

Interfering factors to sensitivity and specificity of bronchial reactivity in children

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Bronchial reactivity to carbachol, estimated by the PD₆₅, the provocation dose of inhaled carbachol inducing a 65% increase of baseline respiratory resistance, was measured in 355 asthmatic children (age 8.7 ± 2.6 years) and 149 healthy children (age 11.8 ± 2.3 years). The frequency distribution of PD₆₅ showed apart from 2 minima at 480 µg and 960 µg 2 significant peaks at 240 µg ($p = 0.003$) and 1200 µg ($p = 0.01$) to carbachol, reflecting a hyperreactive class (PD₆₅ < 480 µg), an intermediate class ($481 \mu\text{g} < \text{PD}_{65} < 960 \mu\text{g}$) and a normo-reactive class (PD₆₅ > 961 µg). The intermediate class is characterised by a considerable overlap between “health” and “disease”. However, taking into account the degree of air pollution in which healthy children are living (best related factor to specificity) and the type of initial lung function disorder in asthmatic children, (the MEF₅₀ to be the best related factor for specificity), weighed specificity and sensitivity improved from 64 to 88%. Evaluating bronchial responsiveness in children, such interfering factors should be strongly considered.

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Bronchial asthma is the most common chronic disease in childhood (1–3), clinically characterized by recurrent episodes of cough, wheezing and dyspnoeic attacks, physiologically expressed by pulmonary hyperinflation and/or reversible airway obstruction (4–6). In addition to genetic factors (7–9), exposure to house-dust mite allergens in early childhood is an important determinant for subsequent development of asthma (10) and, atopy is known to have an important association with asthma (11) and bronchial hyperreactivity (BHR) (12). A quantitative relation between skin prick test reactions has been shown to contribute to the risk of BHR (12, 13) and BHR may continue to persist into late childhood (14). Since there is a relationship between the degree of airway inflammation and bronchial reactivity (BR) at least shown in adults (15–18), detection of BR as an indirect indicator of the onset and degree of an inflammatory process (19–22) also became of interest in children (22–26).

There are, however, different concomitant factors, such as the state of atopy (12, 27), history of bronchiolitis or recurrent croup (28), respiratory infections, rhinitis (or recent allergen exposure) (27, 29) or environmental irritants (15, 30, 31)

which influence specificity and sensitivity of BR. Even healthy subjects may have hyperreactive airways (15, 23, 31, 32). Although predisposing factors for bronchial challenge testing in children has been previously studied (20, 21, 24–26, 33), the question has never been posed whether factors such as age, sex, positive family history of atopy, outdoor air pollution and/or parental smoking may influence the specificity of BR in healthy children or to what extent sensitivity of BR is affected by the type of functional abnormalities in asthmatic children. Therefore, the present study was designed to evaluate the influence of such concomitant factors to BR in healthy and asthmatic children.

Patients and methods

Asthmatic children

A total of 355 children with exogen allergic, mild to moderate asthma, seen over a time span of 12 months at our asthma center of the University Children's Hospital in Berne, Switzerland, (220 boys and 135 girls, mean age 8.7 ± 2.6 years, range 5 to 17 years) were enrolled in this study. Asthma was diagnosed when a history of wheezy attacks,

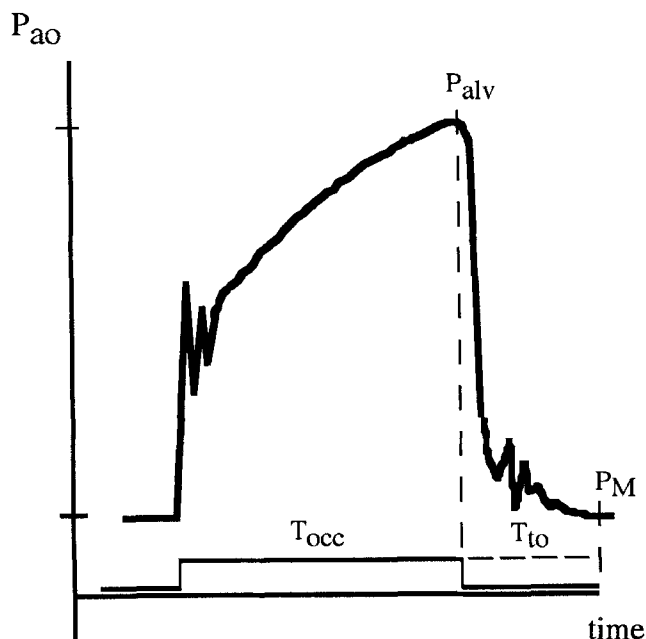


Fig. 1. Setting for calculation of the resistance of the respiratory system (R_{int}) from the interruption curve. (P_{alv} : alveolar pressure equal to open airway pressure P_{ao} at equilibrium; P_M : mouth pressure equal to P_{ao} after shutter opening; T_{occ} : shutter occlusion time = 100 ms; T_{to} : take over time = 75 ms).

elevated IgE-levels (PRIST), positive specific IgE (RAST) for one single or more allergens, and laboratory documentation of airway obstruction, which reversed with sympathomimetics, were present (2, 4–6). At the time of testing, all patients on regular inhalative treatment, were under stable clinical conditions (no signs of asthma clinically detectable) and there was no asthma episode within 2 weeks prior to the investigations. Bronchodilator treatment was withheld 12 h, inhalative steroids or disodium cromoglycate 24 h prior to the test. None of the patients was on xanthine treatment, systemic steroids or oral beta-2-agonists.

Healthy children

As control subjects, 149 healthy, non-atopic and non-asthmatic children (62 boys and 87 girls, mean age 11.8 ± 2.3 years, range 7 to 16 years) were eligible from a population of 312 school children investigated earlier for other purposes (31). According standardized case histories, physical examinations and determination of serum-IgE-levels (Pharmacia) and RAST (Phadiatop) for grass pollen, house dust mites and animal dander, all subjects suspected of having infections, atopic diseases, prematurity and consecutive diseases, cystic fibrosis and cardiac diseases could be excluded. The collective investigated therefore fulfills the criteria that the children were absolutely healthy, free from any signs of respiratory symp-

toms, free from any signs of viral infections and free from any influences stemming from vaccinations during the last 6 weeks prior to the test. All of the subjects' parents gave their informed consent for the examinations and investigations, and the study was approved by the local ethics committee.

Environmental factors

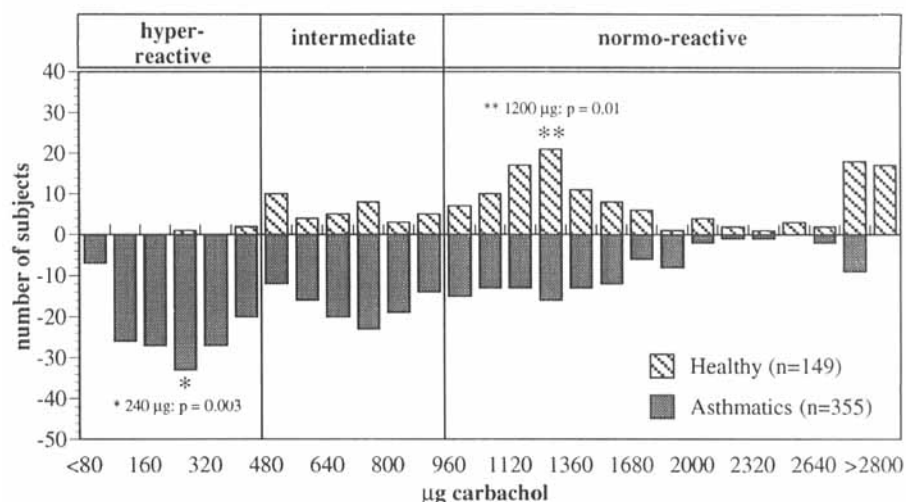
To investigate the potential effects of air pollution on respiratory health, the children were studied according to their living area, which was divided into two zones with significantly different concentrations of atmospheric NO_2 , measured over a period of 10 months prior to the clinical investigations. In the more urban area the mean NO_2 was $36.2 \pm 9.5 \mu g/qm$ compared to the mean of the more rural area of $26.2 \pm 10.4 \mu g/qm$ (31). Although the investigations were taken during one week in May, the atmospheric pollen concentration was low because a rain period of 4 weeks immediately preceded the week of investigations.

Lung function tests and protocol for evaluation of BR

Following the medical interview and a physical examination, pulmonary function tests were performed. The functional abnormalities were evaluated by whole-body plethysmography (4–6). Thoracic gas volume (TGV) was taken as an estimate of the degree of pulmonary hyperinflation, and airway resistance (R_{aw}) as an estimate of the degree of bronchial obstruction. The measurements were done with the subject seated in an air conditioned, constant volume, variable pressure whole-body plethysmograph (Jaeger, Würzburg, FRG) with BTPS (body temperature pressure saturated) conditions. For TGV static respiratory efforts against the shutter were performed at low frequency and TGV was calculated by the Boyle's Law technique. TGV values were corrected for resting expiratory levels of the spirogram. The instrumental dead space of the mouthpiece and the estimated stomach gas volume were subtracted. Based on thoracic gas volume (TGV) at end-expiratory resting level, static lung volumes, the total lung capacity (TLC), the vital capacity (VC), and the residual volume (RV) were obtained. R_{aw} was measured during quiet breathing (no panting) of air from a Douglas bag. Flow-volume curves gave the forced expiratory volume in one second (FEV_1) and the maximal expired flow at 50% of vital capacity (MEF_{50}). Lung function data were expressed as percent of predicted values (34).

Inhalative provocation tests were performed by the Bronchoscreen (Jaeger, Würzburg, FRG), a

Fig. 2. Histogram of 355 asthmatic children compared with 149 healthy controls. The columns indicate the number of children with PD_{65} at the corresponding carbachol level.



computerized apparatus with a combined nebulizer-shutter head measuring with each breath changes in the resistance of the respiratory system (R_{int}) by the interrupter technique. During calm breathing this technique, originally developed by van Neergaard and Wirz (35), further improved by Veuilleumier (36) and others (37–40), measures R_{int} after interruption of the airflow during 100 ms. From the “opening limb” of the interruption curve obtained during each relaxed expiration (Fig. 1), the pressure in the alveolar compartment after closing the shutter (P_{alv}) was obtained indirectly by measuring the airway opening pressure (P_{ao}) during free flow at pressure equilibrium (37, 38). R_{int} was determined using the formula $R_{int} = (P_{alv}/P_M - 1)R_{ref}$ where P_M was the pressure measured 75 ms after opening the shutter (38). R_{ref} was a constant known serial resistance of the calibration set. With the subject seated, noseclip in place, the subject breathing of conditioned ambient air was introduced to allow growing accustomed to the apparatus. Baseline measurements of $R_{int}(0)$ were performed by inhaling 10 to 20 breaths of physiologic NaCl solution (0.9%) from the built-in dosimeter. Then, carbachol-chloride in saline buffered to pH 7.0 with sodium bicarbonate (Merck Inc. Darmstadt, FRG), was used in a low concentration solution of 1.25 mg/ml during the first 10 breaths, then in higher concentration of 2.5 mg/ml until the end of the test procedure. This protocol was used for safety reasons to immediately detect children reacting to very low doses of carbachol. Generation-time for aerosolization was 0.5 s. Multiple nebulizer output measurements revealed a mean output of 44.1 (SD 5.5) mg solution per 10 breaths, and 23.1 (SD 1.7) mg per 5 breaths. Mean values of R_{int} from 5 breathing cycles were calculated in relation to the cumulative dose at each level of carbachol. Special care was taken to increase the dose gradually, to achieve progressive

small changes in R_{int} . Using this procedure a dose-response curve was obtained and the provocation procedure was terminated when an increase of $R_{int} 0$ to 185% was reached or earlier, when symptoms occurred. Thereafter, inhalation was switched to a bronchodilator.

Statistical methods

Data were analyzed by the SAS statistical package for PC and by the SYSTAT 5.2 for Macintosh. Significance of the peaks in the frequency distribution of the PD_{65} has been analyzed by chi-square tests. For decomposing a potential correlation or covariance matrix and hence for the evaluation of concomitant factors influencing the distribution of PD_{65} in healthy and asthmatic children principal component and factor analysis were performed. As multivariate procedure for detecting natural groupings in data, cluster analysis were performed. Relationships were explored using the chi-square analysis of contingency tables and multiple linear regression analysis. Sensitivity (s) defined as the capacity of a test to positively identify those persons with bronchial asthma was calculated as the ratio of the true positive to the sum of the true positive plus false negatives for the asthmatic population. Specificity (f) defined as the capacity of a test to exclude as healthy those subjects who did not have asthma was calculated as the ratio of the true negatives and the sum of the true negatives plus false positive in the healthy population (41). One analytical mathematical approach to establish relevance of such a test is to depict the intersection point of the declining f -curve with the increasing s -curve of cumulative percent response. This intersection point represents the cut off value, where best values for both, the s and the f are obtained, defined as weighed s and f (see figures in the result section) (21, 27).

Table 1. Means and standard deviations (SD) of baseline lung function of 355 asthmatic children within 4 functional groups. (Definition of functional groups see text; MEF₅₀: maximal expiratory flow at 50% vital capacity; Raw: airway resistance; TGV: thoracic gas volume)

Functional groups		n	TLC	TGV	Raw	MEF ₅₀
			mean $\pm$ SD in % pred.			
S	Small airway obstruction MEF ₅₀ < 80% pred.	95	100.3 18.0	110.0 22.0	144.6 49.4	67.9 9.3
C	Central airway obstruction Raw > 130% pred.	90	101.9 14.8	96.9 18.2	158.2 24.7	99.6 17.4
H	Pulmonary hyperinflation TGV > 130% pred.	34	111.5 15.9	144.1 13.1	78.6 24.7	114.6 24.3
N	Normal lung function MEF ₅₀ > 80% pred. and Raw < 130% pred. TGV < 130% pred.	136	100.6 12.8	99.6 15.1	99.1 18.2	107.5 19.1
All		355	102.5 14.3	105.8 22.4	98.3 18.8	95.3 26.8

Results

Frequency distribution of PD₆₅, sensitivity and specificity

The distribution of the PD₆₅ of the 149 healthy and 355 asthmatic children is presented in Fig. 2. There is a multimodal distribution with 2 minima at 480 μ g and 960 μ g and 2 significant peaks at 240 μ g ($p = 0.003$) and 1200 μ g ($p = 0.01$) to carbachol when compared to an equal distribution. Based on this significant maxima-minima distribution and for practical reasons 3 classes of BR can be defined: Class 1 can be defined as "hyperreactive", covering the range of 0–480 μ g carbachol and including 143 subjects, 3 healthy (2.1%) and 140 asthmatic children (97.9%). Class 2 is characterized by a wide overlap between healthy and asthmatic children and, therefore, defined as "intermediate class", covering the range of 481–960 μ g carbachol in which 140 subjects, 35 healthy (25.0%) and 105 asthmatic children (75.0%) are found. Class 3 considered "normo-reactive" children and covers the range >961 μ g carbachol, containing 221 subjects, 111 healthy (50.2%) and 110 asthmatic children (49.8%). Comparing the responder rate of the 3 classes of BR there were significant differences between healthy and asthmatic children (all the three: chi-square = 98.5, $p < 0.0001$; Class 1 versus Class 2: chi-square = 31.9, $p < 0.001$, Class 2 versus Class 3: chi-square = 22.6, $p < 0.0001$). Calculation of cumulative f and s for PD₆₅ 95% true negatives are reached at a level at 480 μ g carbachol and 95% true positive were within the range of up to 1920 μ g carbachol. The weighed f and s was reached at 960 μ g carbachol with a cumulative response of 72%.

Factors influencing BR in asthmatic children

To show individual values of lung function parameters numerically free from sex, age, and height bias, lung function data were expressed as per cent predicted ($\pm$ SD). Having assessed principal component and factor analysis showing that neither by the former, nor by the latter a clear "prima vista" prediction or association of one single or several cumulative influencing lung function parameter could be found, and after having formal proof that also by a cluster analysis no natural grouping in the data could be found, a second approach was chosen by searching for potential differences in BR within clinically and/or physiologically predefined subgroups. Based on individual values of MEF₅₀ (estimate of predominant small airway obstruction), Raw (estimate of predominant bronchial obstruction) and TVG (estimate of pulmonary hyperinflation) the patients are assigned to 4 functional groups (Table 1). Taking the MEF₅₀ as first, Raw as second and TVG as third selection criterium the patients were assigned to the 3 classes of bronchial reactivity. There was a significant association between the classes of BR (1: hyperreactive; 2: intermediate; 3: normo-reactive) and pre-existing alteration of lung function (chi-square 27.1; $p < 0.001$). Asthmatic children with baseline values of a MEF₅₀ < 80% predicted ($n = 95$; 26.8% of all asthmatics) are allocated to 60% in the hyperreactive Class 1, in addition to 44.4% with increased Raw. In contrast 41.2% of patients with increased TGV (pulmonary hyperinflation) or 44.1% with normal baseline lung function are allocated in the normo-reactive Class 3. There was neither a distinction of BR between Class 1 and Class 2 (chi-square 2.05; $p = 0.15$), nor sex difference.

Table 2. Interference of concomitant factors as family history for atopy (FH), parental smoking and out-door pollution on distribution into 3 classes of carbachol induced bronchial reactivity in 149 healthy controls

Bronchial reactivity PD65 (μ g carbachol)	n	High <480	Medium 481–960	Low >961	Chi-square statistics
Family history of atopy					
positive	53	1	16	36	2.06
negative	96	2	19	75	n.s.
Parental smoking					
yes	67	2	14	51	0.96
no	82	1	21	60	n.s.
Air pollution					
high NO ₂ (36.2 $\pm$ 9.5 μ g/qm)*	94	2	26	66	
low NO ₂ (26.2 $\pm$ 10.4 μ g/qm)*	55	1	9	45	2.52 n.s.
All healthy children	149	3	35	111	

*Significant different air NO₂-content ($p < 0.001$)

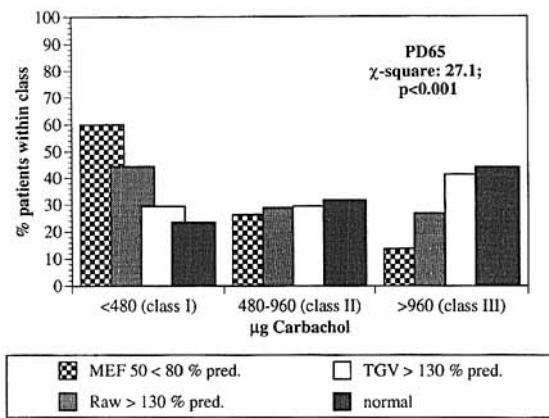


Fig. 3. Percent distribution of asthmatic children within the 3 classes of bronchial reactivity according initial functional abnormalities. (PD₆₅: provocation dose inducing an increase of the resistance of the respiratory system (R'_{m}) from 100 to 165% of baseline value; MEF₅₀: maximal expiratory flow at 50% expired vital capacity; Raw: airway resistance; TGV: thoracic gas volume).

Predisposing factors in healthy children

A positive family history of atopy (parents or brothers and sisters) was found in 53 (35.6%), exposure to parental smoking in 67 (45.0%) and/or exposure to increased air pollution in 94 (63.1%) of the healthy controls. The concomitant factors potentially influencing BR in healthy children are given in Table 2. Principle component and cluster analysis revealed no singular or combined effect of

age, sex, parental smoking, or family history of atopy on BR. Although a certain influence of air pollution to BR could be found between healthy children living in a area of high or low outdoor-NO₂, its influence was statistically insignificant ($p = 0.11$).

Sensitivity and specificity

Fig. 4 shows s and f of PD₆₅ to carbachol with reference to the different concomitant factors. In contrast to the findings given by the component and cluster analysis regarding a potential influence of age, sex, positive family history, parental smoking and air pollution on allocation in the 3 classes of BR, s and f of PD₆₅ are clearly dependent on sex (middle panel). The weighed s and f for healthy subjects are higher and at a lower level in boys (78% at 960 µg carbachol) than girls (66% at 1020 µg carbachol). In addition to sex, specificity is also influenced by the presence or absence of a family history for atopy and the condition of air pollution. Cumulating both as shown in the left-hand panel of Fig. 4, one can observe that the subgroup of healthy children having no history for atopy and living in a area with low air pollution in particular showed the best cumulative percent response. The finding whether the child was exposed to parental smoking or not showed no influence on f . Regarding sensitivity in asthmatic children and

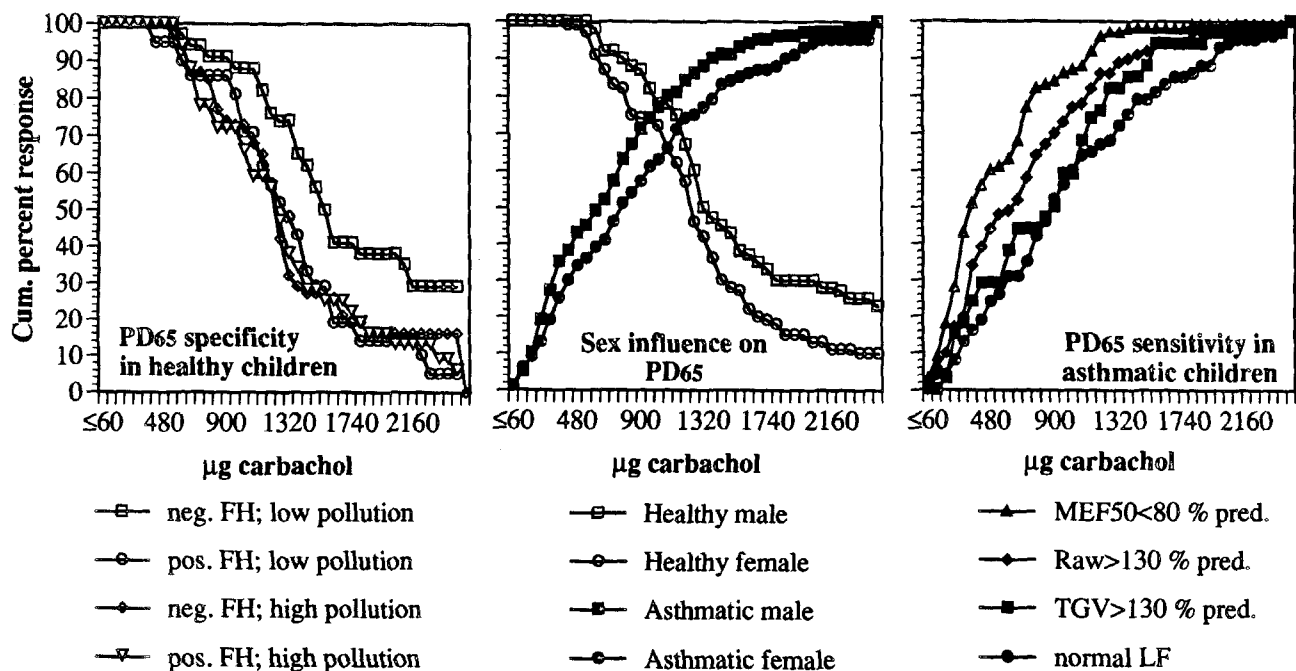


Fig. 4. Sensitivity and Specificity of PD₆₅ to carbachol in 355 asthmatic children and 149 healthy controls in relation to the interference of concomitant factors like sex, family history for atopy (FH), out-door pollution and predominantly abnormal lung function parameters. (PD₆₅: provocation dose inducing an increase of the resistance of the respiratory system (R'_{m}) from 100 to 165% of baseline value; MEF₅₀: maximal expiratory flow at 50% expired vital capacity; Raw: airway resistance; TGV: thoracic gas volume).

Table 3. Weighed sensitivity (*s*) and specificity (*f*) and corresponding thresholds of PD₆₅ to carbachol in relation to interference of concomitant factors in healthy subjects (1st variable) as sex, family history for atopy (FH) and air pollution on the one hand and interference of concomitant factors in asthmatic children (2nd variable) as maximal expiratory flow at 50% vital capacity (MEF₅₀), airway resistance (Raw) and thoracic gas volume (TGV) on the other hand

Healthy subjects 1st variable	Asthmatic children 2nd variable	Weighed <i>s</i> and <i>f</i> for PD ₆₅ in %	Threshold μg carbachol
Boy	Boy	78	960
Girl	Girl	66	1020
Neg. FH;	Boy	82	1080
Low pollution	Girl	75	1140
Pos. FH;	Boy	72	840
High pollution	Girl	65	960
Boy	MEF < 80% pred.	84	900
	Raw < 80% pred.	78	1020
	TGV > 130% pred.	74	1080
Girl	MEF < 80% pred.	82	720
	Raw < 80% pred.	74	900
	TGV > 130% pred.	67	1020
Neg. FH;	MEF < 80% pred.	88	1020
Low pollution	Raw < 80% pred.	82	1080
	TGV > 130% pred.	76	1140
Pos. FH;	MEF < 80% pred.	78	660
High pollution	Raw < 80% pred.	73	900
	TGV > 130% pred.	64	960

(*s*: sensitivity; *f*: specificity; FH: family history for atopy; high air pollution NO₂: 36.2 ± 9.5 μg/qm*; low air pollution NO₂: 26.2 = 10.4 μg/qm*, * significantly different air NO₂-content (p < 0.001))

in addition to dependency from sex it can be demonstrated (righthand panel of Fig. 4) that best *s* is obtained in the group having a MEF₅₀ < 80% and hence characterised by a small airway disease. In contrast, *s* appears negatively be influenced by a state of pulmonary hyperinflation (TGV > 130% pred.) or normal baseline lung function.

Based on the computation of *s* and *f* curves shown in Fig. 4 weighed *s* and *f* as well as the corresponding thresholds of PD₆₅ to carbachol in relation to interference of concomitant factors in healthy subjects (1st variable) as sex, family history for atopy (FH) and air pollution on the one hand and interference of concomitant factors in asthmatic children (2nd variable) as maximal expiratory flow at 50% vital capacity (MEF₅₀), airway resistance (Raw) and thoracic gas volume (TGV) on the other hand are given in Table 3. There is a wide range of weighed *s* and *f* depending on the question which characteristic subgroups of healthy and asthmatic children are compared. Best weighed *s* and *f* (88%) is found for healthy children with a negative family history of atopy and living in a low pollution area with asthmatic children presented with a predominant decrease in MEF₅₀. Worst weighed *s* and *f* (64%) is given for healthy children with positive family history of atopy,

living in a highly polluted area with asthmatic children with pulmonary hyperinflation.

Discussion

The presence of nonspecific bronchial hyperreactivity is widely accepted as an important component of the definition of childhood asthma. Current asthmatic children will have substantial deteriorated lung function to inhaled histamine, methacholine (27, 42), carbachol, hyperventilation of cold dry air and physical effort. In adults, the degree of bronchial response has been shown to be associated with the severity of the asthma (12, 27), and is inversely related to the level of pulmonary function (43, 44). More recently, it has been shown that bronchial reactivity to methacholine in children is independent of age in the pediatric asthmatic population but distinguishes between severity groups (45). The present study confirms these findings. Although it has been shown that BHR occurring with respiratory symptoms may serve as an important indicator for early-onset of atopy (13), the bronchoconstrictor responses to natural and physical challenges are more specific for asthma but less sensitive in detecting non-specific reactivity in non-asthmatic subjects (46, 47). Children seem to have more responsive airways than adults (22, 48).

The original aspect of this study is the complex analysis of potential confounding factors in both the asthmatic group and the healthy children, to determine their influence on bronchial responsiveness. The data confirm the given continuum of nonspecific BR in asthmatic and non-asthmatic, non-atopic and non-infectious subjects with an inevitable overlap of responses in the intermediate dose range (480–960 μg) of carbachol as the challenging agent. The reason for that cannot solely be a definition of diagnosis, since childhood asthma was strictly evaluated in our patients according international standards and in the collective of healthy subjects every well know factor on BR has been excluded. It seems to be, rather, a question of several concomitant factors from which some have been looked at. Other speculative mechanisms such as the variability in the shifting of TGV during provocation test, the variability in airway alterations, and the variable time-dose response in different subjects could be responsible. In an analogy to a recent study published by Avital et al. (45), our patients were stratified according to the initial functional abnormalities. Other suggested that stratification should be based on therapeutic requirements for optimal control of symptoms. Since the individualized therapeutic interventions are based on the aim to have the child free of

symptoms within its individual best range of lung function similar results can be expected from both stratification procedures. The only difference in these approaches is that under the condition of optimal cooping and optimal drug compliance the latter classification takes into account the course of the disease, whereas the former stratification, based on lung function data, features a more objective but instantaneous estimation of the disease state.

There are certain lung physiological prerequisites which definitively influence response to the provocative agent, and the present study shows that a decrease in MEF_{50} , which can easily be measured (even in the ambulatory praxis), is related to bronchial reactivity. There is an inverse relation between the prechallenge level of the MEF_{50} and the determinant of carbachol airway responsiveness in these asthmatic children as it was previously shown by Woolcock and colleagues (43) for the prechallenge FEV_1 and the FEV_1/FVC values and histamine threshold in a random population of adults. Furthermore, the results are in line with findings of Sparrow et al. (44), showing a significant relationship between airway responsiveness to methacholine and the prechallenge FEV_1 examined among 914 male adults. Since the MEF_{50} was independently measured from the R_{int} it is probably correct that preexisting alterations in airway geometry (49, 50) must be respected as confounding factors for BR measurements.

Pulmonary hyperinflation, present in almost 16% of asthmatic children as previously found and less responsive to sympathomimetics (4), seems to be less related to BHR in asthmatic children. Although we have carefully looked at different influencing factors like the kind of allergy (pollen, animals or house dust mites), the degree of allergen exposure, the IgE levels, the combination of drugs prescribed (especially topical corticosteroids, suggested to reduced nonspecific bronchial reactivity (51–53) and the degree of drug compliance, we cannot explain definitively why asthmatic children with pulmonary hyperinflation showed less sensitivity to BR. One explanation could be that measurements of non-specific BHR predominantly reflect the subject's immediate type reaction of allergy. Therefore, it could be speculated that patients with pulmonary hyperinflation predominantly represent asthmatics where late phase reactions or a combination of both are operational.

There are very few studies which have investigated a random population of healthy children (48, 54) and in most settings the identification of cases with asthma has not been the primary goal of bronchial provocation testing. In addition, the use of bronchial provocation testing to detect adverse ef-

fects of environmental exposures and to identify persons susceptible to such exposures has been studied only in adults. Three of 149 (2.1%) of the healthy children were allocated in Class 1 (hyper-reactive) and 35 (23.4%) were from the intermediate type of BR. There are a number of possible explanations for the occurrence of asymptomatic BHR in healthy children. Firstly, BHR may be transitory after an upper respiratory tract infection, although, in this study such cases were excluded from the study. Other explanations include ignorance or denial by the parents of the child's actual symptoms and poor perception of attacks of airway narrowing by the child. In the latter the degree of the BHR might be low despite the necessity of a large provoking stimulus prior to the occurrence of symptoms. It has been reported that children with asymptomatic BHR have "transitory abnormalities" (55). The prognostic significance of BHR in the absence of symptoms is unknown. It is possible, however, that such BHR in some children is a marker for the development of asthma in later years.

In conclusion, BR is only one functional aspect contributing to the complex mechanism of asthma. The present study shows that adequate response in a bronchial challenge test is to be expected when baseline lung function (MEF_{50} , Raw or TVG) is already abnormal. The study furthermore shows that asthmatic children with mixed functional disorders as pulmonary hyperinflation and central bronchial obstruction may have impaired response to the bronchial challenge test. We therefore conclude in consequence that it is of great importance to know precisely the exact type and degree of lung function abnormality for each individual before making a bronchial challenge test and evaluating BHR in asthmatics accordingly. Taking into account the factors confounding BR in children, sensitivity and specificity can be brought up to a more intense level of acceptance.

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