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Lung diffusing capacity for nitric oxide at lowered and raised ambient pressures

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ABSTRACT

Lung diffusing capacity for NO (DL_{NO}) was determined in eight subjects at ambient pressures of 505, 1015, and 4053 hPa (379, 761 and 3040 mmHg) as they breathed normoxic gases. Mean values were 116.9 ± 11.1 (SEM), 113.4 ± 11.1 and 99.3 ± 10.1 ml min^{-1} hPa^{-1} at 505, 1015, and 4053 hPa, with a 13% difference between the two higher pressures (P = 0.017). The data were applied to a model with two serially coupled conductances; the gas phase (Dg_{NO}, variable with pressure), and the alveolo-capillary membrane (Dm_{NO}, constant). The data fitted the model well and we conclude that diffusive transport of NO in the peripheral lung is inversely related to gas density. At normal pressure Dm_{NO} was approximately 5% larger than DL_{NO}, suggesting that the Dg factor then is not negligible. We also conclude that the density of the breathing gas is likely to impact the backdiffusion of naturally formed NO from conducting airways to the alveoli.

1. Introduction

1.1. Background

Measurements of pulmonary nitric oxide (NO) are of scientific and clinical interest mainly in two areas; (1) to monitor the activity of inflammatory airway disease from exhaled NO of endogenous origin (Persson et al., 1994; Olin et al., 2007; ATS Guidelines, 2011), and (2) to measure lung diffusing capacity (DL_{NO}) from the rate of uptake of NO from an external source. In the former case the NO level is several orders of magnitude lower (ppb) than that inhaled during a DL_{NO} maneuver (ppm) (Guénard et al., 1987; Zavorsky et al., 2008).

1.1.1. Exhaled NO

Nitric oxide in the lungs has been implicated as a potentially protective factor against pulmonary manifestations of acute mountain sickness (Busch et al., 2001; Duplain et al., 2000; Erzurum et al., 2007). It has therefore been considered of interest to establish what the effects are of a reduced ambient air pressure on pulmonary NO in healthy individuals. A second consideration is to define normal values for altitude residents, when exhaled NO is used to monitor the activity of inflammatory airway diseases such as asthma (Olin et al., 2007; Taylor et al., 2006). Potential effects of altitude include the influence of hypoxia per se (Brown et al., 2006; Hemmingsson et al., 2009), of the reduced gas density (Shin et al., 2006; Van Muylem et al., 2003) and a combination of these two factors (Hemmingsson and Linnarsson, 2009). The latter authors showed that a short-lasting exposure to normobaric hypoxia down to 10% of an atmosphere had no significant effect on exhaled NO. We recently addressed the effects of gas density on exhaled NO by exposing healthy subjects to both reduced and increased ambient pressures while at the same time maintaining normoxia in the breathing gases. Based on previous experiments with helium-oxygen breathing (Shin et al., 2006; Van Muylem et al., 2003) we had expected that the lowered gas density at altitude and the associated increased diffusivity for NO in the lung gas would increase backdiffusion of NO to the alveoli. In turn this would increase the uptake of NO to the blood resulting in a reduced partial pressure of NO in the exhaled gas (PE_{NO}). We also expected corresponding mechanisms to increase PE_{NO} at hyperbaric pressure. However, recent work from our laboratory has shown that PE_{NO} values were strikingly similar in an ambient pressure range from 0.5 to 4.0 atmospheres (Hemmingsson et al., 2012). We hypothesized that diffusive transport of NO in the gas phase in the lungs would indeed be influenced by changes of the gas density, but that these effects would be relatively small and possibly be concealed by simultaneous effects of density on convective gas transport acting

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in opposite direction (Hemmingsson et al., 2012). We reasoned that direct determinations of the lung diffusing capacity for NO (DL_{NO}) would be a way to test this hypothesis, so we undertook an additional study, now focusing on DL_{NO} while using an otherwise identical experimental design and studying the same subjects.

1.1.2. Lung diffusing capacity

Pulmonary edema is a feared component of acute mountain sickness and determinations of DL_{NO} have been used to detect interstitial pulmonary edema in otherwise healthy individuals during their stay at high altitude (de Bisschop et al., 2010, 2012). Conventionally, DL_{NO} is used to estimate the capacity for diffusive transport through the alveolo-capillary membrane, under the assumption that any effects of diffusive transport through the gas phase in the respiratory zone of the lung can be neglected (Guénard et al., 1987; Zavorsky et al., 2008). We hypothesized that such an effect indeed is small but not necessarily negligible at all ambient pressures.

We reasoned that by comparing DL_{NO} data obtained with background gases of widely differing diffusivity for NO, we would be able to estimate the quantitative role of diffusive transport through the gas phase for DL_{NO}.

1.2. Theory

The general way to characterize the passage of inhaled or endogenously formed gases from the airways to the pulmonary-capillary blood is to determine the diffusing capacity. Forster and Roughton (Forster et al., 1957) developed an algorithm to determine the diffusing capacity of the lung for a specific gas (DL), or transfer factor as it is usually called outside the United States. Below, the term DL will be used and, according to the classical model, expressed as serially connected resistances to diffusion, from the airway gas to the inside of the erythrocyte. Each subcomponent of the serial reaction chain is hence the inverse of a conductance involving both passive diffusion and chemical binding with hemoglobin (Hb). The complete transfer reaction of the lung DL for a gas x can be explained as three serially linked resistances (Cotton and Graham, 2005; Johnson et al., 1996);

$$\frac{1}{DL} = \frac{1}{Dg_x} + \frac{1}{Dm_x} + \frac{1}{(\theta_x \cdot V_c)} \quad (1)$$

defined by the conductance for x in the background gas of the respiratory zone (Dg_x), the conductance for x through the alveolo-capillary membrane (Dm_x), the conductance for uptake of gas x within the erythrocyte (θ_x), and by the volume of the pulmonary-capillary blood (V_c). The factor describing diffusion in the gas phase (Dg_x) is usually considered negligible for inhaled test gases such as CO, whereas both the amount and the oxygen saturation of the hemoglobin in the lung capillaries have measurable impacts on DL_{CO}. The reaction of NO with hemoglobin in the erythrocyte is about 280 times faster than that of CO (Meyer and Piiper, 1989; Tamhane et al., 2001; Zavorsky et al., 2008). Therefore, the resistance to the NO transport within the erythrocyte has been considered negligible (Johnson et al., 1996), and the third term in Eq. (1) ($1/\theta_x \times V_c$) could therefore be omitted in a corresponding equation for NO (Zavorsky, 2010). However, other authors have considered this term to have a finite value (Borland et al., 2010), see Martinot et al. (2013) for a detailed discussion. For the purpose of the present analysis, however, the results are based on the assumption that this third term could be neglected, thereby obtaining:

$$\frac{1}{DL_{NO}} = \frac{1}{Dg_{NO}} + \frac{1}{Dm_{NO}} \quad (2)$$

Since DL for NO is about four times that for CO (Guénard et al., 1978; Zavorsky et al., 2004), we thought that the impact of diffusivity in the gas phase (as quantified by the $1/Dg_{NO}$ term) would be possible to detect if conditions with sufficiently large differences in density and therefore also diffusivity were compared.

Eq. (2) may be further developed by assuming that:

(a) Dg_{NO} should vary in proportion to the diffusion coefficient for NO in the breathing gas. This parameter in turn varies inversely with the ambient pressure (Chang, 1985).
(b) The term $1/Dm_{NO}$ is constant across conditions.

For a range of pressures and breathing gas mixtures consistent with the above assumptions we then obtain for a given pressure:

$$\frac{1}{DL_{NO}} = \left(\frac{1}{Dg_{NO}}\right)^{*} \left(\frac{P}{P_0}\right) + \frac{1}{Dm_{NO}} \quad (3)$$

where P/P_0 is the ratio between the ambient pressure and the pressure for which Dg_{NO} is defined. The equation has the format of a linear relationship where the slope is $1/Dg_{NO}$ and the intercept with the Y axis is $1/Dm_{NO}$.

2. Methods

2.1. Subjects

Eight healthy non-smoking subjects without a history of inflammatory airway disease participated in the current study. The subjects came to the laboratory once for familiarization, physical examination, spirometry and baseline FE_{NO} measurement. They returned twice for experiments in increased or decreased ambient pressure in a combined hyperbaric and hypobaric pressure chamber. All eight subjects, four women, completed all tests at all pressure levels. Their age, height and weight ranged 21–37 years, 1.60–1.93 m and 58–87 kg, respectively.

2.2. Instrumentation and measurements

Experiments were performed at 505 ± 0 (mean ± SEM), 1015 ± 3 and 4053 ± 0 hPa ambient pressure. The corresponding values in mmHg were 379, 761 and 3040. The pressure chamber (internal volume 8 m^3) was pressurized with air but subjects breathed normoxic gas mixtures with oxygen fractions of 0.421, 0.2095 and 0.052 at 505, 1015 and 4053 hPa, respectively. Subjects were investigated at hypobaric and hyperbaric pressures on different days, in random order and always starting with control measurements at 1015 hPa. The lung diffusing capacity for NO (DL_{NO}) was measured in two subjects at a time, seated together with one of the test supervisors in the pressure chamber, using the standardized ATS/ERS techniques for diffusing capacity (ATS/ERS, 2005), which is based on the Jones and Meade methodology (Jones and Meade, 1961), with a modified 5 s breath-holding time (Zavorsky et al., 2008). After a change in ambient pressure, a 15 min waiting time was allowed to accommodate to the new environment while breathing the normoxic gas mixture. Decompression after the hyperbaric experiments was performed according to Swedish Navy standard tables and with correction for the increased nitrogen partial pressure compared to air breathing. There were no decompression symptoms in any of the subjects.

Between DL_{NO} determinations subjects were breathing through an oronasal mask and a non-rebreathing valve (Hans Rudolph Inc., Shawnee, KS, USA) from a 200 l Douglas bag via 40 mm inner diameter hoses. The supervisor kept the bag adequately filled by means of a needle valve connected to the reducing valve of tanks with compressed gas housed inside the chamber and quick-connect fittings allowed for simple change of gas source when required. Gas for argon (Ar) analysis was sampled to a mass spectrometer (Innovision A/S, Odense, Denmark) located outside the pressure chamber. Gas for NO analysis was sampled to a chemiluminescence analyzer (Eco