# Risk factors for bronchial hyperresponsiveness in teenagers differ with sex and atopic status

**Rachel A. Collins, PhD,[a] Faith Parsons, MSc,[a,b] Marie Deverell, PhD,[a] Elysia M. Hollams, PhD,[b] Patrick G. Holt, DSc,[b] and Peter D. Sly, DSc[a,c]** *Perth and Brisbane, Australia*

**Background: Sex-related differences in bronchial hyperresponsiveness (BHR) have been reported in adolescents, but the mechanisms remain obscure.**
**Objective: To investigate the risk factors for BHR in the Raine Study, a community-based longitudinal birth cohort.**
**Methods: At 14 years of age, children underwent a respiratory assessment including a questionnaire, lung function testing, methacholine challenge, and determination of atopic status.**
**Results: A total of 1779 children provided data for assessment, with 1510 completing lung function and methacholine challenge testing. Current asthma was present in 152 (10.4%), 762 (50.5%) were atopic, and 277 (18.6%) had BHR. BHR was more common in girls, whereas atopy was more common in boys, with no sex differences in asthma or current wheeze. Independent risk factors for BHR were being female (odds ratio [OR], 3.45; $P < .001$), atopy at 14 years (OR, 1.27; $P = .004$), and current asthma (OR, 2.15; $P = .005$). Better lung function was protective against BHR (forced expiratory flow between 25% and 75% of forced vital capacity/forced vital capacity, OR, 0.09; $P < .001$). Risk factors differed with sex and atopic status. Early-life factors were generally not independent risk factors for BHR at 14 years of age, with the exception of being smaller at birth in boys (birth length, OR, $6 \times 10^{-9}$; $P = .017$) and maternal asthma in girls (OR, 1.84; $P = .041$). Current asthma was not a risk for BHR in nonatopic children.**
**Conclusion: Bronchial hyperresponsiveness was more common and more severe in girls. These differences could not be explained by differences in lung function or atopic status.**
**(J Allergy Clin Immunol 2011;128:301-7.)**

***Key words:*** *Asthma, atopy, longitudinal birth cohort, lung function, sex differences*

*Abbreviations used*
BHR: Bronchial hyperresponsiveness
$FEF_{25-75}$: Forced expiratory flow between 25% and 75% of forced vital capacity
FVC: Forced vital capacity
HDM: House dust mite
OR: Odds ratio

Bronchial hyperresponsiveness (BHR) is considered to be a cardinal feature of asthma, although it is only loosely associated with symptomatology and the clinical course of the disease. Rather than being a direct marker of asthma, BHR may in fact be a parallel pathologic process[1,2] with distinct genetic inheritance.[3] Indeed, BHR during childhood has been shown to be related to deficits in the growth of airway function, independent of current asthma symptoms.[4] Longitudinal epidemiologic studies may assist in the identification of the factors determining BHR and help us to understand its role in respiratory health and disease. We have recently published a cross-sectional analysis of the risk factors for asthma and BHR in 14-year-old children participating in a longitudinal community-based birth cohort study.[5] In that study, we identified the close association between current atopy, current asthma, and BHR at age 14 years. More severe atopy was associated with higher risks of current asthma and BHR and also with more severe asthma. However, in those analyses, early-life factors were not considered.

The early years of life are thought to present an important developmental window in which future respiratory health is determined.[6] Early insults to the respiratory system may result in structural remodeling or deficits in pulmonary function that persist into later life. Biopsies of airway epithelium in wheezy infants (1 year old) and wheezy preschoolers (3 years old) have shown that structural changes can occur during this time; specifically, epithelial reticular basement membrane thickening and eosinophilic inflammation have been reported.[7] Similarly, eosinophilic inflammation and remodeling can both be present even in early school-age children, and some features of remodeling are maximal even at this young age.[7] Other studies suggest that epithelial reticular basement membrane thickening, which occurs early in the asthma process and correlates with BHR, is maximal between 6 and 16 years of age.[8]

Asthma is more common in boys in early life but more common and more severe in women. The change in sex balance occurs during adolescence and seems to be related to pubertal development. Tantisira et al[9] have demonstrated that sex-specific differences in airway responsiveness exist in relation to age and stage of puberty, with a divergence in responsiveness occurring between boys and girls at approximately 11 years of age. This finding emphasizes the need for longitudinal studies spanning puberty in the examination of BHR and associated factors.

From [a]the Division of Clinical Sciences and [b]the Division of Cell Biology, Telethon Institute for Child Health Research, Centre for Child Health Research, University of Western Australia, Perth; and [c]the Queensland Children's Medical Research Institute, University of Queensland, Brisbane.

Supported by NHMRC, Australia #211912, #458513; and Stanley Trust UK.
Disclosure of potential conflict of interest: P. G. Holt and P. D. Sly receive research support from the National Health and Medical Research Council, the Stanley Trust, and Phadia AB. The rest of the authors have declared that they have no conflict of interest.
Received for publication October 28, 2010; revised March 13, 2011; accepted for publication March 17, 2011.
Available online April 17, 2011.

Reprint requests: Peter D. Sly, DSc, Queensland Children's Medical Research Institute, Level 4, Foundation Bldg, Royal Children's Hospital, Herston Rd, Herston, Qld 4029, Australia. E-mail: p.sly@uq.edu.au.

0091-6749/$36.00

doi:10.1016/j.jaci.2011.03.016

The current study was performed to examine early-life risk factors for BHR in adolescents. We addressed this aim by using the Western Australian Pregnancy Cohort (Raine Study), a longitudinal community-based birth cohort that included prospective collection of data on respiratory health and major respiratory assessments at the ages of 6 and 14 years. We hypothesized that early-life risk factors for BHR in adolescence would differ between boys and girls.

## METHODS

### Study population/participants

Subjects were from an unselected longitudinal birth cohort[10] undertaken in Perth, Australia (see the Methods section in this article's Online Repository at www.jacionline.org for cohort details). The 2337 children available at the 14-year follow-up were invited to participate in a questionnaire and clinical examination including a physical examination, pulmonary function testing, methacholine challenge, and a blood test. Total serum IgE; Phadiatop IgE (Phadia AB, Uppsala, Sweden); specific IgE and $IgG_4$ antibodies to house dust mite (HDM), rye grass, cat, couch grass, mold, peanut, and food mix; and specific IgG antibodies to HDM, cat, and grass mix were measured with ImmunoCAP (Phadia AB, Uppsala, Sweden).[5] A similar assessment had been undertaken when the children were 6 years old. Participant numbers at the 6-year and 14-year follow-ups are available in this article's Table E1, *A and B,* in the Online Repository at www.jacionline.org.

### Variable definitions

*Current asthma* was defined as all 3 of the following: (1) physician-diagnosed asthma (ever), (2) wheeze in the last 12 months, and (3) use of asthma medications in the last 12 months. *Current wheeze* was defined as wheeze in the last 12 months.

*Atopy* was defined at 14 years as specific IgE levels of >0.35 kU/L (ImmunoCAP, Phadia AB) to 1 or more aeroallergens (HDM, rye grass, couch grass, cat, mold mix). The impact of severity of atopy was determined by including specific IgE levels as continuous variables in regression analyses. At 6 years of age, atopy was defined by skin prick test, and severity scores were calculated as described previously[11] (see Methods in the Online Repository for details).

### Spirometry and bronchial responsiveness

Spirometry and methacholine challenge were performed to American Thoracic Society standards (see Methods in the Online Repository for details). In addition to baseline and percent predicted spirometric outcomes, $FEV_1$/forced vital capacity (FVC) and forced expiratory flow between 25% and 75% of forced vital capacity ($FEF_{25-75}$)/FVC were calculated as relatively size-independent indicators of airway obstruction[12] and airway caliber,[13] respectively.

All subjects capable of performing acceptable spirometry participated in methacholine challenge. All spirometry measurements were performed in the morning (10 AM to 12 PM) and conducted according to American Thoracic Society guidelines with the subjects seated.[14,15] Subjects were eligible to participant in challenge testing if they obtained reproducible spirometry, had an $FEV_1$ at least 80% of predicted, had no respiratory illness in the last 14 days, and had been withheld from their asthma medications for the standard period.[16] Methacholine challenges were performed with a Koko digidoser (Pulmonary Data Services, Louiseville, Colo) by using a modified dosimeter technique.[16] Subjects were given an initial saline dose followed by doubling doses of methacholine (0.0625, 0.125, 0.25, 0.5, 1, 2, 4, and 8 mg/mL) delivered via DeVilbiss 646 nebulizers (Devilbiss, Australia). Testing was stopped once a patient's $FEV_1$ had fallen by ≥20% or the highest dose was administered. BHR was defined as a binary outcome with subjects who had a $PD_{20}$ <8 mg/mL classified as positive.

### Statistical analysis

The $\chi^2$ test of association and the Mann-Whitney test of significant difference were performed to assess the relationship between BHR at age 14 years and categoric and continuous variables, respectively. Univariate logistic regression was performed in the whole population, in boys and girls separately, and in atopic and nonatopic subjects separately. Variables were selected from univariate results for inclusion in multivariate logistic regression according to the following selection criteria: $P \leq 0.1$, Nagelkerke $R^2 \geq 0.01$, and missing samples ≤363 for the whole population or missing samples ≤183 after sex stratification. Redundant variables were eliminated from multivariate models by using multivariate logistic regression. Variables were included in forward-step multivariate logistic regressions (see this article's Table E2, *A-D,* in the Online Repository at www.jacionline.org). Variables were selected as described before forward-step multivariate logistic regression in subgroups with and without respiratory symptoms.

## RESULTS

One thousand seven hundred ninety-nine adolescents participated in the 14-year follow-up with 1510 completing lung function and methacholine challenge testing and forming the basis of the current report. Methacholine challenge was not performed in 72 children (Table E1, *B*). The demographic characteristics of the participants are shown in Table I. Current asthma was present in 152 (10.4%; 81 boys, 71 girls), and 762 (50.5%) were atopic, with positive IgE to HDM most common (30.2%). Overall, 277 (18.6%) had a positive response to methacholine, with 120 (15.6%) boys and 157 (21.7%) girls classified as having BHR. Overall, there was a strong relationship between the presence of current symptoms (current wheeze or current asthma) and BHR ($P = .037$ and $P = .005$, respectively). However, of those with current asthma, only 41.4% had BHR, and 35.7% of those with current wheeze had BHR. Alternatively, of those with BHR at age 14 years, 23.2% had current asthma, and 25.8% had current wheeze. Body mass index ranged from 12.3 to 43.5, with 75.6% considered to be normal or underweight, 17.8% overweight, and 6.6% obese. The majority of children classified with asthma used short-acting bronchodilators on an as-needed basis, with only 42 of 152 (27.6%) taking inhaled steroids (with or without long-acting β-agonists). Personal smoking was uncommon, with only 2.5% of children reporting that they currently smoked cigarettes.

Comparing the prevalence of asthma, current wheeze, atopy, and BHR in the same children seen at 6 years and at 14 years showed that the prevalence of current asthma fell from 18.2% to 10.6%, current wheeze decreased from 23.7% to 14.5%, and the proportion with BHR decreased from 53.5% to 16.4% (see this article's Table E3 in the Online Repository at www.jacionline.org). It must be noted that only a subsample of children were offered BHR testing at 6 years of age, and data were available for only 383 children. In contrast, the proportion of children with atopy increased from 34.5% at 6 years of age to 52.8% at 14 years. Some children "lost" asthma, current wheeze, atopy, and BHR between 6 and 14 years, and some gained new diagnoses (Table E3).

### Univariate associations with BHR at 14 years

**Family and early-life risk factors for BHR at 14 years.** Adolescents with BHR were more likely to be girls ($P = .002$) and to have had low birth weight ($P = .009$) and length ($P = .021$). No association was found between body mass index at any follow-up age (1, 3, 6, 8, 10, 14 years) and BHR at 14 years (see this article's Table E4 in the Online Repository at www.jacionline.org).