

## Short papers

## Decreased exhaled nitric oxide in subjects with HIV infection

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**Abstract**

**Background** – Nitric oxide (NO) may be an important component of the host defence against infections. Endogenously produced NO is present in exhaled air and may be representative of respiratory tract production of NO. Since subjects infected with HIV are prone to develop respiratory infections, it was postulated that exhaled NO might be reduced in such individuals.

**Methods** – The exhaled concentration of NO (nl/l) and minute ventilation (l/min) were measured and exhaled NO release (nl/min/m<sup>2</sup>) calculated in 36 subjects infected with HIV (20 non-smokers, 16 smokers) and 31 non-smoking subjects with no active medical conditions.

**Results** – Exhaled NO from HIV positive individuals was less than from control subjects of similar age, height, and weight. Cigarette smoking did not account for the decreased exhaled NO in HIV positive individuals as both smoking and non-smoking HIV positive subjects had decreased exhaled NO compared with control subjects.

**Conclusion** – Exhaled NO is decreased in subjects infected with the HIV. Since NO functions in host defence against bacterial, viral, and fungal infections, reduced exhaled NO may indicate a mechanism of impaired host defence in HIV infection.

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Keywords: nitric oxide, human immunodeficiency virus.

Nitric oxide (NO) may participate in defence against viral, bacterial, fungal, and protozoal infections.<sup>1</sup> Such infections occur with increased frequency in individuals infected with the human immunodeficiency virus (HIV). Endogenously produced NO is present in the exhaled air of normal humans<sup>2</sup> and exhaled NO is increased in inflammatory lung diseases such as asthma,<sup>3,4</sup> bronchiectasis,<sup>5</sup> and viral infections.<sup>6</sup> Treatment of asthmatic subjects with corticosteroids reduces exhaled NO.<sup>7</sup> Exhaled NO is also reduced in cigarette smokers,<sup>3</sup> a population susceptible to respiratory infections. Since HIV positive individuals are at risk for respiratory tract infections, we postulated that exhaled NO might be reduced in these subjects.

**Methods**

We measured NO in the exhaled air of 36 HIV positive subjects (34 men; 20 non-smokers) recruited from an ambulatory care centre and 31 healthy non-smoking control subjects (23 men). The control subjects were recruited from hospital colleagues and coworkers, had no risk factors for HIV infection, and were taking no prescription medications. The most recent CD4 lymphocyte count for each HIV positive subject was recorded. Ten of the HIV positive subjects were receiving trimethoprim-sulphamethoxazole as prophylaxis against *Pneumocystis carinii* infection and none were receiving corticosteroids. Control subjects and HIV positive subjects were similar in terms of age (35.7 (1.8) versus 37.8 (1.4) years), height (5.9 (0.1) versus 5.8 (0.1) feet), and weight (163 (7) versus 167 (5) pounds).

Supine subjects breathed through a two way valve and mouthpiece with nose clips in place for five minutes. The inhalation port of the mouthpiece was connected to a compressed air source (NO <1 ppb) and the exhalation port was connected to a mylar collection bag. The concentration of NO in the collection bag was measured by chemiluminescence (Sievers, 270B, Boulder, Colorado, USA) and the volume of the bag was measured in a Tissot gasometer. Exhaled NO (nl/min) was calculated as the product of NO concentration (nl/l or ppb) and minute ventilation (l/min). In healthy subjects exhaled NO is proportional to body surface area (unpublished observations) so we expressed exhaled NO per m<sup>2</sup>.

Comparisons between groups were made using unpaired Student's *t* tests with *p*<0.05 being considered significant.

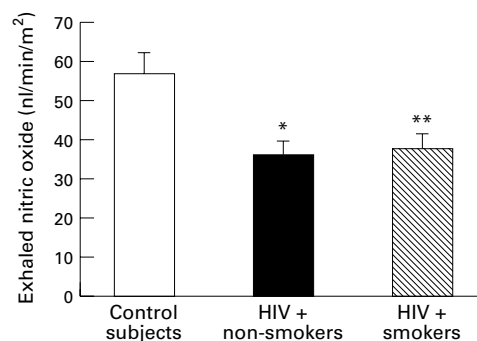
**Results**

The amount of exhaled NO from HIV positive subjects (39 (3) nl/min/m<sup>2</sup>) was less than that from the control subjects (57 (6) nl/min/m<sup>2</sup>; *p*<0.001). The reduction in exhaled NO in HIV positive subjects was present in both smokers (42 (5) nl/min/m<sup>2</sup>; *p*<0.03) and non-smokers (37 (3) nl/min/m<sup>2</sup>; *p*<0.008; fig 1). The distribution of CD4 lymphocyte counts in HIV positive subjects was 207 (34)/mm<sup>3</sup> with 19 of 36 having a count of less than 200/mm<sup>3</sup>. Four of the subjects with less than 200/mm<sup>3</sup> CD4 lymphocytes had a history of prior AIDS

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**Figure 1** Exhaled nitric oxide from healthy control subjects and HIV positive subjects (non-smokers and smokers) recruited from an outpatient HIV clinic. Levels of exhaled NO from both HIV positive smokers and non-smokers were lower than those from control subjects (control versus non-smokers,  $p < 0.008$ ; control versus smokers,  $p < 0.03$ ). There was no difference between exhaled NO values for HIV positive smokers and non-smokers.

defining opportunistic infection. There was no relation (linear regression,  $r = 0.04$ ) between exhaled NO and the most recent CD4 lymphocyte counts.

### Discussion

There is reason to suspect that endogenous NO production in the respiratory tract may be reduced in HIV infected subjects. Nitric oxide may participate in the immune response against viral, bacterial, fungal, and protozoal infections, and against tumours,<sup>1</sup> all of which occur with increased frequency in HIV infected individuals. In addition, a syndrome very similar to primary pulmonary hypertension occurs in HIV infection.<sup>8</sup> Since endogenous NO may be reduced in some types of pulmonary hypertension,<sup>9</sup> it is possible that reduced endogenous NO plays a role in the pulmonary hypertension of HIV infection.

Endogenously produced NO is present in the exhaled air of normal humans.<sup>2</sup> Both the upper and lower respiratory tracts release NO.<sup>10</sup> Exhaled NO is increased in patients with diseases associated with respiratory tract inflammation such as asthma,<sup>3,4</sup> bronchiectasis,<sup>5</sup> and viral infections.<sup>6</sup> In contrast, exhaled NO is reduced by corticosteroid therapy in asthmatic subjects<sup>7</sup> and by cigarette smoking.<sup>3</sup> The latter

may be due to high levels of NO in cigarette smoke which downregulate NO synthase. Although the relationship between exhaled NO and total respiratory tract NO production is not known, the suggestion has been made that exhaled NO may be a monitor of respiratory tract inflammation.<sup>4</sup>

Our results show that exhaled NO is reduced in HIV infection. The effect of smoking did not account for our findings since exhaled NO was reduced in both smoking and non-smoking HIV positive subjects. The lack of correlation between decreased exhaled NO and CD4 lymphocyte counts in HIV infected subjects suggests that reduced NO is not simply a function of the numbers of these immune effector cells. Our findings are consistent with the hypothesis that individuals with HIV infection have reduced endogenous NO in the respiratory tract. The causes could be multifactorial and be due to either reduced constitutive or inducible NO synthase function from progressive immunological deactivation or other mechanisms. If this hypothesis is correct, reduced endogenous NO may represent a host defence defect in HIV infection.

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