Microgravity decreases and hypergravity increases exhaled nitric oxide

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Karlsson LI., Kerckx Y, Gustafsson LE, Hemmingsson TE, Linnarsson D. Microgravity decreases and hypergravity increases exhaled nitric oxide. J Appl Physiol 107: 1431–1437, 2009. First published September 10, 2009; doi:10.1152/japplphysiol.91081.2008.—Inhalation of toxic dust during planetary space missions may cause airway inflammation, which can be monitored with exhaled nitric oxide (NO). Gravity will differ from earth, and we hypothesized that gravity changes would influence exhaled NO by altering lung diffusing capacity and alveolar uptake of NO. Five subjects were studied during microgravity aboard the International Space Station, and 10 subjects were studied during hypergravity in a human centrifuge. Exhaled NO concentrations were measured during flows of 50 (all gravity conditions), 100, 200, and 500 ml/s (hypergravity). During microgravity, exhaled NO fell from a ground control value of 12.3 ± 4.7 parts/billion (mean ± SD) to 6.6 ± 4.4 parts/billion ($P = 0.016$). In the centrifuge experiments and at the same flow, exhaled NO values were 16.0 ± 4.3, 19.5 ± 5.1, and 18.6 ± 4.7 parts/billion at one, two, and three times normal gravity, where exhaled NO in hypergravity was significantly elevated compared with normal gravity ($P \leq 0.011$ for all flows). Estimated alveolar NO was 2.3 ± 1.1 parts/billion in normal gravity and increased significantly to 3.9 ± 1.4 and 3.8 ± 0.8 parts/billion at two and three times normal gravity ($P < 0.002$). The findings of decreased exhaled NO in microgravity and increased exhaled and estimated alveolar NO values in hypergravity suggest that gravity-induced changes in alveolar-to-lung capillary gas transfer modify exhaled NO.

estimates of alveolar NO; analysis of exhaled NO; human centrifuge; lung diffusion; respiratory system; weightlessness

EXHALED NITRIC OXIDE (NO) is increasingly used to evaluate the inflammatory state of the lung. Endogenous NO in the lung results from the activity of three isoforms of NO synthase (NOS), one of them being triggered by inflammation. In future spaceflights, humans will risk being exposed to toxic dust on the moon and Mars, which may cause airway inflammation (12). Analysis of exhaled NO (FeNO) is a simple method to monitor airway inflammation not only in asthma (3, 8) but also after dust inhalation (e.g., Refs. 16, 27). In addition, estimates of alveolar NO (CalvNO) can be obtained from FeNO measurements (10, 20). In preparation for possible future monitoring of lung health during long-term space missions, we wanted to investigate the effects of weightlessness (microgravity) and gravity on FeNO.

Gravity influences the blood distribution within the lung (18), which in turn will induce changes in the rate of alveolar-to-lung capillary transfer [lung diffusing coefficient (Dl./) of blood-soluble gases such as NO and carbon monoxide (CO)]. This is because alveolar-to-blood transfer of gas is highly dependent of the magnitude of the contact area between the blood and the alveolar air. Thus Prisk et al. (19) and Verbanck et al. (32) have shown that, in zero gravity, Dl. for carbon monoxide (DlCO) is enhanced by 11–27%. For very short-lasting microgravity, Vaida et al. (30) showed that both DlNO and the membrane component of DlCO were increased by >40%. Furthermore, Rohdin et al. (21) showed that DlCO is decreased by 21% at two times normal gravity (2 G) and by 34% at three times normal gravity (3 G) (21). We therefore exposed healthy subjects to both decreases and increases of gravity and studied their exhaled NO. We hypothesized that there would be reduced FeNO levels in microgravity due to increased alveolar uptake of NO, caused by an increased lung diffusing capacity (19, 30, 32). Mass and time constraints typical of space experimentation would not allow other than simple FeNO0.05 (exhalation at 50 ml/s) measurements in space. We therefore performed parallel experiments on ground in normal and increased gravity with more complex equipment. We reasoned that such parallel studies of the effects of gravity on FeNO and related parameters would help in the analysis of the space data. As a corollary of the effects of weightlessness, we hypothesized that not only FeNO but also estimates of CalvNO would be increased in increased gravity due to a reduced Dl. (21) and a resulting slowed alveolar uptake of NO.

METHODS

The experiments were approved by the European Space Agency (ESA) Medical Board (070-4, 092, 117-6, 133-3, and 147-7) and by the Regional Ethical Review Board in Stockholm (2005/507-31 and 2005/658-31).

Microgravity

Subjects. This part of the study was performed onboard the International Space Station (ISS) during the period 2005–2008. Five healthy male ESA astronauts and Russian cosmonauts were studied. All were nonsmokers and had no history of asthma or other inflammatory airway diseases. Age, body mass, and height ranged from 34 to 52 yr, 68 to 78 kg, and 1.72 to 1.82 m, respectively.

Equipment. Portable NO analyzers (NIOX MINO, Aerocrine, Solna, Sweden) (9, 29) were procured from Aerocrine and were certified for space use (Damec Research Aps, Odense, Denmark). Functionally equivalent instruments were used on ground for training and preflight controls. The NIOX MINO analyzer is equipped with an exchangeable NO sensor, and both the analyzer and the sensor have an electronically controlled lifetime with preprogrammed termination dates. The semi-disposable NO sensor is precalibrated at manufacturing to ensure correct measurements within the specified lifetime. Compliance control was repeated by independent experts before use. When the expiry date is reached, the hardware goes into a lock-up state, and no further measurements can be made.

Procedures. Training and preflight measurements (1 G) were performed in Russia or in Germany. Postflight measurements (1 G) were made in Russia or in the US. Astronauts performed at least four
control measurements at one to three occasions before the spaceflight and then approximately every 6 wk during their 23- to 28-wk-long stays onboard the ISS. Here, the gravitational force of the earth is counterbalanced by the centrifugal force resulting from the circular trajectory of ISS (weightlessness or microgravity). After return to earth, they performed daily measurements during the first week after landing.

The subjects performed the FENO maneuver sitting and in duplicates. Since ingested food and beverages rich in nitrite and nitrate have shown to affect exhaled NO (33), the subjects had to refrain from food and beverages rich in nitrite and nitrate 24 h before the tests. They rinsed their mouths with water before each test. The subjects performed the following respiratory maneuver: 1) exhalation to functional residual capacity (FRC), 2) inhalation of NO-free air from the NIOX MINO to close to total lung capacity (TLC), and 3) controlled exhalation into the NIOX MINO during 10 s at a flow rate of 50 ml/s. The expiratory pressure was kept between 1.0 and 2.0 kPa to keep the soft palate closed and hence avoid nasal contamination of the exhaled air. The measurements were fully compiled with the joint ERS/ATS recommendations (1).

Data acquisition and analysis. During the last 3 s of the 10-s-long exhalation of the FENO maneuver, the last 3-s portion of the exhaled air was stored in a buffering unit from where the sample is led to the NO analyzer. The NO concentration result is then presented on the LCD screen of the analyzer. The readings are stored on individual memory cards for later assessment. The NIOX MINO analyzer has a lowest detection limit of 2.3 parts/billion (9).

Hypergravity

Subjects. Ten healthy subjects (seven men and three women) were studied. All were nonsmokers and had no history of asthma or other inflammatory airway diseases. Age, body mass, and height ranged from 23 to 42 yr, 53 to 87 kg, and 1.64 to 1.91 m, respectively.

Equipment. The subjects were studied while sitting in a human centrifuge with a radius of 7.25 m. The backrest of the seat was at a 28° angle to the direction of the gravitational vector. Standard monitoring included audiovisual communication between the gondola of the centrifuge and the test supervisor, assessment of brain perfusion by 133Xe inhalation, an ECG, and a pulse oximeter coupled to an infrared CO2 analyzer (type DPT 6003, PVK, Kirchseeon, Germany), a pressure transducer (type CD12, Validyne, Northridge, CA), and a 3500 series heated pneumotachograph (Pall, East Hills, NY). Gas concentration readings are stored on individual memory cards for later assessment. The subjects rested at 1 G for 3 min, followed by two more sets of four maneuvers. During the 2- and 3-G sessions, subjects rested at 1 G between the sets. The choice of 1.4 G rather than 1 G between the sets of four maneuvers was counterbalanced by the centrifugal G vectors on the differential pressure recordings. One side port in the mouthpiece was connected to a pressure transducer (type DPT 6003, PVK, Kirchseeon, Germany). 

Procedures. Subjects performed the experiments at 1, 2, and 3 G. Thirty minutes before the first centrifuge run, subjects took a tablet to prevent motion sickness (Promethazine 25 mg). Once seated in the centrifuge, the vital capacity (VC) was determined (22). Once VC measurements were complete, the following respiratory maneuver was performed in triplicate for each combination of the four gravity conditions (1 G pre, 2 G, 3 G, and 1 G post) and for the four expired flows: exhalation to residual volume, activation of the rotary valve, inhalation of NO-free air to total lung capacity, and then controlled exhalation at a flow rate of 50 ml/s for 3 min, followed by two more sets of four maneuvers. During a typical session, the subject first sat for 1 min at the target 6 G level and then repeated the above maneuver 12 times; four maneuvers with different expired flows in random order were performed with a 1-min interval in between. Thereafter, the subject rested for 3 min, followed by two more sets of four maneuvers. During the 2- and 3-G sessions, subjects rested at 1 G between the sets. The choice of 1.4 G rather than 1 G between the sets of four maneuvers at 2 and 3 G was made in an effort to avoid the vestibular stimulation caused by accelerating and breaking the centrifuge repeatedly. The subjects rested at 1 G for ~30 min between repeated sessions. The order of the 2- and 3-G sessions was randomized.

Fig. 1. Schematic diagram of equipment for exhaled nitric oxide (NO) analysis in the human centrifuge.
from one maneuver is shown in Fig. 3. Subjects were instructed to abstain from food and beverages rich in nitrite and nitrate 24 h before the tests. Before each test session, they rinsed their mouth with water.

Data acquisition and analysis. All physiological signals were stored continuously, together with G data from an accelerometer, on a digital data acquisition system (Biopac, Goleta, CA) at a rate of 200 Hz. Offline evaluation included the extraction of calibrated NO and CO2 concentration readings as time averages during the middle third of the expiration at 50, 100, 200, or 500 ml/s, i.e., during the middle third of the second part of the exhaled VC for flows of 50, 100, and 200 ml/s and during the middle third of the whole exhalation for flows of 500 ml/s. An individual mean value was computed for each combination of G condition and flow rate. Flow and pressure recordings were checked for deviations from target levels. For each subject and condition, plots were generated for FENO as a function of 1/expired flow. As described by Pietropaoli et al. (17), estimates of CalvNO were derived from the 1/flow \( = 0 \) intercept (theoretical infinite flow). The rate of NO given off from the conductive airways to the expired gas \( J_{awNO} \) was assessed from the slope of FENO vs. 1/flow. Heart rate (HR) was obtained during 20-s periods starting 30 s before each FENO maneuver.

Microgravity and Hypergravity

Probability plots indicated that both microgravity and centrifuge data were normally distributed. Analysis of variance (Statistica 7.1, Statsoft, Tulsa, OK) with a repeated-measures design was used to test for differences between G levels at each flow. In most cases, and when the sphericity assumption held, Tukey’s honestly significant difference post hoc test was performed. In other cases, paired t-tests with Bonferroni correction were used. Results were considered statistically significant at \( P < 0.05 \), and all tests were two sided. Data are given as means ± SD if not stated otherwise.

RESULTS

Microgravity

Figure 4 shows group mean data for preflight, in-flight, and postflight measurements. Data from all five subjects were obtained for the first 14 wk in space. For one subject, the analyzer malfunctioned after 14 wk, and the analyzer was subsequently exchanged when the next subject arrived at ISS. For another subject, the stay on ISS lasted only 23 wk. Preflight FENO was 12.3 ± 4.7 parts/billion (mean ± SD). There was no clear trend for changes of FENO over time during the stays on ISS. Thus all in-flight data for each subject were pooled, and in-flight FENO averaged 6.6 ± 4.4 parts/billion. This was significantly lower than the preflight value (\( P = 0.016 \)). Similarly pooled data for the first week postflight was

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\text{Fig. 3. Fraction of exhaled NO (FNO), partial pressure of expired CO2 (PtCO2), expired flow, and mouthpiece pressure (MPP) during a typical breathing maneuver at 1 G in the human centrifuge. Phase I, tidal breathing with a final exhalation to residual volume; phase II, full inhalation of NO-free air; phase III, exhalation of the first half vital capacity (VC) at 500 ml/s; phase IV, exhalation of the remainder of the VC at 50 ml/s; phase V, normal tidal breathing. NO and CO2 readings were obtained from the middle third of phase IV, where the PtCO2 value was close to but not identical to the end-tidal value.}
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\text{Fig. 4. FNO before, during, and after a 23- to 28-wk-long stay at the International Space Station. Exhalation rate was 50 ml/s. Values are means ± SD before (Pre), every 6 wk during, and after (Post) spaceflight.}
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9.7 ± 2.8 parts/billion, which did not differ significantly from either preflight (P = 0.30) or in-flight (P = 0.28). Figure 4 also shows group mean values of measurements obtained every 6 wk during spaceflight. There was no clear time trend of FE\textsubscript{NO} after landing.

**Hypergravity**

All 10 subjects completed the experiments. Data on levels of NO and CO\textsubscript{2} from the four conditions (1 G pre, 2 G, 3 G, 1 G post) and for exhalation rates of 50, 100, 200, and 500 ml/s (0.02, 0.01, 0.005, and 0.002 s/ml) are presented in Figs. 5 and 6, respectively. Exhaled NO at the flow of 50 ml/s were 16.0 ± 4.3, 19.5 ± 5.1, and 18.6 ± 4.7 parts/billion at 1, 2, and 3 G, respectively. There was a significant effect on FE\textsubscript{NO} of the main factor G (P = 0.001 for all flows). FE\textsubscript{NO} values for a given flow were higher at 2 and 3 G than at 1 G pre but did not differ between 1 G post and 1 G pre (Fig. 5). As also shown in this figure, there was a linear relationship between FE\textsubscript{NO} and the inverse of expired flow in the group mean data. Group mean values of parameters for the regression analysis of FE\textsubscript{NO} on 1/flow values are given in Table 1. CalvNO was significantly higher (65%) at both 2 and 3 G compared with 1 G pre. Estimated CalvNO was 2.3 ± 1.1 parts/billion in 1 G and increased significantly to 3.9 ± 1.4 and 3.8 ± 0.8 parts/billion at 2 and 3 G (P < 0.002). J’awNO tended to be elevated at 2 G compared with 1 G pre.

As with FE\textsubscript{NO}, expired partial pressure of CO\textsubscript{2} (P\textsubscript{aCO\textsubscript{2}}) changed significantly with the main factor G (P < 0.001) but only for the three lowest flows. When this change was analyzed (Fig. 6), it was found that P\textsubscript{aCO\textsubscript{2}} was significantly lower at 3 G compared with 1 G pre at all exhalation flows except 500 ml/s (Fig. 6C). P\textsubscript{aCO\textsubscript{2}} at 2 G did not differ from 1 G pre. P\textsubscript{aCO\textsubscript{2}} tended to be higher at 1 G post than 1 G pre at 50 ml/s.

HR increased from 63 ± 11 beats/min at 1 G before hypergravity to 82 ± 18 and 99 ± 19 beats/min at 2 and 3 G, respectively. Post hypergravity HR was 65 ± 10 beats/min.

**DISCUSSION**

Our principal observation was that FE\textsubscript{NO} is gravity dependent; its values were lowered in microgravity and elevated in seated humans during hypergravity (Fig. 7). According to current models of NO transport, NO originates from conductive airways and alveoli, each source having a different impact on FE\textsubscript{NO}. Thus a change in FE\textsubscript{NO} may come from an alteration of the overall balance between production and blood recapture in the alveolar compartment as well as by a change in the airway production. Indeed, in the airways, the influence of

![Fig. 5. FE\textsubscript{NO} as a function of the inverse expired flow.](http://jap.physiology.org/)

**Table 1. Linear regression of fraction of exhaled NO as a function of the inverse of expiratory flow in normal and increased gravity**

<table>
<thead>
<tr>
<th>Condition</th>
<th>Intercept (flow → 0)</th>
<th>Slope</th>
<th>P value</th>
<th>( r^2 )</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 G pre</td>
<td>2.3 ± 1.1</td>
<td></td>
<td>0.996–1</td>
<td></td>
</tr>
<tr>
<td>2 G</td>
<td>3.9 ± 1.4</td>
<td>0.001</td>
<td>0.983–1</td>
<td></td>
</tr>
<tr>
<td>3 G</td>
<td>3.8 ± 0.8</td>
<td>0.002</td>
<td>0.987–0.999</td>
<td></td>
</tr>
<tr>
<td>1 G post</td>
<td>2.2 ± 0.9</td>
<td>0.996</td>
<td>0.996–1</td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SD of regression parameters obtained separately for each subject (n = 10). \( r^2 \), Ranges of the individual \( r^2 \) values; CalvNO, alveolar NO concentration; JawNO, rate of net transfer of NO from airway wall to lumen; 1 G, normal gravity; 2 G and 3 G, 2 and 3 times normal gravity; pre, before spaceflight; post, after spaceflight. P values are from a post hoc test for differences from 1 G pre (Tukey’s honestly significant difference post hoc test).

HR increased from 63 ± 11 beats/min at 1 G before hypergravity to 82 ± 18 and 99 ± 19 beats/min at 2 and 3 G, respectively. Post hypergravity HR was 65 ± 10 beats/min.
uptake to the blood is likely negligible under the present experimental conditions (10, 20). In our hypergravity experiments, the method proposed by Pietropaoli et al. (17) with multi-flow measurements allowed discrimination between the two effects.

Alveolar NO in Hypergravity

Assuming no change in alveolar production in hypergravity, observed C_{alvNO} changes allow estimating a loss of DLNO of 41% between 1 and 2 G and a quasi-steady state between 2 and 3 G (+2.6%). In a separate study (21), DLCO was seen to decrease by 21% between 1 and 2 G and by 16.6% between 2 and 3 G. Recently, Glenet et al. (7) proposed an interpretation of the DLNO-to-DLCO ratio. They showed that this ratio is proportional to the Dm_{CO}-to-Vc ratio, where Dm_{CO} is the membrane component of DLCO and Vc is the capillary volume. Consequently, pure recruitment or de-recruitment of capillaries will change the contact surface with blood and, thus, will affect Dm_{CO} and Vc the same way, leaving DLNO/DLCO unchanged. Any change of DLNO/DLCO will, thus, reflect a change in alveolo-capillary membrane thickness (affecting Dm_{CO}) and/or a change in the thickness component of Vc (i.e., a change in the perfusion of already recruited capillaries). Assuming no change in the alveolo-capillary membrane thickness in hypergravity, a 41% decrease in DLNO suggests a de-recruitment of capillaries from 1 to 2 G and the decrease (−26%) in DLNO-to-DLCO ratio suggests an increase in the thickness component of Vc, i.e., an over-filling in the still recruited capillaries. This is in line with the increase in tissue volume found by Rohdin et al. (21) and is compatible with an increase of zone I (not perfused) at the apex of the lung with a redirection of capillary blood flow to the dependent zones. From 2 to 3 G, an increase (+23%) in DLNO-to-DLCO ratio would indicate some perfusion decrease, in contradiction with the further increase in tissue volume found in Ref. 21. Regardless, whatever the level of perfusion, it little affects DLNO (NO is quasi perfusion-independent), and the quasi-unchanged DLNO strongly suggests no further capillary de-recruitment between 2 and 3 G. However, the above reasoning assumes constancy of alveolar NO production and alveolo-capillary thickness, which has not necessarily been the case.

Conductive Airway NO in Hypergravity

The present values of J’a_{awNO} at 1 G are in line with corresponding data in the literature (6, 11, 17). There was a trend for J’a_{awNO} to be increased at 2 G compared with 1 G (P = 0.054). Quantitatively, this increase could account for half of
the increase of $\text{FENO}$ (at 50 ml/s) from 1 to 2 G. There are a number of factors, which theoretically can influence $J_{\text{awNO}}$ in hypergravity. Tissue stretch may induce increased NO synthesis (4, 5), and during hypergravity lung tissue is certainly both stretched in some parts and compressed in others (22). Both stretch and compression may cause reductions of peripheral airway caliber. A recent theoretical study (31) showed that such reductions may influence the net NO production as measured at the mouth (and thus $\text{FENO}$). Particularly, a reduction in the cross-sectional area of acinar airways may cause an increase of the observed net NO production by impairing the peripheral effect of molecular diffusion.

Effects of Stress in Hypergravity

The increased HR levels in hypergravity indicated that subjects were under physiological and probably also psychological stress in hypergravity with arterial hypotension in the head and neck (13) and with associated increases of plasma catecholamine levels (24, 25). Adrenaline infusions have been shown to increase exhaled NO in a rabbit model (2). Therefore, had stress been a major factor for the $\text{FENO}$ increase, higher values should have been found at 3 G than at 2 G (25), and that was not the case. In contrast, Persoons et al. (15) showed that the NO production from alveolar macrophages was suppressed by stress in a rat model. If also true for humans, such a mechanism may have accounted for the lack of increase of exhaled NO from 2 to 3 G.

Exhaled CO₂ in Hypergravity

CO₂ has been shown to inhibit NO formation (26); therefore, such an interaction should also be considered in the present study. When 1 G pre and 2 G are compared, CO₂ levels were practically identical, strongly suggesting that CO₂/NO interactions is not a major mechanism for the increased $\text{FENO}$ at 2 G. The lowering of CO₂ at 3 G compared with 1 and 2 G would rather result in further elevation of $\text{FENO}$ compared with 2 G, and since that was not the case, it is not likely that CO₂/NO interactions play an important role in the present hypergravity experiments.

The CO₂ values observed in the present study can be regarded as a function of different degrees of relative hyperventilation. Thus the $\text{FENO}$ maneuver with a relatively rapid deep inhalation and then a relatively rapid full exhalation at 500 ml/s, naturally results in an instantaneous lowering of alveolar and expired CO₂ compared with normal tidal breathing. Since expired flow is decreased to 200, 100, and or 50 ml/s, during the second half of the VC expired CO₂ has time to rise toward a eucapnic level (Fig. 6). This was true for all conditions except 3 G, where sustained hyperventilation was underway before the $\text{FENO}$ maneuvers started, leading to generally lower CO₂ values during the maneuvers. The much lower CO₂ values at 3 G are in agreement with a previously described G-induced hyperventilation at 3 G (23).

Lung Deformation and Ventilation/Perfusion Distribution in Hypergravity

In addition to the effect on the blood distribution in the lung, gravity and even more so hypergravity will lead to deformation of the lung tissue so that apical lung units in the seated human are stretched, basal units are compressed, and vascular hydrostatic gradients within the pulmonary circulation are increased (18). These alteration lead to regional heterogeneities of ventilation and perfusion. Recent studies (28, 31) showed that large heterogeneities of ventilation have only little effect on $\text{FENO}$. At the same time, there is a gravity-induced caudal displacement of blood in the systemic circulation resulting in decreases of cardiac preload, stroke volume, and cardiac output (21, 23). Because the perfusion is more affected by gravity than ventilation, this will lead to an impaired matching between ventilation and perfusion in the lungs, manifesting itself as an increased alveolar-to-arterial oxygen partial pressure difference, arterial desaturation (23), and reduced DLCO (21). Lung expansion and emptying during a single-breath maneuver becomes increasingly sequential at 2 and 3 G, so that basal well perfused lung units are subject to airway closure toward the end of expiration (22). Thus the air at a given moment of an expiration may not be representative for the average alveolar gas composition. The impact of these phenomena for $\text{FENO}$ would be an increased relative contribution of nonperfused apex zones, even without further capillary de-recruitment between 2 and 3 G. Since the closing volume continuously increases from 1 to 3 G, it would tend to similarly increase the alveolar part of $\text{FENO}$ with gravity. Such an increase, in fact, is not observed, at least not between 2 and 3 G. One reason can be the counter-pressure applied at the mouth during the $\text{FENO}$ maneuver to avoid nasal NO contamination. This pressure would tend to partially prevent airway closure otherwise seen during VC exhalations in hypergravity (22). It cannot be excluded, however, that gravity-induced sequential emptying and airway closure could have contributed to elevate $\text{FENO}$ in hypergravity.

Exhaled NO in Microgravity

Each of the five subjects had lower $\text{FENO}$ values during the first measurement inflight than before the flight, and decreases ranged 21–76% from the preflight value. In this specific experimental setup, only $\text{FENO}$ was measured; therefore, alveolar and airway contributions may not be estimated separately. However, the decrease averaged more than 5 parts/billion, which is a numerically larger value than what is a normal value for $C_{a\text{awNO}}$. Thus a reduction of $C_{a\text{awNO}}$ could have contributed to, but could not explain the full extent of, the $\text{FENO}$ reduction in microgravity. Again, assuming unchanged alveolar NO production in sustained microgravity, the previously documented improvement of the membrane component of DLCO by 28% in microgravity (19) could only have resulted in a proportional reduction of $C_{a\text{awNO}}$.

With only $\text{FENO}_{0.05}$ data at hand, we can just speculate about additional explanations for the fall in $\text{FENO}$. Tissue relaxation may lead to peripheral airway lumen increase improving peripheral molecular diffusion and decreasing the net NO production as measured at the mouth. In addition, decreased mechanical stress of lung tissue might have resulted in less induction of NO formation in the lung tissue (4). Also, $\text{FENO}$ in microgravity might represent a much better estimate of the NO concentrations in the lungs because a more representative sample for all lung units is obtained during exhalation. These potential effects of tissue relaxation as well as the DLCO increase would be “mirror effects” compared with hypergravity. Finally, one should consider the possibility of systematic
differences between the analyzer units used in space and on
ground. However, the analyzer units and the sensors on ISS
were exchanged at the required intervals and they were com-
pliance controlled before launch to ISS, which renders such
an explanation less likely.

Conclusions and Perspectives

We have found that exhaled NO is lower in microgravity and
higher in hypergravity than in normal gravity. Exhaled NO
changes were in the direction that would be expected from
previously determined gravity-induced alterations in lung dif-
fusing capacity. Thus these data are consistent with the hy-
pothesis that alveolar NO uptake is increased in microgravity
and decreased in hypergravity. However, changes in lung
diffusing capacity can explain neither the full extent of the
lowered exhaled NO in microgravity nor the leveling off of
exhaled NO from 2 to 3 G, so further studies are required to
find additional mechanisms.

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DISCLOSURES

L. E. Gustafsson is an inventor of patents on exhaled NO. L. E. Gustafsson
is a minority shareholder (<0.5% of total shares) in Aerocrine, which markets
instruments for exhaled NO measurements for monitoring of airway inflam-
mation. T. Hemmingsson is a former employee of Aerocrine.

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