvement in his symptoms. It is important to note that TC could present with pyrexia and systemic inflammatory symptoms/markers, which could delay management or lead to worsening complications if unrecognized.

Competing interests

The authors declare that they have no competing interests.

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International Journal of Biomedicine 8(1) (2018) 79-80
http://dx.doi.org/10.21103/Article8(1)_ShC1

SHORT COMMUNICATION

INTERNATIONAL JOURNAL OF BIOMEDICINE

Effects of Fish Bone Meal Flour and Mineral Water «Abalakhskaya» on Bone Mineral Density

A. M. Palshina, PhD; S. L. Safonova, PhD; E. A. Emel'janova, PhD; V. E. Borisov*; V. V. Epanov, PhD

M.K. Ammosov North-Eastern Federal University
Yakutsk, the Republic of Sakha (Yakutia), the Russian Federation

Abstract

We present the results of the complex application of fish bone meal flour (FBMF) and mineral water «Abalakhskaya» (AMW) for correction of calcium-phosphorus metabolism disorders in patients with abnormal bone mineral density (BMD) and biliary tract pathology. Significant improvement in the quality of life of patients in the postmenopausal period against the background of complex oral administration of FBMF and AMW was accompanied by a positive effect on BMD, and on the functional state of the biliary tract and intestine. **(International Journal of Biomedicine. 2018;8(1):79-80.)**

Key Words: fish bone meal flour • bone mineral density • calcium-phosphorus metabolism • osteopenia

Introduction

The high prevalence of the diseases of the musculoskeletal system and digestive system in the Republic of Sakha (Yakutia) (RS(Y)) requires the development of new technologies for health preservation, including the rational use of ecologically clean, biologically active natural resources available in the territory of RS(Y). The ichthyofauna of Yakutia is represented by 50 species belonging to 18 families.

The rivers and lakes of Yakutia are not only picturesque, but still virgin clean, transparent and full of fish. Every year, fishing companies of Yakutia catch about 5,000 tons of fish. The bone fraction from fish is still regarded as waste. However, fish bones are a high value by-product from the fish farm industry and due to the high calcium content this resource can conveniently be utilised as a high quality food ingredient or supplement.[1-3] In the study by M.K. Malde and colleges, the calcium in enzymatically rinsed bones from Atlantic salmon and Atlantic cod was demonstrated to be a well absorbed source of Ca in young, healthy men.[4]

The purpose of the study was to study the effects of FBMF and AMW on BMD in patients with osteopenia and biliary tract pathology.

***Corresponding author:** *Vasiliy E. Borisov. M.K. Ammosov North-Eastern Federal University. Yakutsk, the Russian Federation. E-mail: vasborisov@yandex.ru*

Material and Methods

The study protocol was reviewed and approved by the Ethics Committee of North-Eastern Federal University named after MK Ammosov (Protocol No. 9 of February 15, 2017). All participants provided the written informed consent.

We used FBMF from northern fish of the whitefish species, which is rich in macro- and microelements, in particular calcium and phosphorus, omega-3, and omega-6, as well as polyunsaturated fatty acids, proteins, fat and water-soluble vitamins to regulate calcium-phosphorus homeostasis in a complex with AMW (low-mineralized hydrogen-carbonate sodium water with a slightly alkaline reaction). The duration of the course of treatment was 6 months (from March to August). The complex of therapeutic and preventive measures also included physical activity and educational programs for correcting overweight.[5]

Statistical analysis was performed using the statistical software «Statistica» (v6.0, StatSoft, USA). A probability value of $P<0.05$ was considered statistically significant.

Results and Discussion

The study included nine women volunteers of Yakut nationality, aged between 53 and 69 years (mean age: 62.9±4.9 years). We determined the levels of ionized calcium in blood and daily urine, as well as 25(OH)D and phosphorus in the

blood. BMD was measured using dual x-ray absorptiometry (DXA) by the GE Lunar iDXA Bone Densitometer in 3 standard projections (lumbar spine, femoral neck, forearm bone). According to the WHO criteria, osteopenia is defined as a BMD T-score between -1 and -2.5 standard deviations (SDs) below the healthy young adult norm, while osteoporosis is defined as a BMD T-score of -2.5SDs or lower.[6]

The assessment of blood pressure by Korotkov's method, ultrasound examination of the abdominal cavity, and esophagogastroduodenoscopy was performed on all patients.

At the beginning of the study, patients complained of pains in the knee and hip joints, in the lumbar spine during the prolonged static load, bitterness in the mouth, bloating, pain in the right hypochondrium, and unstable stools when taking fatty, fried foods. All of them had chronic acalculous cholecystitis and focal atrophic gastritis, and were under medical supervision.

After the course of complex intake of FBMF and AMW, patients reported an improvement in their overall health, normalization of the stool, a reduction of pain in the joints and lumbar spine, an increase in physical activity, an improvement in the condition of the nail plates and hair.

There was a tendency toward in a decrease in the level of ionized blood calcium from 1.23±0.05 mmol/l to 1.16±0.02 mmol/l and daily urinary calcium excretion from 5.65±2.62 mmol/day to 4.76±2.2 mmol/day after the course of complex intervention. We found an increase in blood level of 25(OH)D from 23.7±5.29 g/ml to 28.01±5.29 g/ml ($P<0.05$).

According to DXA, the average increase in BMD was 3.36% (0.03 g/cm^2), and the increase in the T- and Z-scores was 0.3 SD ($P<0.05$).

Postmenopausal osteoporosis is one of the most important problems of modern health care because of the high prevalence and severity of fractures arising from minor injuries. Ca-fortified foods are likely to play an important role in helping consumers to meet the calcium requirements needed to reduce the risk of osteoporosis. Significant improvement in the quality of life of patients in the postmenopausal period against the background of complex oral administration of FBMF and AMW was accompanied by a positive effect on BMD, and on the functional state of the biliary tract and intestine. The results obtained require further study of the effects of the combined use of FBMF and AMW.

Competing interests

The authors declare that they have no competing interests.

Sources of Funding

The study was supported by the Grant of the Head of the Republic of Sakha (Yakutia) for young scientists, specialists and students.

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International Journal of Biomedicine 8(1) (2018) 81-82
http://dx.doi.org/10.21103/Article8(1)_ShC2

SHORT COMMUNICATION

INTERNATIONAL JOURNAL OF BIOMEDICINE

Epidemiologic Aspects of Syphilis among Pregnant Women in the Republic of Sakha (Yakutia)

Snezhana S. Sleptsova, PhD, ScD*; Lyubov I. Petrova; Maksim N. Andreev; Anastasiya N. Petrova

M.K. Ammosov North-Eastern Federal University
Yakutsk, the Republic of Sakha (Yakutia), the Russian Federation

Abstract

The article is devoted to the analysis of the syphilis morbidity rate in pregnant women in Yakutia. Syphilis morbidity in Yakutia in 2014-2016 did not have a sustainable trend toward decreasing in comparison with the Russian Federation mean indicators. **(International Journal of Biomedicine. 2018;8(1):81-82.)**

Key Words: syphilis • pregnant women • newborns • morbidity rate • congenital syphilis

Introduction

According to the Ministry of Health of the Russian Federation, the highest syphilis morbidity in pregnant women was registered in 1997 (277.3 per 100,000). By 2016, the syphilis morbidity rate decreased and fell to 21.3 per 100,000.

Unfortunately, the decreasing syphilis morbidity rate did not solve a problem associated with treatment of pregnant women with syphilis and a high infection risk for newborns. Some authors have noted the involvement of pregnant women and newborns in epidemical processes with direction to high morbidity in these groups in different regions.[1,2]

The aim of our study was to assess the syphilis morbidity rate in pregnant women in Yakutia according to information of the Yakut Republician Dermato-Venerologic Center in 2014-2016.

Results and Discussion

Features of Yakutia that should be taken into account when planning medical care are extremely harsh climate, low and unequal population density (0.1-0.9 per 1 km^2, undeveloped communications, the low capacity of hospitals, and long distances. Syphilis morbidity in Yakutia in 2014-2016 did not have a sustainable trend toward decreasing in comparison with Russian Federation mean indicators (Fig.1). According to data, the active population of reproductive age was involved in the epidemic process. The highest morbidity rate was found in the 21 to 29-year-old age group (55.2-58.2%). In pregnant women, the highest morbidity was also in the same age group. In 2015, 10% of underage women were diagnosed with syphilis. Every year, syphilis was diagnosed in pregnant women of 40 years of age and older, reflecting the tendency towards late pregnancy. The highest syphilis morbidity (41.5%) among pregnant women was registered in 2014. In 2016, we observed a decrease in the morbidity rate of 4% in the past year (26.7% vs. 30.7% in 2015). The majority of these women have been directed to the Center by ob/gyn doctors (85% in 2015, 88.8% in 2016, and 81.4% in 2014). In 2016, 11.1% of pregnant women came directly, and 14.8% in 2014. During medical dyspanseration, syphilis was diagnosted in 5% of cases in 2015 and in 3% of cases in 2014.

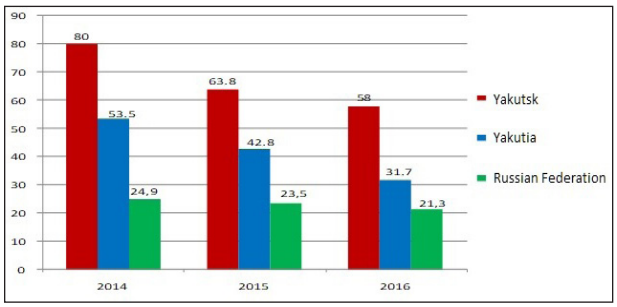

Fig. 1. *Syphilis morbidity in Yakutsk city, the Republic of Sakha (Yakutia), and the Russian Federation (per 100 000).*

***Corresponding author:** Prof. Snezhana S. Sleptsova, PhD, ScD. M.K. Ammosov North-Eastern Federal University. Yakutsk, the Sakha Republic, Russia. E-mail: sssleptsova@yandex.ru*

During the 3-year period, about of 41,5% of cases were diagnosed in the second trimester (51.8% in 2014), 33.8% of cases in the first trimester (25.5% in 2014, 30% in 2015, and 50% in 2016) and 24.6% in the third trimester.

The social status of pregnant women with syphilis was as follows: 55%-61% of women were married; 77.7% of women had a middle educational level; 37.8% were unemployed; 65% of women were childless at the time of the current pregnancy; and 26.4% of women had children. The latent (hidden) stage of syphilis was identified in 62% of cases. We observed a tendency towards an increase in the hidden stage of syphilis: 54% in 2014, 60% in 2015, and 72% in 2016. Simultaneously, secondary syphilis decreased (40% in 2014, 22.5% in 2015, and 18.5% in 2016).

One of main prevention measures in the fight against syphilis is the detection of the source and treatment of all cases and contacts. We analyzed about 45% of detected sources in 2014, 61.1% in 2015, and 62.9% in 2016. Among pregnant women with untreated early syphilis who did not receive *adequate* therapy, we observed high fetal and neonatal mortality rates. According to our study, all of the observed women gave birth. Between 2014 and 2016, we had no medical abortions or miscarriages, but the premature birth rate was 7.6%. Every year, about 83.3%-96.2% of pregnant women recieved specific and preventive treatment. Untreated cases were related to premature births.

Congenital syphilis is an indicator of inadequate antenatal care services and poor quality of programmes to control sexually trasmitted infections.[3-5] We identified one case of congenital syphilis in 2016 vs. 17 cases in 2014. Thus, the data obtained require further studies on epidemiologic features of syphilis prevalence in Yakutia.

Competing interests

The authors declare that they have no competing interests.

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