Paediatric Respiratory Reviews 16 (2015) 68–74

Contents lists available at ScienceDirect

Paediatric Respiratory Reviews

Review Article

Exhaled nitric oxide and other exhaled biomarkers in bronchial challenge with exercise in asthmatic children: current knowledge

Mario Barreto *, Rosanna Zambardi, Maria Pia Villa

Pediatric Unit, Sant'Andrea Hospital, NESMOS Department, Faculty of Medicine and Psychology, Sapienza University of Rome, Rome, Italy

EDUCATIONAL AIMS

- To discuss the potential role of the fractional concentration of exhaled nitric oxide (FENO) in assessing exercise-induced bronchoconstriction (EIB).
- To illustrate changes in FENO and airway-NO exchange dynamics following exercise challenge.
- To review other exhaled biomarkers currently being investigated to assess EIB in asthmatic children.

ARTICLE INFO

Keywords:
Exercise challenge test
Bronchoconstriction
Exhaled nitric oxide
Biomarkers
Asthma
Atopy
Children.

SUMMARY

The fractional concentration of exhaled nitric oxide (FE_{NO}), a known marker of atopic-eosinophilic inflammation, may be used as a surrogate to assess exercise-induced bronchoconstriction (EIB) in asthmatic children. The predictive value of baseline FE_{NO} for EIB appears to be influenced by several factors, including age, atopy, current therapy with corticosteroids and measurement technique. Nonetheless, FE_{NO} cut-off values appear to be able to rule out EIB. FE_{NO} levels decrease during EIB, apparently through neural mechanisms rather than by decreased airway-epithelial surface. Partition of FE_{NO} into proximal and peripheral contributions of the respiratory tract may improve our understanding on NO exchange during exercise and help to screen subjects prone to EIB.

Other biomarkers of inflammation and oxidative stress contained in exhaled gases and exhaled breath condensate (EBC) may shed light on the pathophysiology of EIB. Exhaled breath temperature is a promising real-time measurement whose routine use for assessing EIB warrants further investigation.

INTRODUCTION

The fractional concentration of nitric oxide in exhaled air (FE_{NO}) closely reflects atopic-eosinophilic airway inflammation [1–4]. FE_{NO} measurement is currently being investigated on account of its potential role as a means of monitoring asthmatic patients in addition to its lung function testing and clinical roles [5,6]. Given its non-invasive nature, this marker appears to be particularly suited to the assessment of asthma in children. FE_{NO} may also help to assess exercise-induced bronchoconstriction (EIB) since the severity of EIB correlates with airway eosinophilic inflammation in EIB patients [7,8].

Exercise is a common trigger of airway narrowing in asthmatic children, typically within the first 5-10 minutes of exercise cessation [9]. Post-exercise symptoms such as cough, chest pain or dyspnea may be present, though the patient's and parents' perception of these symptoms may be in contrast to results of the exercise challenge test [10,11]. EIB, which occurs in 70–90% of asthmatic children, is indicative of poor asthma control, but improves with appropriate asthma therapy [12–15]. A post-exercise FEV_1 fall of at least 10% from baseline is diagnostic of EIB [9,16]. The FEV_1 fall is more pronounced as asthma severity increases; however, baseline FEV_1 does not predict outcomes of exercise testing [11]. The most likely mechanism of EIB in asthmatic subjects is airway dehydration and hyperosmolarity from hyperventilation, with the subsequent release of inflammatory mediators such as histamine and leukotrienes. Hyperventilation, particularly in healthy subjects (e.g. athletes), can also induce EIB through airway cooling and rewarming, followed by bronchial edema and plasma exudation [17,18].

* Corresponding author. Clinica Pediatrica, Ospedale Sant'Andrea, Via di Grottarossa 1035-1039, 00189 Roma (Italia). Tel.: +39/0633775279; fax: +39/0633775941.
E-mail addresses: mario.barreto@uniroma1.it, mariobarreto@tiscali.it (M. Barreto).

1526-0542/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.prrv.2013.11.006

The main advantage of the exercise challenge test is that, unlike other tests (e.g. methacholine or histamine), it does not stimulate the bronchial receptors directly, but elicits an inflammatory response indirectly, mimicking the natural pathophysiology of asthma [19]. The main disadvantage of this test is the fact that it needs to be used repeatedly to assess asthma control in children, and hence becomes unpractical, expensive and time-consuming. The measurement of FE_{NO} has been suggested as a surrogate of EIB because it reflects asthmatic-airway inflammation, it can be measured easily in real-time, and it can be repeated in outpatients at every follow-up examination [20]. Assessing FE_{NO} could reduce the need for the exercise challenge test, thereby avoiding the use of this test in subjects in whom EIB is unlikely.

This brief review focuses on the relationship between baseline FE_{NO} measurements and EIB in asthmatic children. Moreover, we discuss post-exercise FE_{NO} changes, which provide insights into the airway sources of nitric oxide (NO) during EIB. We also comment on the use of other exhaled biomarkers in bronchial challenge with exercise in asthmatic children, including biomarkers of inflammation, oxidative stress, acidity and temperature in the airways. The few reports available on these biomarkers in EIB are discussed in the final part of this manuscript.

ORIGIN AND SOURCES OF NITRIC OXIDE IN THE AIRWAY

Nitric oxide (NO) is a widely distributed free radical that exerts a number of functions in the human body, including vasodilation, bronchodilation and neurotrasmission, as well as defence against several microorganisms [21,22]. Production of NO in the respiratory system arises from oxidation of L-arginine to L-citruline via several synthase isoforms, two of which are constitutive, i.e. endothelial (eNOs) and neuronal (nNOs). The activity of nNOS in non-adrenergic non-cholinergic nerves induces bronchial muscle relaxation and prevents bronchial obstruction [23]. Both constitutive NOs are calcium- and calmodulin-dependent and can be stimulated by several mediators such as bradykinine, acetylcholine, histamine, leukotrienes and platelet-activating factor [24]. The third NO synthase is inducible (iNOs) and is found in airway epithelial cells, endothelial and smooth muscle cells, macrophages and neutrophils; iNOs is upregulated by allergens, infections and toxins [24].

The expression of iNOs increases during inflammatory processes and is induced by several interleukins, interferon and tumor necrosis factor [24].

Increased eosinophil activity in the mucosal airway is related to high NO concentrations in the airway lumen [3,25,26]. Excessive NO production leads to the formation of NO derivatives, particularly peroxynitrite, a potent oxidant that further enhances airway inflammation [27].

Most of the NO in the airway lumen diffuses through the airway epithelium, though the oropharynx may also contribute significantly to NO diffusion; NO concentrations are lower in the alveoli, which display a high NO-reabsorption rate by capillary haemoglobin [5,28]. When NO is released into the airway lumen, diffusive and convective forces interact to drive the rate of NO reabsorption by blood vessels (mainly in the distal airways and alveoli) and the rate of NO excretion at the mouth opening, where it can be measured at a determined expiratory flow, i.e. the so-called FE_{NO}. Low expiratory flow rates increase accumulation of NO in the airway lumen, which in turn results in high FE_{NO} levels; conversely, high expiratory flow rates dilute NO in the airway lumen with the low-NO-concentrated alveolar gas, which results in low FE_{NO} levels [5,29–31].

FE_{NO} MEASUREMENT

For the preceding reasons, FE_{NO} should be measured during a constant expiratory flow; expired gas is analysed in real time, usually by chemiluminescence or electrochemical devices. The recommended maneuver consists in a single vital capacity exhalation against a resistance of at least 5 cmH_2O to ensure closure of the soft palate, thus avoiding contamination with the high NO concentrations coming from the nasal and paranasal cavities [5,31]. Most children over 6 years of age can maintain a constant expiratory flow of 50 mL/s for at least 3 seconds [32]. For younger cooperative children, the target expiratory flow can be obtained with the help of a resistor, which can counteract airflow variations close to 50 mL/s [20]. Other techniques for measuring FE_{NO}, which are used above all on infants and less cooperative patients, are not suitable for FE_{NO} assessment during airway challenge with exercise.

Several other factors besides expiratory flow can influence FE_{NO} measurements. These factors include age, atopy (especially during allergen exposure), current respiratory infections, passive smoking, recent food intake and ambient conditions. Thorough reviews on standardized procedures for FE_{NO} measurements are available [5].

FE_{NO} TO ASSESS BRONCHIAL CHALLENGE WITH EXERCISE

Several studies based on a treadmill or bicycle ergometer have investigated the relationship between baseline FE_{NO} and bronchial response to exercise in asthmatic children. [11,33–37] Their results suggest that increased baseline FE_{NO} levels could be used as a surrogate of EIB, [11,33,34,36] as findings from studies on asthmatic adults also indicate [38–42]. Baseline FE_{NO} levels correlate with the degree of post-exercise fall in FEV_1; [11,33,34] consequently, the baseline FE_{NO} is higher in subjects with EIB than in those without [11,33,34,36].

Most studies have used a post-exercise FEV_1 fall of either 12% [33,35] or 15% [11,34,36] as a cut-off for defining EIB. Scollo et al. studied 24 asthmatic children between 6 and 13 years of age (20 atopic, 17 treated with inhaled corticosteroids, ICs); they also included an age-matched control group (n=18) for baseline comparisons alone. [33] Patients with EIB displayed a higher baseline FE_{NO} than patients without EIB or control children, though differences reached statistical significance in the control group alone (12.3 ± 1.6 vs 9.1 ± 1.0 vs 6.1 ± 0.2, $p<0.01$ vs control group). The authors reported a correlation between baseline FE_{NO} and the maximum post-exercise fall in FEV_1 ($r=0.61$, $p<0.01$) [33]. Similar findings were described by Terada et al. in 39 atopic asthmatic children (25 treated with ICs) and 6 healthy controls; patients with EIB yielded a significantly higher baseline FE_{NO} than controls though not than asthmatic children without EIB [34]. We did not find statistically significant differences in baseline FE_{NO} between patients with EIB and those without in 46 ICs-naïve (21 atopic) asthmatic children in our study either [35]. By contrast, a significantly higher baseline FE_{NO} in subjects with EIB than in those without has been reported in two other studies; approximately half of the patients in both those studies were being treated with ICs and over 70% were atopic [11,36].

The contrasting results in all the afore-mentioned studies suggest that several factors may affect the relationship between baseline FE_{NO} and exercise challenge outcomes. Although there were differences between the various studies in the subjects' characteristics (atopy and therapy with ICs), in the methods (FE_{NO} technique and expiratory flow) and in the criteria used to define EIB (percent fall in FEV_1), overall the information they yield supports the usefulness of FE_{NO} measurement in exercise challenge testing (Table 1).