

Paradoxical low nasal nitric oxide in nasal polyposis

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Summary

Background Nitric oxide (NO) is synthesized in the respiratory tract. Three isoforms of NO synthase have been described in man, with the inducible form related to inflammatory disease. In the paranasal sinuses constitutive production of nitric oxide has been demonstrated, with levels of 20–25 p.p.m. being found in sinus puncture. Nasal polyposis is a chronic inflammatory condition in which inducible nitric oxide synthase (iNOS) expression is elevated in nasal polyp epithelium.

Objectives 1. Measurement of upper airway nitric oxide in nasal polyposis patients compared with those with allergic rhinitis, and with normal controls. 2. To assess the effect of polyp treatment on nasal NO levels.

Methods NO levels (parts per billion) were measured in nasal and pulmonary exhaled air using a LR 2000 Logan Sinclair nitric oxide gas analyser. This utilizes the chemiluminescence principle. Eighty-two patients were studied: 44 with rhinitis, but without polyps, and 38 with nasal polyps. NO levels were compared with those of 20 normal controls. In 23 further polyp patients, levels were measured pre- and post-treatment and the changes were compared with alterations in polyp size, as assessed by rigid nasendoscopy.

Results Nasal NO levels were significantly lower (Kruskal–Wallis, $P = 0.000$, $\chi^2 = 27.5$, d.f. = 3) in patients with polyps than those found in uncomplicated allergic rhinitis. NO levels were correlated directly with extent of polyposis as graded by the Lund-McKay index. Successful treatment, with reduction in polyp volume, was associated with a rise in NO levels ($P = 0.042$).

Conclusion NO levels are low in nasal polyposis, despite high levels of iNOS, possibly related to blockage of the ostiomeatal complex and failure of NO generated constitutively in the sinuses to reach the nasal airway. A rise in the NO levels is seen with successful polyp treatment, and is proportional to the reduction in endoscopically assessed polyp size, suggesting that with both medical and surgical therapy, the ostiomeatal complex obstruction is decreasing. We propose the following scenario. Nasal NO levels are the result of two processes: inducible NO production by inflamed nasal mucosa plus constitutive sinus mucosal production, detectable in normals. In uncomplicated allergic rhinitis with patent sinus ostia NO levels tend to be elevated, but when inflammation is sufficient to obstruct sinus ostia (as in nasal polyps), NO levels fall because sinus NO makes the major contribution.

Keywords inducible nitric oxide synthase, leukotriene receptor antagonist, nasal polyposis, nitric oxide, oral corticosteroids, polypectomy

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Introduction

Nasal polyposis is a relatively common disease affecting 0.2 to 1% of the adult population in the United Kingdom at some time in their lives [1]. There are different varieties of polyps [2], but the commonest form is inflammatory in nature with many eosinophils, mast cells and lymphocytes in a stroma covered by an epithelium, which stains heavily for inducible nitric oxide synthase [3, 4]

Nitric oxide, a free radical gas is an important autocrine and paracrine messenger generated from arginine by a family of enzymes, the nitric oxide synthases (NOS) [5]. Some of these enzymes are constitutive, the remainder are expressed by most cells involved in inflammation and are known as inducible NOS (iNOS). Nitric oxide has antiviral and antibacterial properties and plays a major role in innate immunity. Recently nitric oxide has been found to stimulate ciliary mobility and to be absent or very low in primary ciliary dyskinesia [6].

Nitric oxide has been measured in the upper respiratory tract using the chemiluminescence principle [7], and tends to be elevated in inflammatory disorders such as allergic rhinitis [8], although levels are very variable. It would therefore seem likely that with higher epithelial iNOS levels [9] nasal polyposis would

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be associated with higher upper respiratory NO levels. However, constitutive NO production also occurs in the upper respiratory tract as demonstrated by Lundberg, who punctured maxillary sinuses and found continuous production of NO [10]. Presumably this normally diffuses into the nose via the nose/sinus junction, the ostiomeatal complex.

The aim of this study was to investigate NO levels in patients with allergic rhinitis and nasal polyposis, and compare them with normals. We also aimed to see whether NO levels correlated with the extent of polyp tissue present in the nose as assessed endoscopically by an observer who was unaware of the NO result. Thereafter we studied a second group of patients with nasal polyposis to see how nitric oxide levels change with treatment, and with alterations in the quantity of polyp tissue.

Materials and methods

Study group A

One hundred and two individuals had nasal NO measurements in this part of the study. These included patients with allergic rhinitis, but no polyps ($n=44$; age = 39 ± 13.6 years), and those with nasal polyposis ($n=38$; age = 45.7 ± 4.5). They were compared with normal non-rhinitic controls ($n=20$; age = 36.9 ± 11.6). Skin prick tests (Allergopharma, London, UK) were positive in 21 nasal polyp patients. Patients with cystic fibrosis primary ciliary dyskinesia, smokers and individuals with recent (2 weeks) upper respiratory tract infections were excluded. All patients were on topical corticosteroids (fluticasone propionate) at standard dosage (200 µg/day); no patient received oral corticosteroids for at least 3 months prior to testing.

Study group B

This group included 23 patients with nasal polyposis (M:F = 16.7; age = 48.8 ± 4.2) who were managed with conservative medical treatment. All patients received topical corticosteroids. In addition, some were treated with oral corticosteroids ($n=3$) or a leukotriene receptor antagonist ($n=5$). NO levels were measured pre-treatment and repeated 3–6 months after treatment.

Nitric oxide (NO) measurement

NO levels (part per billion) were measured in nasal and exhaled air using a LR 2000 Logan Sinclair nitric oxide gas analyser according to the recommendations of Kharitonov et al. [11]. The subject was sitting relaxed and a probe was inserted into one nostril. The subject took a full inspiration and then held his or her breath, in order to close the soft palate, while the nasal NO measurement was made. The NO value taken was that at the plateau that occurred after 30 to 60 s in most subjects. Carbon dioxide was monitored in parallel in order to confirm that the soft palate was closed and that no contamination was occurring from the lower airways. Measurements were made in duplicate on both sides of the nasal cavity and the mean measurement was used.

Pulmonary NO was measured with an oral probe while the subject exhaled against a resistance sufficient to close the soft palate. Again the plateau measurement occurring after 40–60 s was used. Measurements were made in duplicate.

Rigid nasendoscopy

This procedure was performed using 2.7/4 mm ($0^\circ/30^\circ$) rigid endoscopes. The patients were grouped according to the extent of polyposis using the Lund-Mackay staging system [12]. Grade 0 signifies the absence of polyps, grade I polyps are confined to the middle meatus, Grade II polyps extend below the middle turbinate and grade III polyps occupy most of the nasal cavity.

Data were collected and entered into a statistical package, SPSS v7.0 for Windows 98. Non-parametric tests (Kruskal–Wallis, Mann–Whitney U) were used in analysis with significance level set at $P=0.05$. Graphs were produced using the same program. In group B patients Pearson's correlation was used to study the relationship between change in endoscopy scores and nasal NO levels. Significance level was set at $P=0.01$.

Results

Group A

Repeated measurements in a single individual showed less than 10% variance. The range of NO values for normal individuals was 445–1000 (740.9 ± 148.1). Patients with allergic rhinitis had a wider range of values (150–1300; 659.8 ± 304.8). Our study included 44 grade 0, 14 grade I, 16 grade II and 8 grade III polyp patients. Nasal NO levels were significantly lower in patients with polyps (Kruskal–Wallis, $P=0.000$, $\chi^2=37.6$, d.f. = 4), than those found in uncomplicated allergic rhinitis. NO levels correlated directly with the extent of polyposis (Fig. 1). Within each group there was no difference in NO levels between skin prick positive and negative patients.

Pulmonary NO levels were much lower than those in the nose, as noted by previous workers [11]; they were not significantly different between asthmatics and non-asthmatics ($P=0.51$), probably because the asthmatics were on regular treatment with inhaled corticosteroids (Fig. 2).

Aspirin sensitivity was judged by nasal lysine aspirin challenge [13], and was present in three grade I, five grade II and two grade III polyp patients. NO levels were not significantly

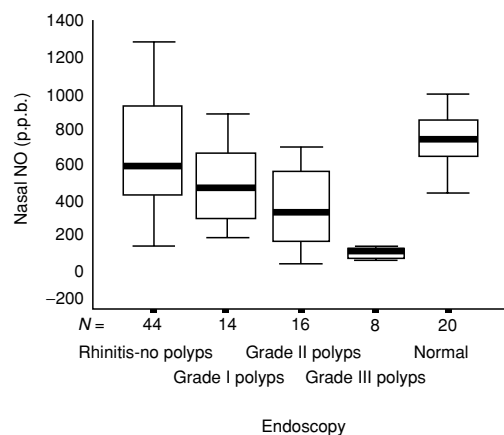


Fig. 1. Nasal NO levels in different patient groups. The median and interquartile values (50% lie within the box) are shown. NO levels were significantly lower in nasal polyp patients compared with those with allergic rhinitis. There was a correlation between NO levels, and extent of polyposis (Kruskal–Wallis, $P=0.000$).

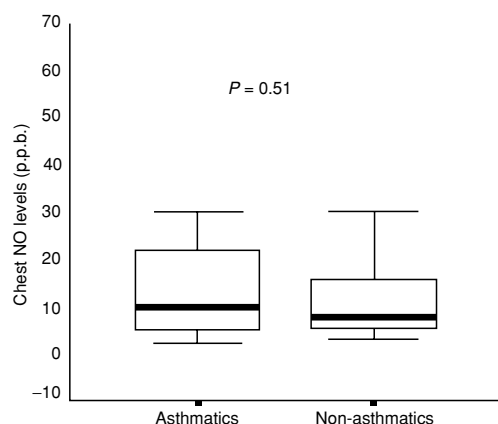


Fig. 2. Chest nitric oxide levels in asthmatics and non-asthmatics. The median and interquartile values (50% lie within the box) are shown.

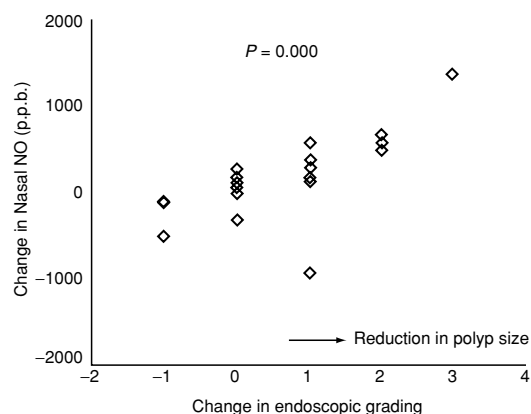


Fig. 3. Change in NO levels seen in 23 nasal polyposis patients after medical treatment. The increase in NO levels correlates with the reduction in extent of nasal polyposis (Pearson's correlation, $P = 0.000$).

different between aspirin-sensitive and aspirin-tolerant patients within each group.

Group B

Treatment of polyps was associated with a rise in NO levels ($P = 0.042$). There was a significant correlation between visual reduction in polyp size (as judged by Lund-Mackay staging system) and an increase in nasal NO levels (Fig. 3; $P = 0.000$).

Discussion

In the lower respiratory tract nitric oxide levels are low, normally less than 20 p.p.b. [14]. Raised levels indicate inflammation, which can have many causes, including allergy and infection [15]. In the upper respiratory tract the situation is more complex as constitutive nitric oxide production in high bactericidal levels (20 to 25 p.p.m.) occurs in the paranasal sinuses [10]. The inducible form of nitric oxide synthase is also present in nasal epithelium and is raised in nasal polyposis, especially the aspirin-sensitive variety [9].

This study shows that the low NO level seen in nasal polyposis probably reflects ostiomeatal complex obstruction and suggests that upper respiratory tract NO is predominantly formed in the sinuses. However, the differences between grade I, II and III polyps are difficult to explain solely on the basis of sinus blockage as ostiomeatal complex obstruction is likely in all three groups. The higher levels seen with smaller polyps may reflect inducible NO synthase activity in the nasal epithelium [3, 4, 9]. We have shown that inflammatory changes including raised eosinophils occur in inferior turbinates of aspirin-sensitive nasal polyp patients [16]. Thus upper respiratory NO levels in nasal polyposis are the result of two processes; constitutive production by sinus mucosa and inducible production by polyp and nasal epithelium.

The fact that successful polyp treatment results in a rise in NO levels suggests that sinus NO is the predominant form, because reduction in the polyp size and therefore epithelium would be expected to reduce iNOS levels and inducible NO production. The dual origin of nasal nitric oxide would also explain the wide variation of values seen in patients with allergic rhinitis. Those with immediate allergic reactions show high values, whereas in those where the late-phase reaction predominates, and ostiomeatal complex obstruction occurs, NO levels fall. This series of events has been found after nasal allergen challenge (Turner P, Scadding G, unpublished observations). Indeed in allergic rhinitis treatment with topical corticosteroids tends to reduce NO levels [17].

In study A our allergic rhinitis patients had NO levels measured whilst taking topical nasal steroids in order to conform with the polyp patients who were on long-term topical nasal steroid treatment to prevent recurrence. This may have decreased the measured NO levels in the allergic rhinitis group; however, it cannot account for the differences between polyp patients with grade I, II and III disease.

The low level of NO seen with nasal polyposis could contribute to the frequent occurrence of sinus infection in these patients because NO plays an important role in host defence and stimulates ciliary mobility. Nitric oxide may play a regulatory role in polyposis as it can disrupt fas-receptor-mediated signalling events, and thus inhibit apoptosis of inflammatory cells [18]. Low NO levels could predispose to the longevity of eosinophils seen in polyp tissue [19].

It remains to be seen whether obstruction of upper respiratory tract constitutive NO production has implications for the comorbid associations of nasal polyposis, such as infective sinusitis and asthma.

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