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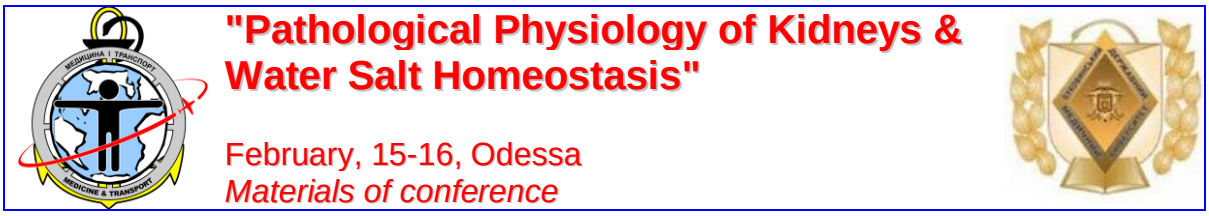

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PECULIARITIES OF BRONCHIAL ASTHMA MANAGEMENT IN CHILDREN WITH ALLELIC *GSTT1*, *GSTM1* GENE POLYMORPHISM

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Abstract

Content of peculiarities of basic anti-inflammatory therapy and its efficacy in children with available or absent deletion polymorphism of *GSTM1* and *GSTT1* genes coding II phase enzymes of *GSTM1* and *GSTT1* xenobiotic detoxication considering their acetylation status. It is established that in patients of a school age suffering from bronchial asthma with available deletion polymorphism of *GSTT1* and *GSTM1* genes associated with slow acetylation phenotype anti-inflammatory therapy should be intensified emphasizing on much higher "step" or by means of addition of other anti-inflammatory drugs. In patients without deletion polymorphism of the examined genes in terms of quick acetylation phenotype average daily doses of iGCS have a tendency to prevailing over the similar ones in patients from the groups

of comparison, and their triple administration is reliably higher.

Key words: bronchial asthma, *GSTT$_1$*, *GSTM$_1$*, gene polymorphism.

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Introduction. Nowadays the fact that chronic inflammation of the respiratory tract with the development of immune disorders underlies bronchial asthma (BA) pathogenesis is generally accepted. To achieve control over the disease, which is the purpose of treatment, requires a continuous anti-inflammatory therapy. In recent years “Global Strategy for Asthma Management and Prevention” [4], National Protocols of Giving Medical Aid to the Population [7**]** determine the notion of “achieving control over the disease**”** as a final purpose of an adequate therapy. Preventive (basic, anti-inflammatory, controlling) treatment of BA involves the following groups of medicines: inhalation glucocorticosteroids (iGCS), anti-leukotriene drugs, prolonged methylxanthines, antibodies against IgE and systemic glucocorticosteroids (sGCS), as well as inhalation β_2-agonists of a slow release considered as the means of the basic therapy only in combination with iGCS.

At the same time, BA basic therapy in children, directed to the increase of inflammatory activity of the respiratory tract which is clinically manifested by achieving and continuous maintaining the control over the disease, is often characterized by insufficient efficacy of the standard schemes. One of the important factors influencing the efficacy of the therapy are genotypic and phenotypic peculiarities of the child organism which are not considered by the standard therapeutic schemes, although, patients with different gen- and phenotypes of the disease require an individual approach to symptomatic and preventive treatment. Thus, genetic component is one of the valid ones in achieving control over the disease [9], as it forms a certain phenotype of BA, stipulates peculiarities of response to the environmental stimuli and pharmacotherapy [1, 10]. In particular, the spread of *GSTM$_1$* and *GSTT$_1$* gene deletions is found to increase in patients with BA enabling to draw a conclusion concerning an increased risk of BA and decreased lung function in case polymorphism of these genes is available [3, 5]. Certain relations of *GSTM$_1$* and *GSTP$_1$* genotypes with the indices of forced expiratory volume (FEV) are found, although current scientific literature does not present clear pathogenetic explanation of the effect of these genes [2, 8]. N-acetyltransferase is known to participate not only in the second phase reactions of xenobiotic biotransformation and metabolism of substances containing amino groups in their molecule [11], but it also plays an important role in metabolism of endogenous substrates

regulating the processes of bronchial spasm and initiating inflammatory response, such as serotonin, dopamine, and leukotriene E4 [6].

Objective. To investigate peculiarities of basic anti-inflammatory therapy and its efficacy in children with available or absent deletion polymorphism of *GSTM1* and *GSTT1* genes coding II phase enzymes of *GSTM1* and *GSTT1* xenobiotic detoxication considering their acetylation status.

Materials and methods. Keeping to the principles of bioethics 33 school children suffering from BA were comprehensively examined. They were divided into 2 clinical groups of comparison by the results of detection of the deletions in *GSTT1* and *GSTM1* genes and the rate of acetylation processes. The I group included 18 patients with the signs of quick acetylizers and absent deletions in the examined genes coding glutathione-S-transferase T_1 and M_1 (genotype *GSTT1+M1+)*. The second (II) group included 15 patients with the genotype *GSTT1+M1-*, *GSTT1-M1+* and *GSTT1-M1-* and the signs of a slow acetylation status. The groups were comparable by the main clinical characteristics. Thus, the onset of the disease in early childhood was determined in 21,0 % of children from I group and 33,3 % in II group of comparison, at the pre-school age – in 5,79 % and 13,3 % of patients respectively, at the school age – in 63,16 % and 53,3 % of the study respectively (in all the cases P>0,05).

To determine deletions in *GSTT1* and *GSTM1* genes capillary blood was tested by means of multicomplex polymerase chain reaction at the Department of Molecular Genetics and Biotechnologies at Yuriy Fedkovych Chernivtsi National University (Chief – Doctor of Biological Sciences, Professor R.A. Volkov).

The type of acetylation was determined by the results of sulfadimine urine test by means of photoelectrocolorimetric method. The output of acetylated sulfadimine was calculated by the difference between general and free fractions, and patients with the contents of acetylated sulfadimine less than 75% were considered as slow “acetylizers”, more than 75% - to quick ones respectively.

The level of BA control was determined by means of the following questionnaires:

1) By ACT-test (Asthma Control Test), containing 7 questions (3 of them are answered by parents) for children under 11, and 5 – for patients over 12. Every answer was estimated from 1 to 5 points. The total score higher than 20 was indicative of a complete control achieved, from 16 to 19 – a partial control, 15 and less – absent control over the disease.

2) By GINA-test containing six questions with alternative answers. The absence of daily, nocturnal symptoms, physical restrictions, administration of rapidly acting β2-agonists

less than twice a week and FEV_1 higher than 80% were estimated in 1 point; the above characteristics and FEV_1 less than 80% out of age norm were estimated in 2 points. Therefore, the total score 5 and more was indicative of a complete control over the disease, 6-8 points – a partial control, and higher than 8 points – uncontrolled BA.

3) By clinical-instrumental evaluation (CIE) scale assuming answers to 7 questions estimated from 0 to 4 points, and the indices of FEV_1 and peak volumetric rate (PVR). 10 and less points enabled to identify controlled BA, 11-16 points were associated with partially controlled BA, and higher than 17 – with uncontrolled course of BA.

Results and discussion. Atopic form of BA was found to prevail in the group of patients with changed II phase processes of xenobiotic transformations, mixed form – in children with high acetylation phenotype and without deletion changes in the examined genes. Thus, atopic form occurred in I group A in 36,84 %, mixed form – in 63,16 % of cases, in the group of comparison this distribution was the following: 86,67 % and 13,33 % respectively (in all the cases $P<0,001$). Therefore, the efficacy of basic therapy directed first of all to the control of atopic inflammatory process was expected to possess better indices in II group.

At the same time, in I clinical group moderately severe persisting course of BA was found in 57,9 % of cases, severe – in 42,1 % of cases, and mild persisting disease was not detected at all. In II group of comparison mild persisting asthma was found in 6,67 % cases, moderate and severe – in 40,0 % and 53,33 % of cases (in all the cases $P>0,05$). Therefore, in children with changed efficacy of xenobiotic detoxication system a tendency to more severe course of the disease is determined probably at the expense of deletion changes in genes of glutathione-S-transferase family and slow process of acetylation of allergens and other xenobiotics.

Similar tendencies were found in peculiarities of basic anti-inflammatory treatment according to Global Initiative for Asthma (GINA) [4], which were indicative of the necessity of more active controlling therapy in children of II group of comparison. Thus, therapy within the range of the first step was indicated for 22,2 % of patients from I group and only 13,3 % of children from II group of comparison, therapy within the range of the second range was indicated for practically 1/3 of patients from I group and half of children from II group (27,8 % and 46,8% respectively), treatment assumed by the third step was administered for half of patients from I group and 26,7% individuals from II group, and therapy included into the fourth step – only 13,3% patients from II clinical group of comparison. 77,78 % representatives of I clinical group 86,67 % patients from the group of comparison received basic treatment in the form of monotherapy with iGCS ($P>0,05$); 14,29 % and 23,08 %

patients respectively – therapy combined with prolonged β_2-agonists (P>0,05). Among patients without deletion gene polymorphism in terms of quick acetylation phenotype available beclametasone dipropionate was administered for 78,57 %, budesonide – only 7,14 % patients, the same number of children took fluticasone and combination of other medicines. In the group of patients with changes in xenobiotic detoxication system these drugs of the basic anti-inflammatory therapy were indicated with the following frequency: beclametasone dipropionate – in 76,92 % cases (P>0,05), fluticasone propionate – in 23,08 % of cases (P<0,05). Thus, fluticasone propionate combined with iGCS and β_2-agonists of a slow release was indicated more often for patients with available deletion changes of the examined genes and slow processes of acetylation.

At the same time, an average daily dose of iGCS was higher in patients without deletion polymorphism of the examined genes in terms of quick acetylation phenotype. Thus, an average daily dose of iGCS in the representatives of I group was (289,29±39,63) mcg, and in II group of comparison – (244,23±36,05) mcg (P>0,05).

Considering the fact that more than a half of children from II group of comparison had the signs of severe persisting BA and only 13,3 % of them received therapy according to the fourth step, the content of a comprehensive anti-inflammatory therapy deserves to be examined in details. The Table presents a qualitative frequency distribution of the use of low, mean and high doses of iGCS in children from the groups of comparison as medicines of basic anti-inflammatory controlling therapy of BA.

Table

Frequency of use of different doses of iGCS in basic treatment of BA in clinical groups of comparison (P±m)

Clinical groups	iGCS were not used	Low doses of iGCS	Mean doses of iGCS	High doses of iGCS
I group	22,2	42,9	35,7	21,4
II group	13,3	61,5	30,8	7,7
P	>0,05	>0,05	>0,05	>0,05

Note: P – probability of difference

In spite of the absence of statistically significant differences a clear tendency to prevailing use of iGCS low doses in II group of comparison is seen. The representatives of this group received high doses of these drugs practically three times as less than children with

preserved processes of xenobiotic transformation. There were no statistically significant differences found in the frequency of indication of iGCS during twenty-four hours in the groups of comparison: (1,67±0,26) times a day against (1,6±0,2) times a day (P>0,05). At the same time, 14,29 % children from I group and 23,08 % representatives of II clinical group received iGCS as a single inhalation (P>0,05), these basic medicines were indicated twice a day for 57,1 % and 69,2 % patients (P>0,05), and three times a day – 28,6 % and 7,7 % patients from the clinical groups of comparison respectively (P<0,05).

In estimation of the degree of control over the disease according to GINA questionnaire in I group the obtained results coincided with and were distributed in half between partially controlled and uncontrolled BA. According to ACT-test in I group of children aged under 11 and over 12 in 100% of cases the course of asthma was found to be partially controlled. On the contrary, by the results of CIE scale a controlled course of BA occurred in this group in 17,65 % of patients, partially controlled asthma – in 29,41 %, and uncontrolled disease – in 52,94 % of cases.

In the group of patients with deletion polymorphism of $GSTT_1$ and $GSTM_1$ genes and slow acetylation status by ACT-test in all the children under 11 the course of asthma appeared to be partially controlled (similar to that of I group), in patients over 12 in 33,33 % of cases the course of asthma was found to be partially controlled, and in the rest 66,6 % - controlled. Although by the results of CIE scale controlled BA was found in 16,67 % cases (P>0,05), partially controlled – in 25,0 % (P>0,05), and uncontrolled – in 58,33 % of cases (P>0,05). Figure presents the results of assessment of a controlled course of BA in children depending on the availability or absence of deletion polymorphism of $GSTT_1$ and $GSTM_1$ genes considering the rate of acetylation processes.

Thus, ACT-test enabled to determine the largest number of cases of a controlled course of BA, and CIE scale – uncontrolled course, which is likely associated with incomplete self-estimation of the condition (teenagers over 12 in particular) while using ACT questionnaire, and at the same time, objectification of the results of the questionnaire by means of spirometry examinations using CIE scale. Both the latter scale and GINA questionnaire enabled to determine uncontrolled BA course in half of the cases among representatives of both clinical groups of comparison, requiring the correction of the recommended means of the basic therapy.

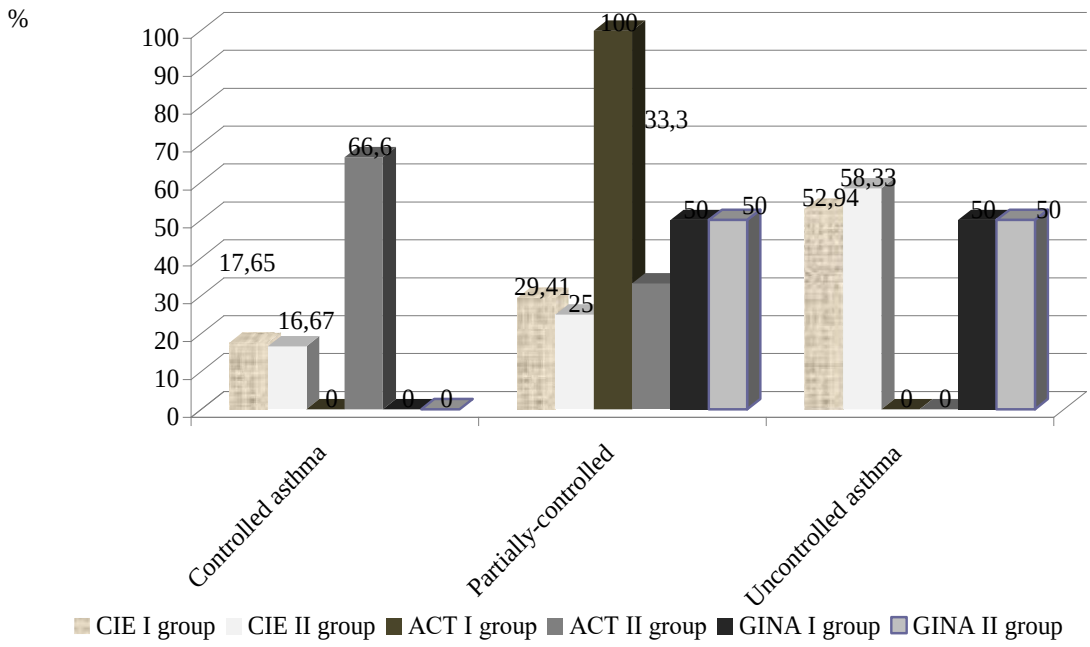

Figure. Frequency of registration of different variants of bronchial asthma course by the level of control in children of the clinical groups of comparison.

Conclusions

1. In patients of a school age suffering from bronchial asthma with available deletion polymorphism of *GSTT1* and *GSTM1* genes associated with slow acetylation phenotype anti-inflammatory therapy should be intensified emphasizing on much higher “step” or by means of addition of other anti-inflammatory drugs.
2. In patients without deletion polymorphism of the examined genes in terms of quick acetylation phenotype average daily doses of iGCS have a tendency to prevailing over the similar ones in patients from the groups of comparison, and their triple administration is reliably higher.
3. Uncontrolled course of bronchial asthma in both clinical groups was determined in half of the patients by means of CIE scale and GINA questionnaire, and in every case while using ACT-test, which is indicative of the necessity to objectivize the control over the disease by means of spirography method.

References:

1. Blakey J., Halapi E., Bjornsdottir U.S. Contribution of ADAM33 polymorphisms to the population risk of asthma. Thorax. 2005; 60: 274-276.

2. Carolan B.J., Sutherland E.R. Clinical phenotypes of chronic obstructive pulmonary

disease and asthma: Recent advances. The Journal of Allergy and Clinical Immunology. 2013; 131(3): 627-634.

3. Ivaschenko T.E., Sideleva O.G., Baranov V.S. Glutathione- S-transferase micro and theta gene polymorphisms as new risk factors of atopic bronchial asthma. J. Mol. Med. 2002; 80: 39–43.

4. Global strategy for asthma management and prevention (GINA 20124). Access mode: http://www.ginasthma.org/local/uploads/files/GINA_Report_2014_Aug12.pdf

5. Kamada F., Mashimo Y., Inoue H. The GSTP1 gene is a susceptibility gene for childhood asthma and the GSTM1 gene is a modifier of the GSTP1 gene. Int. Arch. Allergy Immunol. 2007; 144 (4): 275-286.

6. Makarova S.Y., Vavylyn V.A., Liakhovych V.V. Allel NAT2*5 – faktor ustoichyvosty k zabolevanyiu bronkhyalnoi astmoi u detei. Biulleten эksperymentalnoi byolohyy y medytsynы. 2000; 6: 677-680.

7. Nakaz MOZ Ukrainy № 868 vid 08.10.2013 r. «Unifikovanyi klinichnyi protokol pervynnoi, vtorynnoi (spetsializovanoi) medychnoi dopomohy. Bronkhialna astma». 2013. Rezhym dostupu http://moz.gov.ua/ua/portal/dn_20131008_0868.html

8. Reddy P. Naidoo R.N., Robins T.G. GSTM1 and GSTP1 gene variants and the effect of air pollutants on lung function measures in South African children. Am. J. Ind. Med. 2012; 55 (12): 1078-1086.

9. Ruscoe J.E., Rosario L.A., Wang T. Pharmacologic or genetic manipulation of glutathione S-transferase P1-1 (GSTpi) influences cell proliferation pathways. J. Pharmacol. Exp. Ther. 2001; 298: 339–345.

10. Taylor D.R., Bateman E.D., Boulet L-P. A new perspective on concepts of asthma severity and control. Eur. Respir. J. 2008; 32(3): 545-554.

11. Windmill K.F., Gaedigk A. Localization of N-acetyltransferases NAT1 and NAT2 in human tissues. Toxicological Sciences. 2000; 54: 19-29.