

Airway Hyperresponsiveness to Methacholine in 7-Year-Old Children: Sensitivity and Specificity for Pediatric Allergist-Diagnosed Asthma

Chris Carlsten, MD MPH,^{1*} Helen Dimich-Ward, PhD,¹ Alexander Ferguson, MD,² Allan Becker, MD,³ Anne DyBuncio, BSc,¹ and Moira Chan-Yeung, MB¹

Summary. Background: The operating characteristics of PC₂₀ values used as cut-offs to define airway hyperresponsiveness, as it informs the diagnosis of asthma in children, are poorly understood. We examine data from a unique cohort to inform this concern. Objective: Determine the sensitivity and specificity of incremental PC₂₀ cut-offs for allergist-diagnosed asthma. Methods: Airway reactivity at age 7 was assessed in children within a birth cohort at high risk for asthma; PC₂₀ for methacholine was determined by standard technique including interpolation. The diagnosis of asthma was considered by the pediatric allergist without knowledge of the methacholine challenge results. Sensitivity and specificity were calculated using a cross-tabulation of asthma diagnosis with incremental PC₂₀ cut-off values, from 1.0 to 8.0 mg/ml, and plotted as receiver operator characteristic (ROC) curves. The "optimal" cut-off was defined as that PC₂₀ conferring maximal value for sensitivity plus specificity while the "balanced" cut-off was defined as that PC₂₀ at which sensitivity and specificity were most equal. Results: 70/348 children (20.1%) were diagnosed with asthma. The optimal and balanced PC₂₀ cut-offs, both for all children and for females alone, were respectively 3 mg/ml (sensitivity 80.0%, specificity 49.1%) and 2 mg/ml (sensitivity 63.1%, specificity 64.7%). For males alone, the "optimal" and "balanced" PC₂₀ cut-offs were both 2 mg/ml. Conclusion: For this cohort of 7-year olds at high risk for asthma, methacholine challenge testing using a cut-off value of PC₂₀ 3 mg/ml conferred the maximal sum of specificity plus sensitivity. For contexts in which higher sensitivity or specificity is desired, other cut-offs may be preferred.

Pediatr Pulmonol. © 2010 Wiley-Liss, Inc.

Key words: asthma; pediatric; methacholine; PC₂₀; sensitivity; specificity.

Funding source: none reported

INTRODUCTION

Objective measurement of lung function and/or airway hyperresponsiveness (AHR) in the diagnosis of asthma is encouraged by asthma guidelines.^{1,2} While a provocative concentration of methacholine resulting in a 20% drop in FEV₁ (PC₂₀) of <8 mg/ml has often been considered a reasonable threshold for supporting the diagnosis of asthma (absent compelling information to inform pre-test probability) in both adults and children,³ the diagnosis of asthma based on objective testing of children presents particular challenges.⁴ As is true with adults, there remains is the absence of a consensus "gold standard" for the diagnosis of pediatric asthma, particularly in younger children.

We utilized the opportunity provided by a high-risk birth cohort, which took part in a randomized controlled clinical trial for the primary prevention of asthma and returned for assessment of asthma at the age of 7 years, to study the relationship between PC₂₀ and the diagnosis of asthma; specifically, we describe how these data can be used to determine various cut-off points for AHR, in terms

of sensitivity and specificity for a pediatric allergist-based diagnosis of asthma, depending on the clinical or research scenario.

¹Department of Medicine, Chan-Yeung Centre for Occupational and Environmental Respiratory Disease, University of British Columbia, Vancouver, BC.

²Division of Allergy, Department of Pediatrics, University of British Columbia, Vancouver, BC.

³Section of Allergy and Clinical Immunology, Department of Pediatrics and Child Health, Manitoba Institute of Child Health, University of Manitoba, Winnipeg, Manitoba, Canada.

*Correspondence to: Chris Carlsten, MD MPH, UBC Department of Medicine, Vancouver General Hospital, 2775 Laurel Street, 7th Floor, Vancouver BC, Canada V5Z 1M9. E-mail: chris.carlsten@vch.ca

Received 8 June 2010; Revised 29 July 2010; Accepted 2 August 2010.

DOI 10.1002/ppul.21347

Published online in Wiley Online Library (wileyonlinelibrary.com).

METHODS

This cohort has been described in a previous communication.⁵ The children, at high risk for developing asthma (defined as those with at least one first-degree relative with asthma or two first-degree relatives with other IgE-mediated allergic diseases), were identified during their mothers' third trimester of pregnancy. Families were randomly allocated to the multi-faceted intervention group (N = 279) or to the control group (N = 266) prior to the child's birth. The multifaceted intervention program, focused on dietary modification, allergen remediation, and minimization of tobacco smoke exposure,⁵ was applied before birth and during the first year of life. Three hundred and eighty (70.0%) children returned at age 7 for assessment in each city by a single pediatric allergist and for laboratory testing. The diagnosis of asthma was a clinical decision made by the pediatric allergist, without knowledge of the results of methacholine test, based on symptoms of wheeze and cough, use of medications, and physical findings. The presence of recurrent wheeze was defined by questionnaire (Has your child had two or more episodes of wheezing in the past year?) administered to the children's parents at 7 years of age. Allergy skin tests were performed using the epicutaneous method with a prick lanceter (Bayer Inc., Toronto, Canada) with the following allergens (Omega Laboratories, Montreal Vancouver, Canada): house dust mite (*D. pteronyssinus* and *D. farinae*), cat, dog, cockroach, *Alternaria alternata*, *Cladosporium herbarum*, mixed tree, mixed grass and ragweed pollens, cow's milk, egg white, wheat, soy, and peanut. Atopy was defined as positive skin test reactivity (mean wheal diameter 3 mm or greater than the negative control) to one or more of these allergens.

Spirometry was carried out, after physician assessment, according to ATS standards. Methacholine challenge testing and PC₂₀ determination was based on the method of Cockcroft et al.⁶ using a hand-held Bennett Twin-jet nebulizer, with the following modification (motivated by our desire to limit the number of doses in these young children): the starting dose varied according to pre-set criteria, that is, 1.0 mg/ml if child had no asthma; 0.25 mg/ml if mild or moderate asthma; 0.06 mg/ml if severe asthma. We measured the output of each nebulizer before the study and standardized them by using different flow rate to generate the same output between the nebulizers.

ABBREVIATIONS

AHR	Airway hyperresponsiveness
ATS	American thoracic society
AUC	Area under curve
FEV ₁	Forced expiratory volume in 1 sec
PC ₂₀	Provocative concentration of a substance causing a 20% fall in FEV ₁
ROC	Receiver operator characteristic.

FEV₁ was measured after administration of methacholine at the starting dose and doubling doses thereafter up to 8.0 mg/ml and PC₂₀ was defined by interpolation (PC₂₀ was interpolated using log-linear graph paper for a 20% fall in FEV₁ from post-saline value). Sensitivity and specificity were calculated using a cross-tabulation of asthma diagnosis with PC₂₀ cut-off values, in 0.5 mg/ml increments from 1.0 to 8.0 mg/ml methacholine, and plotted as receiver operator characteristic (ROC) curves. This was done for all children and then stratified by gender and by atopy status. The "optimal cut-off" was defined as that PC₂₀ conferring maximal value for sensitivity plus specificity. The "balanced cut-off" was defined as that PC₂₀ associated with the most closely approximated sensitivity and specificity and was so defined from the perspective of those wanting to maximally balance sensitivity with specificity (i.e., that farthest "up and left" on a standard ROC curve). However, realizing that different clinical and research circumstances make both specificity and sensitivity of variable priority, we present specificity and sensitivity associated with each discrete methacholine concentration. Sensitivity and specificity of PC₂₀ cut-offs for recurrent wheeze, a secondary analysis performed primarily for comparison to the analyses using allergist-diagnosed asthma, were derived similarly.

This study has the approval of the Ethics Committees of the University of British Columbia and the University of Manitoba approved the study. Parents signed informed consent and children gave verbal consent at age 7.

RESULTS

Of the 348 children (161 females; 187 males) who completed testing at year 7, 70 (20.1%) were diagnosed with asthma. Although gender and age were similar in children whether or not they had asthma, those children with a clinical diagnosis of asthma had a greater prevalence of atopy (73.4% vs. 39.2%), a family history of asthma (89.2% vs. 74.6%), and lower percent-of-predicted FEV₁ (91.7 vs. 95.0) compared to children without asthma.

The optimal cut-off was a PC₂₀ of 3 mg/ml methacholine (sensitivity 80.0%, specificity 49.1%; Table 1). Increasing concentrations of methacholine conferred progressively greater sensitivity and progressively lesser specificity. For example, if <8 mg/ml was used as the cut-off, a much higher sensitivity (92.3%) was observed but at the cost of much lower specificity (26.6%). Of those children not diagnosed with asthma, 50.9% would have AHR when using the 3 mg/ml cut-off value compared to 73.5% if 8 mg/ml was used instead. The area under the ROC curve (AUC) was 0.699, with the "balanced" cut-off (closest point to the ideal classification point of 100% specificity and 100% sensitivity) being 2 mg/ml (Fig. 1) which conferred a sensitivity of 63.1% and specificity of 64.7%. Of those with a PC₂₀ <2 mg/ml yet without a

TABLE 1—Sensitivity and Specificity of AHR According to Physician Diagnosed Asthma at Age 7 Using 0.5 mg/ml Cut-Offs for PC₂₀

Cut points PC ₂₀ (mg/ml)	All children at year 7			
	No asthma N = 283	Asthma N = 65	Sensitivity	Specificity
8.0	208 (73.4)	60 (92.3)	92.3	26.6
7.5	206 (72.8)	60 (92.3)	92.3	27.2
7.0	204 (72.1)	60 (92.3)	92.3	27.9
6.5	198 (70.0)	60 (92.3)	92.3	30.0
6.0	191 (67.5)	59 (90.8)	90.8	32.5
5.5	187 (66.1)	58 (89.2)	89.2	33.9
5.0	183 (64.7)	58 (89.2)	89.2	35.3
4.5	173 (61.1)	57 (87.7)	87.7	38.9
4.0	164 (58.0)	53 (81.5)	81.5	42.0
3.5	156 (55.1)	53 (81.5)	81.5	44.9
3.0	144 (50.9)	52 (80.0)	80.0	49.1
2.5	131 (46.3)	47 (72.3)	72.3	53.7
2.0	100 (35.3)	41 (63.1)	63.1	64.7
1.5	86 (30.4)	32 (49.2)	49.2	69.6
1.0	35 (12.4)	24 (36.9)	36.9	87.6

diagnosis of asthma, 32% were diagnosed with allergic rhinitis. Stratified by gender, the optimal and balanced cut-offs were unchanged in females, but for males the optimal cut-off decreased to 2 mg/ml (while the balanced cut-off remained 2 mg/ml; data not otherwise shown). Stratified by atopy, neither the optimal nor the balanced cut-offs changed.

For recurrent wheezing, the optimal cut-off point was a PC₂₀ of <4.5 mg/ml methacholine (sensitivity 85.7%, specificity 36.6%) while the balanced cut-off point was a

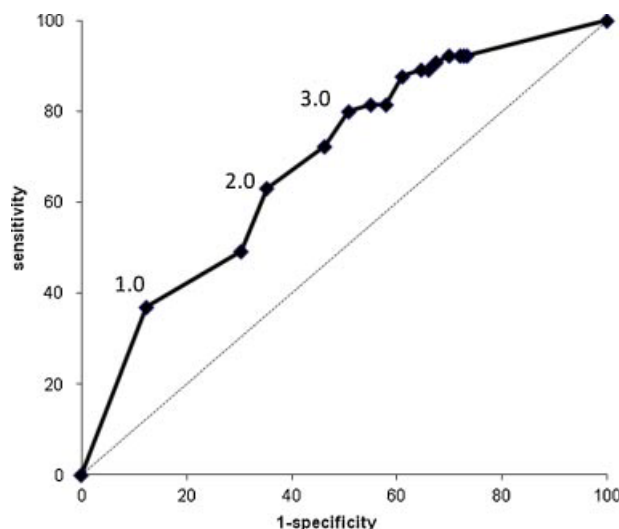


Fig. 1. Receiver-operator-characteristic curve comparing sensitivity and (1-specificity) percentages of PC₂₀ resulting from comparisons to physician diagnosed asthma at age 7. Legend: Each closed square represents a concentration of methacholine beginning at 1.0 mg/ml (lower left) and increasing in 0.5 mg/ml increments to 8 mg/ml (upper right).

PC₂₀ of <1.5 mg/ml methacholine (sensitivity 59.5%, specificity 62.1%).

DISCUSSION

In our cohort of 7-year-old children at high risk of asthma, we found that the PC₂₀ cut-off that maximizes the sum of sensitivity and specificity of AHR in the diagnosis of asthma, in children within a high-risk birth cohort, was 3 mg/ml. The cut-off conferring the most balanced sensitivity and specificity was 2 mg/ml. Either of these cut-offs for PC₂₀ to define AHR is considerably less than the value of 8 mg/ml which we feel is often offered too-simplistically as a cut-off value⁷ though in some circumstances this may understandably be with particular deference to sensitivity over specificity. A cut-off of 2–3 mg/ml does concur with a number of recent publications evaluating pediatric populations.^{8–11} For instance, Liem et al.¹⁰ reported that in 8-year olds a PC₂₀ of ≤4 mg/ml had the most balanced sensitivity (66.1%) and specificity (64.0%) for the diagnosis of all asthma irrespective of atopic status. Niggemann et al.,¹¹ using titrated bronchial histamine challenges of the reservoir method, derived an even lower cut-off for PC₂₀ of <1.0 mg/ml to define AHR in 7-year-old children. In our study, if the cut-off point of 8 mg/ml was used, the majority (73.5%) of the children without diagnosed asthma would be considered to have AHR. Conversely, there were several children with pediatric allergist-diagnosed asthma who did not have AHR even at the any of the lower PC₂₀ cut-off values. This may be due to the fact that methacholine reactivity is known to be normal in some individuals with clinical asthma and/or because AHR may vary considerably over relatively short time-frames based on temporal proximity to exposures such as allergens and/or irritants.¹² We did not have the short-term temporal resolution in our data to explore this directly. However, we suggest that measurement of AHR is most appropriate when performed concurrent with asthmatic symptoms.

Using wheeze as the outcome, the specificity at any given PC₂₀ cut-off was lower than that for asthma; accordingly, the optimal and balanced PC₂₀ cut-points for wheeze were slightly higher and lower than those for asthma as the outcome (data not shown). In contrast, another rare study able to compare the balanced cut-off for asthma versus wheezing in 7-year olds¹¹ found the cut-offs to be more similar, suggesting that our pediatric allergists were appropriately basing the diagnosis of asthma both on perceived functional (though they were blinded to PC₂₀) as well as symptomatic elements.

Others have speculated on differences in PC₂₀—asthma relationships between children and adults. Le Soeuf¹³ suggested that children are likely to receive a larger dose, relative to lung size, of methacholine than adults if the “adult” protocol is used, and furthermore, the younger the

child, the larger the “overdose” may be, and differential gender effects may be attributed to gender-specific anatomic growth rates.¹⁴ The increased responsiveness to methacholine would thereby be attributed to the larger dose rather than increased intrinsic responsiveness. Nonetheless, our data may be clearly used to derive sensitivity and specificity for the pediatric allergist’s diagnosis of asthma. There are limitations in generalizing these findings, as not all pediatric allergists may operate clinically as ours did, and we suggest that, Because all of our children were 7 years old at the time of methacholine challenge, these data be applied principally to those of similar age. While we were fortunate to obtain high-quality spirometry associated with our testing, we recognize that in children <7 years old, particularly in settings unfamiliar with administering a methacholine challenge to children, the maneuvers inherent to this test become increasingly difficult to perform.

We appreciate that the cut-off for PC₂₀ at which a given patient should be considered to have asthma is context-dependent; the pre-test probability affects the posterior probability that a given PC₂₀ may be considered representative of asthma and our balanced cut-off of 2 mg/ml would not be an ideal supplement to valid individualized pre-test probability in a child. However, when there is little or no quantifiable pre-test probability (as is particularly frequent to be the case in the epidemiological research setting), it is helpful to have a cut-point at which sensitivity and specificity is most equal along with further data by which to choose cut-points for particular scenarios that prioritize sensitivity over specificity, or vice versa. Accordingly, we emphasize that “optimal” and “balanced” are terms more theoretical than practical, best viewed in the context of our *a priori* definitions, while our data allows the reader to choose a cut-off point that emphasizes sensitivity and specificity as needed; we encourage the full range of data in Table 1 to be applied to specific clinical contexts, as appropriate. For example, in a clinical scenario in which one is doubtful of a clinical diagnosis of asthma (given a reasonable alternative diagnosis) but desires to consider it nonetheless, a lower cut-off such as 1.0 mg/ml may be used to gain specificity. In particular epidemiological settings, one may prefer a cut-off point with higher sensitivity and a lower specificity so as to capture any children with asthma.

In conclusion, our study confirmed that a relatively low cut-off of methacholine dose, 2 mg/ml methacholine, best balances sensitivity and specificity for the diagnosis of asthma in high-risk children. Regardless of the PC₂₀ cut-off utilized to define AHR, AHR should only be used as an

adjunct to common clinical indicators of asthma because of its relatively low sensitivity and specificity for clinical asthma even under ideal circumstances. Furthermore, any cut-off value should be tailored to contextual needs, should account for potential temporal variability, and should be based on data from a population similar to the population of concern. There may be reasons other than asthma, such as allergic rhinitis (present in 32% of those in our cohort without asthma yet with PC₂₀ <2 mg/ml), that explain AHR in children.

REFERENCES

1. Global Initiative for Asthma. <http://www.ginasthma.com> Accessed June 8, 2010.
2. Becker A, Lemière C, Bérubé D, Boulet L-P, Ducharme FM, FitzGerald M, Thomas Kovesi on behalf of The Asthma Guidelines Working Group of the Canadian Network For Asthma Care. Canadian pediatric asthma consensus guidelines, 2003 (updated to December 2004): introduction. *Can Med Assoc J* 2005;173:S12–S14.
3. Backer V, Bach-Mortensen N, Dirksen A. Prevalence and predictors of bronchial hyperresponsiveness in children aged 7–16 yrs. *Allergy* 1989;44:214–219.
4. James A, Ryan G. Testing airway responsiveness using inhaled methacholine or histamine. *Respirology* 1997;2:97–105.
5. Chan-Yeung M, Ferguson A, Watson W, Dimich-Ward H, Rousseau R, Lilley M, DyBuncio A, Becker A. The canadian childhood asthma prevention study: outcomes at 7 years of age. *J Allergy Clin Immunol* 2005;116:49–55.
6. Cockcroft DW, Killian DN, Mellon JJ, Hargreave FE. Bronchial reactivity to inhaled histamine: a method and clinical survey. *Clin Allergy* 1977;7:235–243.
7. American Thoracic Society Guidelines for methacholine and exercise challenge testing—1999. *Am J Respir Crit Care Med* 2000;161:309–329.
8. Godfrey S, Springer C, Bar-Yishay E, Avital A. Cut-off points defining normal and asthmatic bronchial reactivity to exercise and inhalation challenges in children and young adults. *Eur Respir J* 1999;14:659–668.
9. Joseph-Bowen J, de Klerk NH, Firth MJ, Kendall GE, for the Raine Study Group, Holt PG, Sly PD. Lung function, bronchial responsiveness, and asthma in a community cohort of 6-year old children. *Am J Respir Crit Care Med* 2004;169:850–854.
10. Liem JJ, Kozyrsky AL, Cockcroft DW, Becker AB. Diagnosing asthma in children: what is the role for methacholine provocation testing? *Pediatr Pulmonol* 2008;43:481–489.
11. Niggemann B, Illi S, Madloch C, Völkel K, Lau S, Bergmann R, von Mutius E, Wahn U, and MAS-Study Group. Histamine challenges discriminate between symptomatic and asymptomatic children. *Eur Respir J* 2001;17:246–253.
12. O’Byrne PM, Inman MP. Airway hyperresponsiveness. *Chest* 2003;123:411S–416S.
13. LeSoeuf PN. Validity of methods used to test airway responsiveness in children. *Lancet* 1992;239:1282–1284.
14. Neve V, Girard F, Flahault A, Boule M. Lung and thorax development during adolescence: relationship with pubertal status. *Eur Respir J* 2002;20:1292–1298.