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Differences in clinical test results of patients with bronchial asthma and environment of living. Researches in rural and urban area in the east part of Poland

Asthma is currently considered and defined as a chronic inflammatory disorder of airway mucosa resulting in airflow limitation, which – at least in the majority of patients – is reversible. Recent studies have revealed that bronchial biopsies from patients with asthma are characterized by: shedding of the surface epithelium, thickening of the reticular basement membrane, thickening of the bronchial smooth muscle, glandular hyperplasia, and marked cellular infiltration, in most cases dominated by eosinophils (18).

Asthma diagnosis, first suspected on the basis of characteristic symptoms, is facilitated owing to various *in vivo* and *in vitro* tests. Lung function spirometry tests and specific provoking test are conducted in order to determine the level of obturation and reversibility of abnormalities. From other *in vivo* assays, skin prick tests are particularly useful. *In vitro* tests allow for determination of eosinophil level in peripheral blood and sputum as well as sIgE concentration in patient's serum by performing immunosorbent tests with anti-IgEs conjugated with a marker, which can be radioisotopic (RAST), enzymatic (ELISA), fluorescent or luminescent (MAST-CLA).

In spite of major advances in the recognition and treatment of asthma, the prevalence and severity of this condition have continued to increase over the last 20 years, especially among children and adolescents living in industrialized area (4). Western Europe has the highest prevalence of asthma in the world (8). According to the data from a population-based survey, there has been a 2–4% annual increase in asthma morbidity rates in most of the European countries over the past 15 years (15). Despite obvious complexity of the reasons for this increase, environmental exposure to allergens and pollutants, changes in patterns of medical treatment as well as psychosocial stress of living in poor inner-city neighbourhoods can be taken into account. Although higher prevalence of allergic diseases and increased levels of IgE antibodies against inhaled allergens have been reported for subjects living in urban areas than for those living in rural areas, it has to be elucidated whether rural lifestyle protects against allergy or whether urban pollutants contribute to allergy (11).

This paper presents evaluation of immunological and clinical parameters in the course of asthma of patients from rural and urban area in the east part of Poland.

MATERIAL AND METHODS

The research involved 60 subjects with bronchial asthma symptoms, who reported at the Ambulatory Chest Clinic of the Pulmonary Department of the Medical University of Lublin either to confirm illness diagnosis or to continue medical treatment. Thirty-six patients (21 females and 15 males) live within the Lublin boundaries (mean age of 31.13 ± 13.41), and 24 (14 females and 10 males) – in the country (mean age of 37.09 ± 13.87). Studies were carried out in the period of a low pollen count. The subjects were given a detailed questionnaire based on one of the International Union Against Tuberculosis and Lung Disease (IUATLD) to complete (2). It concerned the symptoms, allergies, family history, smoking, home environment, socio-economic status, education and medication use. The patients received skin prick tests with eighteen allergens. Besides blood eosinophil level, serum total and specific IgE level and spirometry testing using standard instruments, reagents and protocols, were examined.

Skin prick tests. For allergy testing, the standardized set of allergens (Alergopharma, Nexter, Katowice, Poland), including a negative and a histamine positive controls, was used. The results of skin pricks were read after 15 minutes. The mean diameter of above four mm was considered as a positive skin test. The examined patients were not treated with antihistaminic drugs.

Eosinophils. Blood eosinophil level was determined in whole blood direct microscopic preparation stained with Dunkier's solution. Eosinophilic leucocytes were counted using Fush-Rosenthal's chamber. The value of 150–250 of eosinophils per blood cubic millimetre was considered as normal.

Total and specific IgE. Total and specific IgE serum concentration was gauged using the Pharmacia CAP System (Pharmacia Diagnostics, Uppsala, Sweden). Specific IgE was measured against single allergens of selected grass (*Secale cereale* – denoted as g12, and *Hordeum vulgare* – g201) and weed pollens (*Artemisia vulgaris* – w6, and *Utrica dioica* – w20), of moulds (*Penicillium notatum* – m1, *Cladosporium herbarum* (*Hormodendrum*) – m2, *Aspergillus fumigatus* – m3, *Mucor racemosus* – m4, *Alternaria alternata* (*A. tenuis*) – m6, and *Aspergillus niger* – m207) and yeast (*Candida albicans*, m5) as well as against allergen mixes of grass pollens (*Dactylis glomerata*, *Festuca elatior*, *Lolium perenne*, *Phleum pratense*, *Poa pratensis* – gx1) and house dust (Hollister-Stier Labs., *Dermatophagoides pteronyssinus*, *Dermatophagoides farinea*, *Blatella germanica* – hx2). Detection of specific IgE (above 0.35 kU_A/l) was treated as the definition of sensitization.

Lung function test. Spirometry testing was performed using a flow-type spirometer (pneumotachometer MES, Kraków, Poland) with computer data analysis (LungTest 100 software), when taking the blood sample. The forced expiratory volume in one second (FEV_1) value below 70% of the predicted normal value (PV) was considered as a serious disturbance of the lung function.

Statistics. The Chi-squared test was used in the univariate analysis. A Men-Whitney U-test determined the magnitude of group differences. p-value <0.05 was considered as significant. All statistical analyses were performed with Statistica 5.0.

RESULTS

The examined groups of patients were similar regarding their age and sex. Cigarette smoking was reported by four individuals living in a rural area, and by two city subjects. All patients reported cough and dyspnoea attacks accompanied by whistling rales heard when breathing in the last 12 months. Allergic rhinitis symptoms, co-existing with asthma, were more frequent ($p<0.05$) in patients living in the city (i.e., 33 individuals, 91.7%) than in those from rural regions (i.e., 17 subjects, 71%). Asthma symptoms occurred insignificantly earlier ($\leq$15

years of age) in the inner-city patients (N=15, 41.7%) than in rural patients (N=7.29%). Positive skin prick tests results were found more often in the city patients (i.e. 31 subjects, 86.1%) compared to the examined country inhabitants (i.e. 20 subjects, 83.3%), though the difference was statistically insignificant. Spirometry testing revealed considerable disturbances (FEV_1<70% of PV) in 25% of patients living in urban area (N=9) and in 50% of subjects from the country (N=12; p<0.05). All patients were treated with $beta_2$-agonists. Inhaled corticosteroids were administered to 53 subjects, 23 from the rural and 30 from the urban area, with statistical insignificance of this variable. When examined, six patients living in the country (25%) and four inner-city subjects (11.1%) required oral corticosteroid administration, again without statistical significance of the variable.

Peripheral blood eosinophil level was analogous in both groups (mean value of 369/mm^3). The inner-city patients had insignificantly higher serum total IgE concentration (687.3 ± 1663 kU_A/l) than the rural subjects (364.2 ± 718.5 kU_A/l). As far as the level of specific IgE in patients' sera is concerned, it can be said that five subjects (20.8%) from rural regions and 21 patients (58.3%) from urban area have positive results of specific IgE against gx1 (p<0.005). The positive result obtained for 11 subjects (50%) living in the country and 21 inner-city patients (58.3%) when testing the level of specific IgE against hx2 was statistically insignificant. The level of specific IgE against grass pollens: g12 and g201 was significantly (p<0.005 and p<0.05, respectively) higher in the city patients (10.5 ± 25,8 kU_A/l, 11.6 ±25.7 kU_A/l, respectively) compared to the rural patients (0.1 ± 0.35 kU_A/l, 0.1 ± 0.4 kU_A/l, respectively). The level of specific IgE against w20 was significantly higher (p<0.05) in the city patients (1.8 ± 6.1 kU_A/l) compared to the rural patients (0.1 ± 0.2 kU_A/l). Analogous differences, although statistically insignificant, referred to the concentration of IgE against w6. The level of IgE against house dust allergens and mould was similar in both examined groups (Fig. 1).

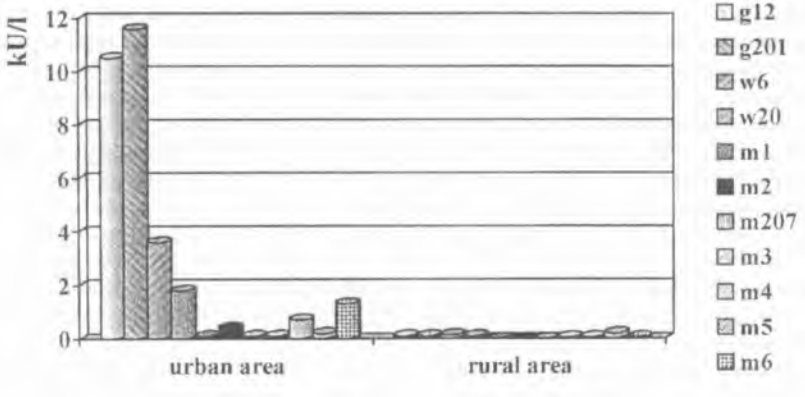

Fig. 1. The level of specific IgE against selected grass, weed, mould and yeast allergens of patients living in rural and urban area

Eosinophil count and level of specific IgE against g12 and g201 were significantly higher (p<0.05) in patients with allergic rhinits symptoms (446 ± 446 cells/mm^3, 9.6 ± 26 kU_A/l and 10.9 ± 26 kU_A/l, respectively) than in subject without symptoms (73 ± 41 cells/mm^3, 0.05 ± 0.05 kU_A/l, 0.05 ± 0.15 kU_A/l, respectively). High levels of specific IgE against g12 nad g201 are also connected with early asthma onset (under 15) in comparison with later beginning of the disease (14 ± 32.5 kU_A/l and 13 ± 30.5 kU_A/l versus 2.5 ± 9.4 kU_A/l and 4.3 ± 13.7 kU_A/l, p<0,05, respectively).

No smoking influence or dependence between the sex and clinical test results was found.

DISCUSSION

Several studies which aimed at comparing asthma patients from the rural and urban areas, both children and adults, revealed that a higher morbidity rate of this condition is reported for the city subjects. On the basis of the research, which indicated that the prevalence of hay fever and asthma was found to be lower in children from rural areas than in children from urban environment, Riedler et al. hypothesized that living on a farm might be protective against allergic sensitisation and allergic diseases development. The results showed significantly lower prevalence of hay fever ($p=0.0002$), asthma ($p=0.017$) and positive skin prick reactivity to at least one of the common local allergens ($p=0.001$) in children living on a farm than in children from an urban area of Austria. The authors' conclusion is that the lower morbidity rate of hay fever, asthma and allergic sensitisation in children from rural areas may be explained by the development of immunotolerance or the stimulation of Th_1 cells and suppression of Th_2 cells by increased exposure of those to microbial antigens in the stables or farmhouses (13).

Some investigators report, however, that asthma is more prevalent in rural than in urban areas. As a research of Woods et al. showed, in comparison with Melbourne, asthma morbidity rate was higher in rural south-western New South Wales, although a pattern of exacerbations differed between these two Australian regions. Subjects living in the country reported significantly higher prevalence of nocturnal dyspnoea ($p<0.01$), chronic bronchitis ($p=0.03$), an asthma attack in the previous 12 months ($p<0.001$), ever having asthma ($p<0.001$) and physician-diagnosed asthma ($p<0.001$) compared to those from Melbourne. New South Wales respondents also reported a higher prevalence of smoking ($p<0.05$) and they smoked more cigarettes on average ($p<0.001$). However, among the subjects with 'asthma attacks in the last 12 months', Melbourne residents experienced higher frequency of attacks ($p<0.05$). Besides, air pollution annoyance was raised when compared with the rural area (19).

The higher asthma prevalence in inner-city patients when compared with subjects from rural areas may also be confirmed by the research, which indicated that in urbanised former rural region, asthma morbidity rate increased. Filipiak et al. suggested that the increasing urbanization, along with the decrease in the number of farms in southern Germany might be associated with growing prevalence of allergic diseases (5). Having compared the morbidity rate of these conditions in farmers, and rural, suburban and urban residents in the small geographical area of Augsburg, they showed that in comparison to rural subjects, urban population had an increased risk of allergic rhinitis, atopic sensitization and sensitization against pollen. No difference in the risk of asthma and a decreased risk in the sensitization against house dust mite was found in this research. It is also worth mentioning that the suburban residents did not differ from urban residents. On the basis of the results, the authors concluded that a farming environment and rural lifestyle might be associated with the earlier-mentioned unknown protective factors affecting the prevalence of allergies (5, 11, 13).

Having examined two groups of subjects (i.e., from rural and urban areas) we could show that in spite of the fact that the examined patients reported similar symptoms of illness, its clinical and immunological manifestation were different. The highest risk of allergic rhinitis, atopic sensitization and sensitization against pollen was observed in inner-city patients, whereas there were no differences in sensitization against house dust allergens as well as moulds and yeast between examined groups. The contradictory findings of Woods et al. may result from the influence of different factors on Australian population as compared to European.

Atopy defined as a hereditary predisposition to abnormal IgE production, and therefore positive skin test reactivity to inhalant allergens is found in the majority of asthma patients, especially children, with stronger implication for inner-city individuals (5). The association between atopy and asthma outcome is, however, unclear. Some studies pointed to a negative effect (3) and some to no effect (12). Many of these studies define asthma outcome as the presence or absence and the severity of respiratory symptoms, especially these reporting a negative atopy effect on outcome. Only a few researches considered the possible impact of

atopy on longitudinal changes in lung function in asthma patients finding that skin test-positivity is associated with an accelerated decline in FEV_1 (7, 14). At the same time, the poor correlation between asthma symptoms and the degree of airway obstruction as determined by FEV_1 was shown (17). Following the treatment, subjective improvement in asthma symptoms may occur without improvement in airway obstruction. In our study, rural area patients with lower IgE levels experienced more serious lung function disturbances in spirometry testing (which should not be treated as reliable enough because of being performed once only) than inner-city subjects, though the prevalence of asthma was found to be higher in the latter environment.

Atopy is a risk factor for symptomatic asthma. However, in both children and adults with established asthma, it seems reasonable to conclude, on the basis of the studies mentioned above, that atopy is not an independent determinant of prognosis considering lung function. It is suggested that inflammatory processes in the airways of asthma patients run their own courses irrespective of the subjects' atopic status with a particular emphasis put on T-cell cytokine production, which seems to be the main factor in determining the degree of airway inflammation and hyperesponsiveness (9). Moreover, the ability of an allergen to induce IgE response is largely independent of biological function. The critical elements appear to be allergen dose, route of exposure and the genetic predisposition of the host, especially the allergen processing mechanisms resulting in Th_2 responses.

Atopic patients are very often diagnosed to have allergic rhinitis co-existing with asthma. Clinical observations suggest that allergic rhinitis and asthma occur together more often than expected by chance alone but the data are scanty. It was noticed that the probability of an asthmatic having allergic rhinitis is higher than the probability of an individual with allergic rhinitis having asthma, which seems to be due to the natural history of two conditions, that is hay fever and perennial rhinitis. Hay fever appears to be associated with seasonal asthma, whereas perennial rhinitis – with house dust mite asthma, as has been confirmed by skin-prick tests. Higher asthma prevalence was observed in subjects with allergic rhinitis confirmed by skin-prick testing to common aeroallergens compared to those with nonallergic rhinitis (1). The examined inner-city patients reported more allergic rhinitis symptoms than rural subjects, which was confirmed by more often determined positive skin-prick tests with allergens against pollens, high sIgE levels against g12 and g201 and increased eosinophil concentration.

Atopic asthma is also characterized by its early onset. However, the results of studies concerning the significance of age at which asthma begins as a predictor of subsequent disease appeared to be contradictory. Some studies showed that an early beginning of asthma is associated with a negative outcome (16). Other researches, however, indicated that an early onset of symptoms is a predictor of a positive outcome (6), whereas some studies found no particular effect of age at onset of symptoms on prognosis (10). Other possible risk factors for the incidence of respiratory symptoms and asthma include heredity, sex, working exposures, smoking habits, air pollution, and socioeconomic status.

Our study shows important differences between the clinical manifestation of asthma in rural and inner-city patients. It also indicates that different aetiology may play an important role in asthma development. Different courses of asthma require different approaches towards patients from these two groups. This is particularly important in the case of country inhabitants with the poorer asthma control, which may lead to stronger lung function disturbances as well as to the necessity of using oral corticosteroids.

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SUMMARY

The objective of the research was to evaluate immunological and clinical parameters in bronchial asthma patients from rural and urban areas in the east part of Poland. The study comprised 55 subjects reporting from November 2001 to January 2002 at the Pulmonary Department in order to have the disease diagnosed or to continue the treatment. The type of symptoms was determined on the basis of the standardized questionnaire. The patients were subjected to spirometry and skin prick tests. Blood eosinophil number as well as total and specific IgE levels were also evaluated. Significant ($p<0.05$) disturbances in respiratory parameters were found in 23% of patients living in the city and in 50% of subjects from the rural area. The level of IgE against grass pollens was significantly ($p<0.01$) higher in urban residents. The level of IgE against house dust was similar in both groups. Our study shows important differences between clinical manifestation of asthma in rural and inner-city patients and indicates that different aetiology may play an important role in asthma development.

Zróżnicowanie wyników testów klinicznych w astmie oskrzelowej a środowisko chorych. Badania w obszarach wiejskim i miejskim regionu lubelskiego

Celem pracy była ocena parametrów immunologicznych i klinicznych, obrazujących przebieg astmy oskrzelowej u osób zamieszkałych w Lublinie i poza terenem miejskim. Badaniami zostało objętych 55 osób z objawami astmy oskrzelowej, które zgłosiły się do Kliniki celem potwierdzenia diagnozy choroby lub kontynuacji leczenia. Badania prowadzone były od listopada 2001 r. do stycznia 2002 r. po sezonie pylenia roślin. Rodzaj objawów występujących u pacjentów został określony na podstawie ankiety sporządzonej na bazie kwestionariusza International Union Against Tuberculosis and Lung Disease (IUATLD). U chorych przeprowadzono badanie spirometryczne i testy skórne, oceniono liczbę eozynofili we krwi oraz poziom całkowitego i swoistych IgE. Badania spirometryczne wykazały istnienie znacznych zaburzeń u 23% chorych żyjących w mieście i u 50% pacjentów ze wsi ($p<0,05$). Poziom IgE swoistych skierowanych przeciw alergenom pyłków traw i zbóż był istotnie wyższy ($p<0,01$) u mieszkańców miasta. Poziom IgE przeciw alergenom roztoczy kurzu domowego był podobny w obu grupach. Uzyskane wyniki wydają się potwierdzać występowanie większego odsetka postaci atopowych astmy oskrzelowej w regionie miejskim. Może to sugerować, że w zależności od środowiska życia różne czynniki etiologiczne wywierają wpływ na rozwój astmy oskrzelowej.