Airway hyperresponsiveness and development of lung function in adolescence and adulthood

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Received 17 December 2012; accepted 15 January 2014
Available online 24 January 2014

KEYWORDS
Adolescent; Adult; Airway hyperresponsiveness; Lung function; Epidemiology; General population

Summary
Background: Long-term longitudinal studies of lung function from childhood to adulthood are important in linking our understanding of childhood risk factors to adult disease. Airway hyperresponsiveness has been shown to independently affect lung function growth in studies of adolescence. The objective of the study was to test the hypothesis that airway hyperresponsiveness has an independent deleterious effect on lung function in adolescence that extends into adulthood.

Methods: A random population sample (n = 983) aged 7–17 from Copenhagen was followed longitudinally for 20 years with four examinations.

Results: A total of 780 (79.3%) subjects contributed with lung function measurements and bronchial provocation testing. Among these, 170 (21.8%) had airway hyperresponsiveness at one examination or more during the study period. There was no difference in initial FEV1 levels between subjects with and without airway hyperresponsiveness. In a repeated measures regression model with adjustment for asthma and smoking, airway hyperresponsiveness was independently associated with reduced rates of growth in lung function in both sexes of 23 ml/year. Reduced growth rates resulted in deficits in maximal attained level of lung function at age 18, which persisted throughout the follow-up until the last examination at age 27–37 years.
**Introduction**

Development of lung function during childhood and adolescence determines the maximal attained lung function in young adulthood, and this level together with the rate of decline in lung function determines the subsequent lung function levels during adult life [1,2]. As such, deficits in maximal attained lung function may potentially contribute to the risk of later development of chronic obstructive pulmonary disease (COPD). Factors that influence lung function growth in children and adolescents are therefore interesting to study, as well as factors with effects on lung function that extend beyond the growth phase.

Only one general population study has studied lung function development from birth to the age of 22 [3]. In this study, the authors found, that children with low lung function in infancy had significantly lower lung function at the age of 22. Furthermore, they investigated the influence of wheeze, smoking, atopy, and parental asthma on this association, and found no modifying effects of these parameters on the development of lung function measures.

Two general population studies have addressed the independent effect of AHR on lung function in adolescents, and found that AHR is related to short-term deficits in lung function growth independently of asthmatic symptoms [4,5]. One study found AHR to be the single most important risk factor for reduced maximal level of lung function [6]. In adult populations, AHR has been related to lung function decline in individuals both with and without asthma [7]. We therefore hypothesized that AHR may be independently associated with lung function growth during adolescence in the general population, and that this association possibly have long-term effects extending into adulthood.

Consequently, the aim of this study was to investigate the association between AHR and lung function development from childhood to adulthood. Analyses were performed on a non-selected cohort of subjects followed over 20 years from childhood to adulthood covering the ages from 7 to 37 years.

**Methods**

**Measurements of lung function**

Measurements of forced expiratory volume in 1 s (FEV$_1$) and forced vital capacity (FVC) were performed on a 7-L dry wedge spirometer (Vitalograph®, Buckingham, UK) at all examinations. Each measurement consisted of at least three forced expiratory manoeuvres from total lung capacity to residual volume with a variation of less than 5%. Manoeuvres were performed with the subjects in standing position without use of nose-clip. The highest FEV$_1$ and FVC values were the reported in absolute values. For the first and second examination, when subjects were still children and adolescents, the reference values by Zapletal were used to calculate FEV$_1$/pred and FVC in percent of predicted value (FVC%pred) [9]. For the examinations three and four, when subjects had become adults, the reference values for pulmonary function testing published by the European Community for Coal and Steel (ECCS) [10], was used to calculate FEV$_1$/pred and FVC%pred.

Before testing, subjects were instructed not to use theophylline for at least 24 h, and short-acting beta-agonist (SABA) for 6 h. When long-acting beta-agonists (LABA) and leukotriene antagonists became available in Denmark, subjects were asked not to use LABA for 12 h, and leukotriene antagonists for 24 h. They were allowed to continue use of ICS. Subjects had no respiratory infections within 6 weeks of testing. Otherwise the examination was postponed.

**Bronchial challenge tests**

Bronchial challenge tests were performed with histamine according to the method described by Cockcroft et al. [11] at the first three examinations, and with methacholine according to the method of Yan et al. [12] at the fourth examination. Cut-off was a fall in FEV$_1$ of 20% or more at a concentration of less than 8 mg/ml at the first examination, due to constraints placed by the ethical committee at the time, 16 mg/ml at the second and third examination, and a dose less than 8 μmol at the fourth examination.

**Height and weight**

Height (in standing position without shoes) and weight were measured, and body mass index (BMI) was calculated.

**Case history**

Case history was obtained in part by questionnaires and in part by semi-structured interviews performed by a trained physician at all examinations. Case history included data on asthma, allergic diseases, and lifestyle factors.

Questions about asthma were adopted from studies by the ATS, Division of Lung Disease of the National Heart, Lung and Blood Institute [13] and Global Initiative for Asthma (GINA) [14].
Smoking history was recorded and subjects were classified as current smokers, former smokers or never smokers. In the analyses, former and current smokers were grouped as ever smokers.

Statistical methods

Data were analysed with the statistical software package SAS (SAS Institute Inc., Cary, NC). Repeated measures regression analysis was used to study the pattern of lung function development. Our model fitted two lines with different slopes joining in a breakpoint at 18 years of age to describe the growth phase followed by a plateau or decline phase. The correlation between the unequally spaced measurements was modelled by a power function of the time between the measurements using Proc Mixed. Dependent variable was FEV_{1} and the explanatory variables were AHR, age, sex, asthma, and smoking. An interaction term between sex and age was added to allow sex differences in lung function growth. The variable AHR was tested in two versions: one that described a positive AHR test on at least one occasion during the study (ever AHR), and one that described AHR at the first examination of the subject. The term "estimated values of FEV_{1}," refers to the fitted values calculated from the model.

The possible confounding of FEV_{1} by height was addressed by fitting a model with height as dependent variable and the same covariates as the model for FEV_{1}. This showed that the pattern of height growth was not influenced by AHR or any other covariates besides age and sex (p > 0.2). Therefore, height is not likely to have confounded the relationship between FEV_{1} and the covariates.

Differences in means were tested using parametric (t-test) and non-parametric (Mann–Whitney) tests where appropriate. Differences in frequencies were tested using Chi-square tests.

P-values below 0.05 were considered significant. All subjects gave informed consent at each examination and the study was approved by the ethics committee of Copenhagen and Frederiksberg ((KF) 01 318737).

Table 1 Characteristics of subjects included in the longitudinal analyses.

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<tbody>
<tr>
<td></td>
<td>Males</td>
<td>Females</td>
<td>Males</td>
<td>Females</td>
</tr>
<tr>
<td>Age (years)</td>
<td>12.2 (2.8)</td>
<td>12.4 (2.9)</td>
<td>18.5 (2.8)</td>
<td>18.7 (2.9)</td>
</tr>
<tr>
<td>BMI (kg/m^2)</td>
<td>18.6 (2.7)</td>
<td>19.1 (3.0)</td>
<td>21.6 (3.3)</td>
<td>21.4 (4.1)</td>
</tr>
<tr>
<td>FEV_{1} (L)</td>
<td>2.61 (1.0)</td>
<td>2.45 (0.7)</td>
<td>4.47 (0.85)</td>
<td>3.46 (0.49)</td>
</tr>
<tr>
<td>FVC (L)</td>
<td>2.95 (1.1)</td>
<td>2.70 (0.8)</td>
<td>5.26 (1.00)</td>
<td>3.97 (0.58)</td>
</tr>
<tr>
<td>FEV_{1}/FVC ^1</td>
<td>0.88 (0.06)</td>
<td>0.91 (0.04)</td>
<td>0.85 (0.07)</td>
<td>0.87 (0.06)</td>
</tr>
<tr>
<td>AHR ^5</td>
<td>22 (5.9)</td>
<td>12 (2.9)</td>
<td>54 (14.5)</td>
<td>49 (12.0)</td>
</tr>
</tbody>
</table>

Values are given as mean (SD).

^1 p < 0.05 in 1986, 1992, 1998, and 2006; ^2 p < 0.001 in 1992, 1998, and 2006; ^3 p = 0.01 in 1986 and p < 0.001 in 1992, 1998, and 2006; ^4 p < 0.001 in 1986, 1992, and 1998 and p = 0.002 in 2006; ^5 p = 0.02 in 2006.

Definition of abbreviations: AHR = airway hyperresponsiveness, BMI = body mass index, FEV_{1} = forced expiratory volume in 1 s, FVC = forced vital capacity.

^a Differences between sexes were tested with t-tests for continuous variables and with Chi-square for dichotomous variables. Differences were non-significant, unless otherwise stated.

Results

For the longitudinal analysis of lung function development, 780 subjects (79.3%) were eligible and included. Of these, 148 subjects (19.0%) participated in all examinations, and thus contributed with four lung function measurements. 244 subjects (31.3%), 246 subjects (31.5%), and 142 subjects (18.2%) contributed with 3, 2, and 1 lung function measurement, respectively.

The 203 subjects that were excluded in the study have never been examined, thus we have no further information on them besides sex and age. There was no significant difference in the proportion of males (56%) and females (44%) (p = 0.07) among the excluded subjects, although there was a tendency of more males than females not participating. With regard to age, there was no significant difference in the age-distribution between included and excluded subjects (p = 0.57).

Among the included subjects, there were equal proportions of males (47.8%) and females (52.2%) (p = 0.22). Table 1 presents the characteristics of the subjects; the proportion of subjects with airway obstruction (defined as FEV_{1}/FVC < 0.70) was 0.4%, 0.8%, 2.3%, and 2.7%, respectively, at the four examinations. Table 2 presents the prevalence of AHR, asthma and smoking. There was no significant difference in prevalence of asthma between sexes (p = 0.06), although there was a slight tendency towards more females having asthma than males. Furthermore, females smoked more frequently than males (p < 0.001).

Among the 780 subjects, 170 (21.8%) had a positive AHR test on at least one occasion during the study (ever AHR). There was no difference in this prevalence between sexes (p = 0.9). Among the 170 subjects, 75 (44.1%) were AHR positive at the first examination of the subject, and significantly more males than females. Among the 95 subjects that had ever AHR during the study but no positive test at their first examination, 34 (35.8%) had positive AHR tests at all their examinations after the first examination, and 61 (64.2%) had positive AHR tests at some of their
subsequent examinations. Subjects with a positive AHR test at the first examination were older at the first examination (mean age [SD] 15.5 [5.4]) than subjects with ever AHR but a negative test at the first examination (mean age [SD] 13.5 [3.9]), \( p = 0.01 \).

### AHR and lung function development

We used a repeated measures regression model to examine the effect of AHR on lung function development adjusted for asthma and smoking. The model demonstrated a difference in growth rates in FEV\(_1\) until age 18 between males and females of 154 ml/year (\( p = 0.015 \)) with males presenting with higher growth rates compared to females, as expected (374 ml/year vs. 219 ml/year).

There was no difference in FEV\(_1\) values at age 7 between subjects with and without AHR, but subjects with ever AHR had a significant reduction in growth rate of FEV\(_1\), of 23 ml/year until age 18 (\( p = 0.015 \)).

Estimates of the effect of asthma (\( p = 0.7 \)) and smoking (\( p = 0.6 \)) on growth rate were insignificant in the model with AHR.

After age 18, both males and females presented with slow annual declines in FEV\(_1\) of 18 ml (\( p < 0.001 \)). No difference could be demonstrated between sexes. AHR had no effect on this rate of decline (\( p = 0.6 \)), thus the slopes for the subjects with and without AHR were parallel after age 18. Although there was a tendency towards smoking accelerating the decline in FEV\(_1\) after the age of 18, it did not reach statistical significance (\( p = 0.1 \)).

### Discussion

In this longitudinal study of lung function development from childhood to adulthood in a general population, we found that AHR is independently associated with reduced rates of lung function growth and causes deficits in maximal lung function attained. Our results were suggestive of AHR being an independent predictor for the subsequent lung function development, which indicates a temporal relation between AHR and lung function growth in children and adolescents with AHR preceding the lung function changes. A temporal relationship between presence of AHR and subsequently reduced FEV\(_1\) level two years later has been demonstrated in adolescents [5], but we were now able to demonstrate the long-term consequences of this relationship. The adverse effect of AHR on lung function growth resulted in deficits in maximal attained level of lung function at age 18, as seen in a previous study [6], which persisted through the rest of our follow-up period. The oldest subjects in our cohort were followed until age 37, and we were not able to demonstrate any effect of AHR on lung function decline in adulthood so far. An association between AHR and lung function decline in adults has been found previously in older general population cohorts [7]. It is most likely, that our follow-up period is still too short to demonstrate this association.

Several general population studies have investigated lung function development from infancy through childhood or from childhood through adolescence, but so far the follow-up periods have not covered ages beyond early adulthood [5,15–17]. Consequently, these studies cannot demonstrate the long-term consequences of lung function development in childhood and adolescence on later lung function. Other studies are initiated at an older age [18,19], and thus cannot demonstrate factors of importance for lung function development during the growth phase. Only few studies on lung function development are based on longitudinal measurements from childhood to older ages [3,20].
Our repeated measures regression model revealed a gender difference in the FEV₁ growth rate until age 18, whereafter males and females presented with similar slow annual declines in FEV₁. In contrast to our findings, Wang et al. [6] have previously reported from a large cohort study based on the Vlagtwedde/Vlaardingen study in the Netherlands that the level of FEV₁ has already peaked by age 15 in females, whereas the peak in males was not observed until age 20. However, our findings are in accordance with previous observations reported by Tager et al. [1] from the East Boston Study. The reasons for these discrepancies in observations are unclear, but may be related to differences in sampling of the cohorts, including amount of data in the age range of greatest interest and age range, and differences in exposures, as also reflected in comparisons of reference values for lung function in young adults [21].

Even though our study was consistent with previous studies, there are some limitations to note. Most notably is the change in use of bronchoprovocational agent during the study from histamine to methacholine. Histamine and methacholine are equivalent in producing bronchoconstriction at nearly the same concentrations and doses, and at the same weight as their molecular weights are relatively close. Accordingly, tests with histamine and methacholine are comparable, and they are most likely related to the same pathological airway processes and probably interchangeable [22,23].

Our findings of AHR from the first examination in 1986 were possibly influenced by the restricted permission on the AHR test in terms of the low-concentration cut-off (20% fall in FEV₁ caused by less than 8 mg/ml histamine). The cut-off employed for the subsequent examinations, i.e. a 20% fall in FEV₁ caused by less than 16 mg/ml of histamine had most likely detected more AHR positive subjects and could have strengthened the results on AHR as a predictor of impaired lung function growth.

Moreover, there was a trend, but no significant effect of smoking on lung function decline could be demonstrated. Again, it is possible that the cohort is still too young for this effect to be demonstrated. Besides AHR, asthma, and active smoking, other factors such as passive tobacco exposure and respiratory infections in infancy have been shown to have an effect on lung function development, including some we have not been able to assess in this cohort. We had no measurements of indoor or outdoor pollution. However, as all subjects in the cohort resided in the capital of Denmark, differences in levels of at least outdoor pollution between subjects were most likely minor.

In conclusion, we found that AHR had an independent deleterious effect on lung function development over the ages from 7 to 37 years resulting in a lower maximal attained lung function and persistently lower levels of lung function after this peak. The effect persisted after adjustment for asthma and smoking.

Conflict of interest statement

All authors declare they have no conflicts of interests to disclose.

Acknowledgements

The authors thank all the participants in the study. The study has received funding from Copenhagen University Hospital, Bispebjerg; The H:S Research Foundation; King Christian X Foundation. The study sponsors have not been involved in the study design; the collection, analysis and interpretation of data; the writing of the manuscript; nor in the decision to submit the manuscript for publication.

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