A pilot study of oral L-arginine in cystic fibrosis

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Received 25 June 2004; accepted 29 October 2004

Abstract

Exhaled nitric oxide has previously been found to be low in cystic fibrosis. The aim of this study was to determine whether exhaled nitric oxide levels would increase in response to oral L-arginine supplementation administered daily for 4 weeks. Exhaled and nasal nitric oxide was measured weekly. Plasma L-arginine levels increased in response to supplementation but this was not reflected in an increase in eNO levels.

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Keywords: Children; Exhaled/nasal nitric oxide

1. Introduction

A number of studies have indicated that levels of exhaled and nasal nitric oxide (NO) are low in patients with cystic fibrosis (CF) [1]. However, the mechanism(s) underlying this observation and the impact, if any, of reduced airways NO are unclear. Possible reasons for low measured levels of exhaled and nasal NO include a primary reduction in ability to synthesis NO; trapping of NO in mucus with subsequent conversion to nitrate and nitrite; impairment of NO synthesis as a consequence of chronic inflammation; pancreatic insufficiency leading to low levels of L-arginine; and possible consumption of NO by the denitrifying organism Pseudomonas aeruginosa, which utilises the enzyme NO reductase. A recent study found that nitric oxide levels, and its metabolites, were no different in young CF patients when compared to controls, and suggested that decreased NOS II expression occurs with increasing airway inflammation [2].

Nitric oxide is produced in the airways from oxidation of L-arginine via nitric oxide synthases (NOS), of which there are three isoforms. It has a number of important functions including vasodilation, bronchodilation, modulation of ciliary activity, and enhancement of neutrophil migration, and it appears to play an important role in the inflammatory response [3]. Impairment of NO synthesis may therefore have implications for the progression of the disease. Further evidence to support the suggestion that NO levels in the airway may have an impact on disease progression comes from the suggestion that nitric oxide synthase 1 (NOS1) may act as a “modifier” gene in patients with cystic fibrosis. The proximal region of the NOS1 gene contains dinucleotide GT repeats. It has recently been shown that patients with more than 27 repeats in this region have a lower annual FEV1 loss [4]. This implies an association between the NOS1 gene locus and the progression of lung disease in CF, which is independent of the CFTR genotype. A previous study showed that CF patients with a higher number of repeats in the NOS1 gene had lower exhaled nitric oxide levels [5], and this may provide the link between the genotype and disease progression. If this is the case, it is possible that increasing airway nitric oxide levels could be of therapeutic benefit for cystic fibrosis patients.

L-arginine is an essential amino acid with a short half-life due to rapid metabolism. It is used therapeutically to treat certain metabolic diseases. A number of studies suggest that levels of NO in the airways may rise in response to administration of L-arginine. Nasally inhaled nebulised L-arginine has been shown to increase nasal nitric oxide levels and decrease nasal mucociliary clearance time in patients with primary ciliary dyskinesia (PCD) [6]. It has also been shown to increase exhaled nitric oxide levels in asthmatic

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doi:10.1016/j.jcf.2004.10.001
In another short-term study undertaken in patients with CF, levels of exhaled and nasal NO increased in response to L-arginine administered by infusion, although subnormal throughout [9]. Pulmonary function tests were unchanged.

The aim of this pilot study was to determine whether oral supplementation of L-arginine administered daily for 1 month would result in increased plasma levels and whether this would be reflected in increased levels of eNO and nNO in patients with CF.

2. Method

Six patients with cystic fibrosis aged 6–16 years of age were recruited. Patients were seen weekly for a total of six visits. At each visit, they had FEV₁ and FVC measured, along with measurements of exhaled and nasal nitric oxide. Exhaled and nasal nitric oxide levels were measured via a chemiluminescence analyser (LR2500; Logan Research, Kent, UK), in accordance with published recommended standards for cooperative children [10,11]. Exhaled levels were recorded at flow rates of 50 ml/s. Three attempts were allowed at each rate and the mean was calculated. All nitric oxide levels were taken within 1 h of ingestion of L-arginine.

Oral L-arginine was commenced on week 2 and continued for 4 weeks. A dose of 150 mg/kg/day was administered in three divided doses, as advised by our metabolic department. Amino acid levels were taken at baseline, week 3, and week 6.

Ethical approval was obtained prior to the onset of this study.

3. Results

The recruited patients included three boys and three girls (mean age 12.9 years; range 6.16–15.4 years). All, but one, were homozygous for the delta F508 mutation and all had been colonised with non-mucoid Pseudomonas for more than a year prior to entry into the study. One patient withdrew at visit 5 due to an infective exacerbation. One patient declined to have amino acid levels checked at visit 3.

The mean plasma L-arginine level at entry to the study was 57 μmol/L (range 43–99 μmol/L), lying within the normal range (0–150 μmol/L). While on supplements, L-arginine levels increased significantly with a mean value at visit 3 of 154 μmol/L (range 36–294 μmol/L) and 174 μmol/L (range 78–262 μmol/L) at visit 6.

No side effects or adverse events were noted and there was no increase in the number of infective exacerbations during the 12 months following treatment.

Despite increases in plasma L-arginine levels, there was no evidence of a significant impact on either exhaled or nasal nitric oxide levels (Fig. 1a and b).

4. Conclusion

Oral supplementation resulted in an increase in plasma levels of L-arginine, the essential substrate for the production of nitric oxide, but this was not reflected in increased eNO levels. This would suggest that lack of substrate is not a major factor and that providing increased substrate does not overcome the mechanism contributing to the relatively low levels of eNO and nNO observed in patients with cystic fibrosis. It remains possible that a higher dose of L-arginine may have been necessary to increase eNO levels. However, plasma L-arginine levels were in excess of the normal range in half of the samples obtained on supplementation, suggesting that higher doses would not have had a greater impact.
Oral arginine was well tolerated by our patients. One possible adverse effect of supplementation may be to promote the growth of *P. aeruginosa*, which utilizes arginine. However, in this small pilot study, we were unable to identify any evidence that L-arginine may adversely affect the clinical status of patients colonised with *Pseudomonas*.

Despite increased circulating levels of plasma L-arginine, we were unable to identify a significant increase in exhaled NO levels. It therefore seems unlikely that significant therapeutic benefits would be derived from L-arginine supplementation in patients with cystic fibrosis.

References