PEDIATRICS

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SIGNS OF INFLAMATION IN THE AIRWAYS IN CHILDREN WITH EXERCISE-INDUCED BRONCHIAL ASTHMA

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Abstract: The study basing on examination of 102 children with bronchial asthma defines indices of inflammatory activity in the airways and sets the diagnostic value of these indices as tests for verification exercise-induced bronchial asthma. The children with no signs of exercise-induced asthma compared to patients having them, were shown to experience some changes in the exhaled breath condensate, indicating a higher activity of inflammation in the airways. The content of nitrogen monoxide metabolites less than 50 mcmol/L or markers of proteolytic activity by azocol lysis less than 0.2 ml/hour in pulmonary expiration products increase the chances of having exercise-induced bronchial asthma phenotype with these tests sensitivity within 66.7-75.7%.

KeyWords: exercise-induced bronchial asthma, children, diagnostics, inflammation of airways, pulmonary expiration.

INTRODUCTION

According to the definition of PRACTALL [1], exercise-induced bronchial asthma (EIBA) is regarded as a type of asthma associated with exercise-induced transient bronchial obstruction [2, 3]. At the same time exercise-induced bronchospasm (EIBS) is objectively defined as reduction of FEV1 by 10% and more compared to the initial value after a bronchoprovocation test [4, 5]. The detection rate of EIBA and EIBS in pediatric populations varies in quite a wide range depending on the terminology, which the researchers prefer, choosing populations, using different objective criteria of the disease with an arbitrary choice of the distribution point of the results, taking into account factors that affect the severity of EIBA. The causal role of inflammation in exercise-induced bronchial obstruction in patients with bronchial asthma (BA) and people without the disease has been confirmed by many studies [6].

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For instance, based on the examination of patients with asthma and otherwise healthy children, we noted that in EIBA the rate of leukotrienes in exhaled breath condensate increased by half and reliably correlated with decreased FEV1 after exercise. It enabled to assume that leukotrienes, along with nitrogen monoxide, are actively involved in the development of exercise-induced bronchoconstriction [7]. Howerer, it should be noted that the role of nitrogen monoxide in the formation of EIBA is ambiguous. For instance, it was noted that its content in the exhaled breath condensate does not correlate with spirographic changes [8], but, in the event of EIBA, its content in exhaled air may increase, decrease or not change significantly [9, 10]. Still, the low level of NO in exhaled air can probably be seen as a marker of the absence of EIBA in screening studies in cohorts of young children suffering from asthma [11]. Thus, the available information concerning airway inflammation markers found in exhaled breath condensate, can now be considered contradictory and unclear.

2.1 Purpose

The aim of the study is to identify the signs of inflammatory activity in the airways of children with exercise-induced asthma.

2.2 Subjects & Methods

For this purpose we examined 102 schoolchildren with asthma at Pulmonary and Allergy department of Chernivtsi Regional Pediatric Hospital. In the article we used the classification and management of asthma according to "Unified clinical protocols of primary, secondary (specialized) medical care" Asthma in Children "(MOH of Ukraine number 868 of October 8, 2013) with the guidelines of The International global initiative for the diagnosis and treatment of asthma (GINA 2006-2010) [12-14], NAEPP-2008 [15], ICON-2012 [16], approvals PRACTALL-2008 [17] and national protocols of Australia (2008) [18], UK (2011) [19], Japan (2010) [20]. The diagnosis of exercise-induced asthma (EIBA) was formulated according to the recommendations of PRACTALL-2008 [17], the European Respiratory Society (ERS) and the European Academy of Allergology and Clinical Immunology (EAACI) in cooperation with GALEN (2008) [21].

The survey was conducted in parallel groups formed on the basis of a simple random selection using the "experiment-control" method. Based on the examination of the children, we formed two clinical groups. The first (Group I, basic) group included 50 schoolchildren diagnosed with EIBA, and the comparison group (Group II) consisted of 52 patients suffering from BA without signs of exercise-induced bronchospasm (EIBS). The average age of children in the first clinical group was 11.2±0.4 (95% CI 10.3-12.1) year. The first group involved 22 girls (44.0%) and 28 (56.0%) boys. Furthermore, 27 children (54.0%) were rural inhabitants and 23 patients (46.0%) were city dwellers. The average disease duration was 6.18±0.45 (95% CI 5.3-7.1) years. The second clinical group comprised 16 girls (30.8%) and 36 (69.3%) boys. The average age of patients in the second clinical group was 12.02±0.46 (95% CI 11.1-12.9) years and 25 children (48.1%) lived in the rural area while 27 patients (51.9%) were city dwellers. The average duration of disease reached 6.77±0.55 (95% CI 5.7-7.9) years. These data give reason to believe that comparison groups did not differ significantly in the main clinical characteristics, and therefore were comparable.

We identified inflammation signs in the exhaled breath condensate (EBC) of all the children: total protein by the method of Lowry O.H.[22]; concentration of nitrogen monoxide metabolites by N.L. Yemchenko [23]; markers of proteolytic activity by azoalbumin, azocasein and azocollagen lysis by K.N. Veremeyenko et al. [24]; the content of aldehid- and keto- secondary products of 2,4-dinitrophenylhydrazines (AKDNFH) of basic and neutral nature by O.E. Dubinina et al. [25]; catalase activity by M.A. Koroliuk et al. [26].

To determine the diagnostic value of the findings obtained in the comprehensive survey of children as the verification tests of EIBA, we determined their sensitivity as a test, specificity, predictive value of positive and negative results by defining confidence intervals (95% CI). Based on these characteristics of the test, we determined the ratio of reliability of positive and negative results, as well as post-test probability of the event with positive and negative test results. Risk assessment of the implementation of events was carried out on the basis of probability of relative, attributive risks and odds ratios events while defining their confidence intervals (95% CI).

Conflict of interests

There is no conflict of interests.

3 RESULTS AND DISCUSSION

It should be noted that the content of nitrogen monoxide metabolites (NMM) in exhaled breath condensate, which can be seen as an integral indicator of inflammatory activity, was reduced in children of the first clinical group compared to the comparison group of patients. For instance, the average content of NMM metabolites in patients with EIBA was 47.27±3.1 (95% CI 49.96-37.54) mmol/L. The representatives of the second clinical group

had the content of NMM metabolites in EBC as high as 48.78±3.67 (95% CI 56.27-41.29) mmol/L (P>0.05). Despite the absence of reliable inter-group differences, the frequency of found NMM metabolites in EBC content more than 50,0 mmol/L in the first group was 24.3% of cases and 39.4% of cases in the comparison group (P>0.05). Perhaps the lack of probable differences on this inflammatory marker is due to the fact that it reflects both the activity of the inflammatory process and its protective role in relation to the inflammation process.

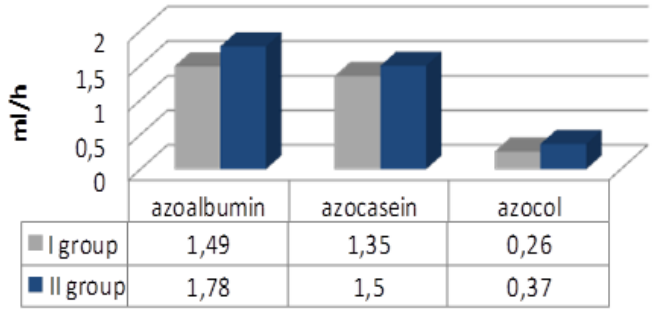
According to the literature, inflammation markers in the bronchi, which are found in EBC were supposed to be more pronounced in children of the second clinical group. Indeed, the content of products of protein peroxidation in EBC, which can be considered as an indicator of activity of oxidative stress in the bronchi, in these children was slightly higher than in patients with EIBA (Table. 1). However, a slight increase of total protein in EBC is likely to be regarded as an indicator of more pronounced vascular permeability that accompanies inflammation and can be seen as a manifestation of "leakage" syndrome.

Table 1.

Indices of lipid peroxidation in EBC in children of the comparison clinical groups (M±m)

Clinical groups	Number of children	Total protein, g/l	AKDNFH of basic nature, o.u.g of protein	AKDNFH of neutral nature, o.u.g of protein
Group I	30	3,76±0,23	48,51±6,69	9,4±2,49
Group II	41	4,22±0,27	50,8±5,62	5,89±0,63
P		>0,05	>0,05	>0,05
Note: P – Student's test; AKDNFH – aldehid- and keto- secondary products of 2,4-dinitrophenylhydrazines				

Proteolytic activity of EBC, expressed in ml/h, which can be regarded as an additional criterion for assessing the activity of inflammation in the bronchi, did not differ significantly in the comparison groups and tended to prevail in the second clinical group (Fig. 1).



Note: in all cases P>0.05

Fig. 1. Results of expired breath condensate proteolytic activity (ml/h on azoalbumin, azocasein and azocol lysis in the children under study

Lower activity of catalase in EBC that serves as a key enzyme of antioxidant bronchial defense must have contributed to the development of more active inflammation in the airways of children from the second clinical group. For instance, in children with no signs of EIBA the catalase activity in EBC amounted to 46.93±5.6 (95% CI 34.0-79.8) mmol/min x mg of protein, and 58.19±3.1 (95% CI 33.1-83.3) mmol/min x mg of protein in children of the first clinical group. The activity of catalase less than 40.0 mmol/min x mg of protein in patients with EIBA was found in 61.9% of cases and in those from the comparison group in 73.7% of cases.

According to current ideas, chronic inflammation of the airways, which in patients with clinically significant EIBS has more pronounced transitory nature is a basis of asthma pathogenesis and its particular phenotype EIBA [27]. Therefore, it was appropriate to study the diagnostic value of some signs of bronchial inflammation in EBC, in particular, the level of nitrogen monoxide metabolites (NMM), total protein and products of oxidative modification (basic and neutral AKDNFH). The results of this analysis are presented in Table 2. It should be noted that more sensitive tests (the content of nitrogen monoxide metabolites in EBC <50.0 mmol/L and total protein <4.0 g / L) increased the likelihood of post-test EIBA with positive result not more than by 5.0%.

Table 2.

Diagnostic value of some indices of the expired breath condensate for EIBA verification

Indices of EBC	Diagnostic value, % (95% CI)				Likelihood ratio	
	Sensitivity	Singularity	Prognostic value		Positive result	Negative result
			positive result	negative result		
Total protein < 4.0 g/l	63.3 (53-72)	48.8 (39-59)	55.3 (46-65)	57.1 (46-68)	1.24	0.75
Basic AKDNFH >55.0 o.u.g of protein	40.0 (30-50)	74.3 (65-83)	60.9 (48-73)	55.3 (46-64)	1.56	0.81
Neutral AKDNFH >10.0 o.u.g of protein	20.0 (13-29)	86.0 (78-92)	58.8 (41-75)	51.8 (44-60)	1.43	0.93
Nitrogen monoxide metabolites <50.0 mcmol/L	75.7 (66-84)	39.3 (30-50)	55.4 (47-64)	61.9 (49-74)	1.25	0.62
Azoalbumine lysis <1.2 ml/h	16.7 (10-25)	87.5 (79-93)	57.2 (38-75)	51.2 (43-59)	1.34	0.95
Azocasein lysis >1.5 ml/h	50.0 (40-60)	56.3 (44-66)	53.4 (43-64)	53.0 (43-63)	1.14	0.89
Azocol lysis <0.2 ml/h	66.7 (56-76)	50.0 (40-60)	57.2 (48-66)	60.0 (49-70)	1.33	0.67

At the same time, the content of basic and neutral AKDNFH in EBC increased the post-test likelihood of this phenotype in case of a positive result by 11.0% and 8.0%, respectively.

However, all of the suggested indices, meant, to some extent, a probable risk of realization of this phenotype of asthma in the examined patients (Table 3).

Table 3.

Clinical and epidemiological risk of EIBA depending on some indices of exhaled breath condensate

Indices of EBC	Odds ratio (95% CI)	Relative risk (95% CI)	Attributive risk
Total protein < 4.0 g/L	1.64 (0.9-2.9)	1.29 (1.0-1.6)	0.12
Basic AKDNFH >55.0 o.u.g of protein	1.93 (1.1-3.5)	1.36 (0.9-2.1)	0.16
Neutral AKDNFH >10.0 o.u.g of protein	1.54 (0.7-3.2)	1.22 (0.6-2.3)	0.11
Nitrogen monoxide metabolites <50.0 mcmol/L	2.02 (1.1-3.7)	1.45 (1.2-1.8)	0.17
Azoalbumin lysis <1.2 ml/h	1.4 (0.6-3.1)	1.17 (0.6-2.3)	0.08
Azocasein lysis >1.5 ml/h	1.29 (0.7-2.2)	1.13 (0.8-1.5)	0.06
Azocol lysis <0.2 ml/h	2.0 (1.13-3.6)	1.43 (1.1-1.8)	0.17

These findings give reason to believe that the probable risk of EIBA is indicated by higher sensitivity tests. However, it should be noted that, from a clinical point of view, the above mentioned risks of EIBA should be seen as rather modest arguments in favor of this phenotype of the disease.

4 CONCLUSIONS

Thus, the children with no signs of exercise-induced asthma compared to patients having them, experience some changes in the exhaled breath condensate, indicating a higher activity of inflammation in the airways. The content of nitrogen monoxide metabolites <50 mcmol/L or markers of proteolytic activity by azocol lysis <0.2 ml/h in pulmonary expiration products increase the chances of having EIBA phenotype with these tests sensitivity within 66.7-75.7%.

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