

Sildenafil inhibits the altitude-induced hypoxemia and pulmonary hypertension.

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With grant from Pfizer S.A. France

Running head: Sildenafil, pulmonary hypertension, hypoxia

Descriptor number: 148

Word count for manuscript: 3816

This article has an online data supplement, which is accessible from this issue’s table of content
online at www.atsjournals.org.

Abstract

Exposure to high altitude induces pulmonary hypertension that may lead to life threatening conditions. In a randomized double-blind placebo-controlled study, we examined the effects of oral sildenafil on altitude-induced pulmonary hypertension and gas exchange in normal subjects. Twelve subjects (sildenafil, SIL n=6; placebo, PLA n=6) were exposed for 6 days at 4,350m. Treatment (3 x 40mg / day) was started 6 to 8 hours after arrival from sea level to high altitude and maintained for 6 days. Systolic pulmonary artery pressure (echocardiography) increased at high altitude before treatment (+29% vs sea level, $p<0.01$), then normalized in SIL (-6% vs sea level, ns) and remained elevated in PLA (+ 21% vs sea level, $p<0.05$). Pulmonary acceleration time decreased by 27% in PLA vs 6% in SIL ($p<0.01$). Cardiac output and systemic blood pressures increased at high altitude then decreased similarly in both groups. PaO₂ was higher and alveolar-arterial difference in O₂ lower in SIL than in PLA at rest and exercise ($p<0.05$). The altitude-induced decrease in VO₂max was smaller in SIL than in PLA ($p<0.05$). Sildenafil protects against the development of altitude-induced pulmonary hypertension and improves gas exchange, thus limiting the altitude-induced hypoxemia and decrease in exercise performance.

Abstract word count: 194

Key words: hypoxia, exercise, cardiac output, gas exchange

Exposure to high altitude (HA) leads to hypoxemia which induces several physiological or pathophysiological responses in normal humans. Among those, the hypoxic pulmonary vasoconstriction (HPV) leads to an increase in pulmonary artery pressure (PAP) which may have adverse consequences. High PAP has been recognized to be one of the main causing factors of High Altitude Pulmonary Edema (HAPE), a serious acute condition which has a mortality rate of 44% in untreated patients (1,2). Moreover, ventilation-perfusion mismatch has been correlated to increasing PAP at high altitude, probably by the development of interstitial and perivascular edema, aggravating the hypoxemia (3). In the early phase of exposure to HA, signs of Acute Mountain Sickness (AMS) may develop and have been shown to be worsened by aggravating hypoxemia, although the precise mechanisms of AMS have not been elucidated yet (4). Altitude hypoxia induces a dramatic decrease in physical aerobic performance, as assessed for example by the maximal O₂ consumption (VO₂max) (4). Several steps in the oxygen transport from the ambient air to the cell can be responsible for the limitation in aerobic performance, among which O₂ transfer within the lungs, cardiac output, and tissue diffusion of O₂ may play an important role (5). Altogether, acute altitude-induced hypoxemia leads to an adverse condition where, at least, overall well being is altered by AMS and reduction in performance, and life is possibly threatened by the development of high PAP and HAPE. Thus, any treatment or condition that limits the increase in PAP and reduces the altitude-induced hypoxemia may be beneficial for humans acutely exposed to HA.

The treatment currently recommended for HAPE is rapid reoxygenation combined with a calcium-channel blocker (6,7). However, the partial efficacy of this treatment and its systemic adverse effects (hypotension) limit its use. Inhalation of nitric oxide (NO) has also been used, and has demonstrated its efficacy in this condition, but its use in the field is difficult (8).

L-Arginine supplementation has also been found to improve gas exchange at high altitude, further suggesting that the NOS-NO system is involved in the hemodynamic changes in the

lungs (9). Recently, sildenafil, a selective inhibitor of type-5 phosphodiesterase has been shown to lower PAP and used successfully in the treatment of severe primary or secondary pulmonary hypertension (10-18). In most cases, sildenafil was not given as a unique treatment but associated with either inhaled NO, IV epoprostenol or inhaled iloprost. Only two studies have evaluated the effect of oral sildenafil (50 to 100 mg, single dose) in normal subjects exposed to acute hypoxia, in a randomised double-blind study (13, 14): hypoxia-induced increase in PAP was almost abolished with sildenafil and no important effect on systemic circulation was observed. However, deleterious effects of high altitude occur after several hours of exposure and, to date, no double-blind controlled study has evaluated the effect of sildenafil on the adverse effects of prolonged altitude exposure in normal humans. Sildenafil has also been reported to increase arterial PO_2 (17) and improve physical performance (14,15,18) in various cases of severe pulmonary hypertension, but no study has explored the effect of a several day treatment by sildenafil on these variables in normal subjects exposed to altitude conditions. The objective of the present study was to explore the effects of oral sildenafil in normal subjects exposed for 6 days to an altitude of 4,350m, in a randomized double-blind placebo-controlled manner. The hypothesis was that sildenafil would reduce the hypoxia-induced increase in PAP, ameliorate the pulmonary hemodynamics and gas exchange conditions, thus increasing the arterial PO_2 , alleviating the clinical symptoms and limiting the reduction in aerobic performance. Some of the results of this study have been previously reported in the form of abstracts (19,20).

METHODS

Subjects

Twelve male normal subjects (aged 29 ± 6 yrs) participated in the study. Anthropometric characteristics were: height 181 ± 6 cm, body weight 79 ± 11 kg. They were healthy,

unacclimatized to altitude, moderately trained subjects, with no particular medical history, and no previous episode of severe altitude sickness. They gave their informed consent to participate in the study that was approved by the Ethics Committee of Necker Hospital, Paris.

Procedure.

The following evaluations were performed during the 11-day experimental period from D-3 to D8 (Figure 1). Sea-level PRE measurements were performed in Bobigny (60m altitude), then subjects were transported to Chamonix (1035m) for one day and to Observatoire Vallot (4,350m) by helicopter (21). Additional details on the methods and equipment used are provided in an online data supplement.

Clinical questionnaire and systemic hemodynamic parameters

A daily questionnaire was filled three times a day (08:00-09:00, 13:00-14:00, 18:00-19:00), including the Lake Louise consensus questionnaire to evaluate the symptoms of AMS (22), and some specific questions related to the possible adverse effects of sildenafil (headache, muscle pain, dyspepsia, flushing). A score of sleep disturbances was evaluated in the morning (from 0, normal sleep to 3, very poor sleep). Ataxia and dyspnea were also evaluated (from 0 to 3) according to the Lake Louise consensus (22). At the same moment, heart rate and O₂ saturation were evaluated by pulse oximetry [Ohmeda Biox 3740, USA], and systemic systolic and diastolic blood pressure were evaluated in a supine position by sphygmomanometry.

Echocardiography

Subjects were examined by two observers on left decubitus or supine position, using a portable ultrasound system equipped with a 2.5 MHz probe (Cypress, Acuson/Siemens, Germany).

Complete 2-dimensional, TM-echography and Doppler parameters for left cardiac function were recorded following classical procedures. Systolic pulmonary arterial pressure (sPAP) was calculated from the tricuspid gradient. The acceleration time of the pulmonary flow (AcT) was taken as an index of pulmonary vascular resistance (23). At each examination, all parameters

were measured at least 3 consecutive times and the subjects were examined 3 times on baseline on days D-3, D-2, D-1, 5 times during the altitude exposure on days D1, D2, D3, D5, D6 and at recovery on day D8 (Sea level post). Base line normoxic values (Sea level pre) were taken as the mean of values obtained at D-2 and D-1. Values at D2 and D3 were pooled and considered as initial values after 1 – 2 days of treatment, values at D5 and D6 were pooled and considered as final values after 4 – 5 days of treatment.

Maximal exercise test

Maximal aerobic performance was evaluated through a step-by-step progressive exercise test performed on a bicycle ergometer [Monark, Sweden] until exhaustion, at D-3, D2, D5 and D8. ECG was monitored continuously (Life Scope 6, Nihon Kohden, Japan) and arterial O₂ saturation was obtained via ear oximetry (Ohmeda Biox 3740, USA) on an ear lobe previously vasodilated by a capsaicin cream. PaO₂, PaCO₂ and pH_a were measured by means of a blood gas apparatus (Model 220, Bayer diagnosis, Germany) from an arterialized blood sample. Cardiac output and an intra-thoracic fluid (ITF) index were measured continuously by trans-thoracic impedencemetry (Physioflow PF-05 lab1, Manatec, France), from electrodes placed on the base of the neck and on the medial line under the xyphoid (24,25).

Color vision test

Modifications in color vision in the red/green axis have been observed at high altitude and correlated with severity of AMS (26). Transient, fully reversible, impairment of color discrimination has also been noticed as side effects of treatment with sildenafil (27). Therefore, color vision was evaluated in the present study, using the Lanthony 15-Hue Desaturated Test at D-1, D1, D3, D5 and D8. A color confusion index (CCI) was calculated. The greater the number and importance of mistakes, the higher the CCI value (28).

cGMP and sildenafil

Blood sampling was performed at rest from an antecubital vein, at D-1, D1, D3 and D6 to measure cGMP, sildenafil concentration and hematocrit, one to two hours after oral administration. cGMP was measured by radioimmunoassay (cGMP RIA kit, Immunotech, Marseille, France). Sildenafil + desmethylsildenafil concentration was measured by a liquid chromatography-tandem mass spectrometry method (29). Hematocrit was measured immediately by means of a microcentrifuge (Sigma 112, Germany).

Treatment

Subjects were randomly assigned to a placebo (PLA, n=6) or sildenafil (SIL, n=6) treated group. Treatment (40 mg) started on D1 at 4,350m at 20:00, 6 to 8 hours after arrival at Observatoire Vallot. Then treatment was taken (40 mg orally) three times a day (08:00, 14:00, 20:00) from D2 to D6. Sildenafil and placebo were provided by Pfizer.

Statistics

Values are presented as mean \pm standard deviation. A Mann-Whitney *U*-test was performed to compare the two groups and analyse the effects of treatment in each condition (symbol #).

Values obtained at high altitude after treatment (from D2 to D6) have also been compacted and analyzed with a Mann-Whitney *U*-test to evaluate the overall effect of treatment at high altitude (symbol +). A Wilcoxon paired test was used between each condition and sea-level, to evaluate the effect of altitude exposure on each group (symbol *). Values of the two groups were pooled at D1 to evaluate, by a Wilcoxon paired test, the overall effect of hypoxia before treatment (symbol §). A *P* value < 0.05 was considered as significant.

RESULTS

Tolerance

Exposure to high altitude and treatment were well tolerated by all subjects. However, subject # 4 (placebo) showed some low values of SaO₂ (under 60%) at various occasions at high altitude,

without any abnormal clinical symptoms, except moderate headache and fatigue. His cardiac and lung auscultation and neurological examination were strictly normal. He was thus maintained in the study and given inhaled O₂ (1 l/min) for 4 hours during sleep from D4 to D5, at distance from any test involved in the study. It is noteworthy that the significance of all results presented is not modified if subject #4 is excluded from the study. Frequency of expected adverse events was not different between the two groups: 1 SIL and 2 PLA subjects suffered from dyspepsia; 3 SIL and 1 PLA subjects showed flushing of the face; muscle pain was noticed by 2 SIL and 3 PLA subjects. All these complaints were occasional. Sildenafil treatment had no effect on color vision. Acute exposure to high altitude (D1) was associated with a slight alteration in color vision score in both groups (D1 vs D-1, $p < 0.05$), then values returned to normal levels (Table 1).

Clinical evaluation (Table 1)

Subjects suffered from AMS until D4, then the Lake Louise score was not significantly different from normoxic base line. Lake Louise score tended to be lower in SIL group at D1 ($P = 0.054$) before treatment and thereafter at D5 and D6 but the difference did not reach significance.

Among clinical symptoms, gastrointestinal symptoms and dizziness were similar in the two groups (results not shown). Headache, which is both a symptom of AMS and a possible adverse effect of sildenafil was not significantly modified by the treatment. Fatigue score appeared slightly higher in PLA than in SIL, even after the return to normoxia, but the differences did not reach significance. Sleep was significantly altered during the first two nights at high altitude, with no effect of sildenafil. Only scarce cases of ataxia or dyspnea scores different from zero were noticed, with no effect of sildenafil (results not shown).

Systemic hemodynamic parameters (Figure 2)

Mean daily heart rate (HR) increased in both groups at high altitude (Figure 2A). HR in SIL was significantly lower than in PLA from D2 to D6 ($p < 0.01$). Systolic and diastolic systemic arterial pressure increased transiently from D1 to D4 but was not modified by the treatment (Figure 2B).

SaO₂ decreased at high altitude and was clearly higher in SIL than in PLA from D2 to D6 ($p < 0.001$, Figure 2C).

Echocardiography (Figure 3 and Table 2)

As expected, sPAP increased with acute exposure to high altitude (D1) before treatment. After one to two days of treatment (D2-D3), sPAP was significantly lower in SIL than in PLA ($P = 0.025$). After four to five days of treatment (D5-D6), sPAP was lower in SIL than in PLA ($p < 0.05$). At D5-D6, when compared to sea-level, sPAP increased by 21% in PLA ($P = 0.03$) and decreased by 6% in SIL (n.s.). Pulmonary acceleration time (AcT) decreased in both groups at D1 (before treatment) and returned to basal normoxic values in SIL but stayed low in PLA at high altitude ($P = 0.001$, PLA vs SIL). All other echocardiographic parameters, especially those exploring left ventricular function, were strictly normal and similar in the two groups (Table 2). The diameter of the left ventricle slightly decreased in diastole and systole, leading to a transient increase in shortening fraction. Left atrium diameter and mitral E/A wave ratio progressively decreased with exposure to high altitude. Cardiac output measured by Doppler, increased from sea-level at D1 and D2-D3 in both groups ($p < 0.05$), then returned to basal values, with no significant effect of treatment.

Aerobic performance and gas exchange (Table 3, figure 2)

As expected, VO₂max decreased at high altitude (D2) and slightly (n.s.) increased with acclimatization (from D2 to D5). The altitude-induced mean decrement in VO₂max was smaller in SIL (- 29% at D2, -25% at D5) than in PLA (- 39% at D2, -35% at D5) ($p < 0.01$, SIL vs PLA, Figure 2D). At high altitude, PaO₂ was higher in SIL than in PLA, either at rest ($p < 0.05$ at D5-D6) or at exercise ($p < 0.01$ at D5-D6). Alveolar-arterial difference in PO₂ at rest and at exercise decreased in both groups at high altitude, but the decrease was lower in PLA than in SIL ($p < 0.001$ at rest, $p < 0.05$ at exercise). On return to sea level, at rest, PaO₂ was lower and alveolar-arterial difference in PO₂ higher than in basal level values. As expected, PaCO₂

decreased and pHa increased at high altitude (hyperventilation-induced hypocapnia and alkalosis), no difference was found between the two groups. Cardiac output at rest, measured by transthoracic impedencemetry transiently increased at high altitude ($p < 0.05$) and was similar in the two groups. At ventilatory threshold, cardiac output was modified, neither by altitude nor by treatment. Heart rate at ventilatory threshold and at maximal exercise decreased at high altitude in both groups. After the return to sea level, maximal heart rate remained lower than before the hypoxic exposure. The intra-thoracic fluid index (ITF) increased in both groups at high altitude (D2), then decreased only in SIL and stayed elevated in PLA ($p < 0.05$ SIL vs PLA).

Acclimatization to high altitude (Table 3)

The physiological parameters, characteristics of acclimatization to high altitude (PaCO₂, pHa, heart rate), were modified from D2 to D5 as expected. No difference was found between SIL and PLA. PaCO₂ at rest and at the ventilatory threshold decreased from D2 to D5 (SIL: $p < 0.03$, PLA: ns), pHa at the ventilatory threshold increased (SIL: $p < 0.03$, PLA: ns). Heart rate at the ventilatory threshold and at maximal exercise decreased from D2 to D5 ($p < 0.05$ for PLA and SIL). Sildenafil treatment had no effect on these parameters.

Serum cGMP, serum sildenafil and hematocrit

Serum cGMP increased from sea level at D1 before treatment, then by 165 % ($p < 0.05$) and 42 % (n.s.) at D6 in SIL and PLA respectively ($p < 0.05$ SIL vs PLA) [Figure 4]. Serum sildenafil + desmethylsildenafil concentration was below detectable limit at N and D1 and increased in the SIL group to 10.3 ± 6.7 ng/ml and to 254.0 ± 146.3 ng/ml at D2 (cumulative dose of sildenafil ingested, 36 hours after first pill: 160 mg) and D6 (cumulative dose of sildenafil ingested, 108 hours after first pill: 520 mg) respectively. Ht was not modified by the treatment. In the whole group, mean Ht increased from 43.2 ± 2.6 % at sea level (D-1) to 46.1 ± 1.8 at D3 ($p < 0.001$), 45.3 ± 1.3 at D6 ($p < 0.01$) and was still high at D8 (45.9 ± 2.0 , $p < 0.001$).

DISCUSSION

This is the first double blind controlled study evidencing the beneficial effect of oral sildenafil (3 x 40 mg / day for 6 days) in normal subjects exposed to prolonged high altitude hypoxia. High-altitude hypoxia induces a specific pulmonary vasoconstriction and an acute sympathetic activation. Hence, after acute exposure to 4,350 m. all the subjects exhibited a decrease in SaO₂ and the expected changes in cardiac hemodynamics with an increase in heart rate, cardiac output, systemic and pulmonary pressures. Then, acclimatization occurred with a decrease in heart rate and an increase in ventilation.

The main observed effect of sildenafil was a suppression of the hypoxia-induced increase in pulmonary artery pressure, associated with an increase in blood oxygenation. No adverse effect, such as systemic hypotension or alteration in color vision, was noticed. Only minor adverse effects (muscle pain, dyspepsia) have been recorded. Lastly, sildenafil hampered the hypoxia-induced decrease in exercise performance and did not interfere with acclimatization.

The effect of sildenafil upon pulmonary artery pressure, already observed in humans suffering from primary or secondary pulmonary hypertension (10-13, 15-18) has thus been found in normal subjects exposed to altitude-induced hypoxia. No adjunct treatment such as NO or epoprostenol has been used in the present study suggesting that the inhibition of PDE5 by itself can have a vasodilatory effect on pulmonary circulation, probably by increasing the availability of cGMP within the pulmonary vasculature (13). In the present study, plasma level of cGMP increased with sildenafil and was associated with a decrease in pulmonary artery pressure without significant decrease in cardiac output. This is in accordance with a direct effect of cGMP on pulmonary vascular smooth muscle cell rather than an effect on cardiac function. Increase in PAP at high altitude was also confirmed by the decrease in pulmonary acceleration time, as shown at D1, which has been considered as an index of pulmonary hypertension (23). Sildenafil restored this index to basal values as soon as in D2-D3, whereas it stayed low in PLA

during the whole stay at high altitude. Although present at D2-D3, the overall hemodynamic effects of sildenafil on pulmonary circulation were more marked on D5-D6 when the plasma concentration of the drug was 25 fold increased.

All parameters of LV systolic function (Table 3) were not modified by the treatment, confirming that sildenafil has no effect on cardiac contractility and LV afterload. Furthermore sildenafil has been shown to have no or modest effects on systemic vasculature after a single dose of less than 100 mg (30). In the present study, sildenafil had no significant effect on systemic circulation since systemic arterial pressure and cardiac output transiently raised then returned to baseline values, similarly in the two groups. Lastly, despite a lower heart rate in the treated group, cardiac output did not significantly change suggesting a lack of negative effect of sildenafil on cardiac inotropism. The lowering effect on heart rate may be indirect, by increasing SaO_2 or direct through a negative chronotropic effect via increased cGMP (31). The decrease in E/A ratio, an index of LV relaxation, observed in the two groups with exposure to high altitude was probably due to a decrease in LV filling as shown by the associated decrease in LA and systolic and diastolic LV diameters. This phenomenon is probably linked to a lower venous return due to an altitude-induced decrease in plasma volume previously observed in the same conditions (32). In the present study, plasma volume was not measured but indirect evidence can be drawn from the acute increase in hematocrit from 43 to 46%.

The increase in PaO_2 and SaO_2 observed at rest and exercise, associated with a lower alveolar-arterial O_2 difference and an unchanged PaCO_2 , is particularly interesting since it evidences a better oxygen transfer within the lungs, probably due to a better ventilation/perfusion adequacy or a decrease in lung diffusion impairment. Hypoxia-induced increase in PAP has been shown to be one of the main mechanisms responsible for the development of the alveolar edema in HAPE (1, 2). Even in normal subjects, ventilation-perfusion mismatch has been shown to increase at high altitude with increasing PAP,

either by a non-uniform pulmonary vasoconstriction or by increasing the interstitial and perivascular edema (3). By lowering PAP, sildenafil could reduce the pulmonary capillary leak and limit the development of interstitial edema. The observed decrease in intra-pulmonary fluid index with sildenafil is in favour of this hypothesis. This amelioration of blood oxygenation, in turn, can have a beneficial effect on pulmonary vasculature, thus enhancing the effect of the drug. Upon return to sea level, all parameters tended to return to sea level basal values.

However, in both groups at rest, PaO₂ was lower and PA-PaO₂ higher than in basal conditions. This may not be linked to hypoventilation since PCO₂ was not elevated. It could be related to a slight persistent interstitial edema or ventilation-perfusion mismatch after altitude exposure.

No significant adverse event was evidenced and the treatment was well tolerated. The dose used (120 mg /day) is comparable to what is now commonly used (100 – 150 mg) in prolonged treatment of pulmonary hypertension (15,18). No clear effect has been shown on the clinical signs of AMS, even if a tendency to lower the Lake Louise score was shown after four days of altitude exposure. However, headache being a possible adverse effect of sildenafil, its probable increase in treated subjects may have jeopardize a possible beneficial effect on overall AMS score due to a better blood oxygenation. The indication of sildenafil in the treatment of HAPE has not been addressed in the present study since none of our subjects suffered from this severe condition. However, the beneficial effect on PAP strongly suggests that this drug could be highly effective in this condition, without adverse systemic effect, contrarily to the classically proposed calcium blockers (6,7).

The beneficial effects of sildenafil on pulmonary circulation and gas exchange have been sufficient to limit the altitude-induced decrease in maximal aerobic performance. To our knowledge, no pharmacological treatment has been previously shown to reduce this debilitating effect of prolonged high altitude exposure. Sildenafil treatment did not interfere with the usual physiological characteristics of acclimatization to high altitude. The decrease in

PaCO₂ and increase in pHa indicating a process of ventilatory acclimatization and the decrease in maximal heart rate, attributable to a progressive desensitization of cardiac beta-receptors (33), observed in the present study from day 2 to day 5, were not modified by the treatment. Similarly, an acute altitude-induced decrease in plasma volume probably accounts for the slight increase in hematocrit, without any significant effect of treatment.

In conclusion, sildenafil, by its vasodilating effect on pulmonary circulation 1) suppresses the altitude-induced pulmonary hypertension, 2) ameliorates pulmonary hemodynamics and gas exchange, thus limiting the altitude-induced hypoxemia and favouring cardiovascular adaptation to exercise, 3) does not alter the normal physiological processes of acclimatization. Further studies will determine if sildenafil could replace calcium blockers in the treatment of high altitude pulmonary edema.

Acknowledgments

We are grateful to Eric Jaudinot and Véronique Chauveau (Pfizer France), Ghazwan Butrous (Pfizer Sandwich) for supporting the project and providing sildenafil citrate, and Richard Hucker (Pfizer Sandwich) for the dosage of sildenafil. We thank all the volunteers for their participation in this study at Observatoire Vallot, the Laboratoire de Glaciologie et Géophysique de l'Environnement (CNRS) for the use of the facilities, Acuson/Siemens for providing the echocardiographic device (Cypress) and Bayer Diagnostics for providing the blood gases analyser.

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Legend of figures.

Figure 1. Schematic diagram of the procedure. VO₂max: maximal exercise test; Echo: echocardiographic examination; B.S.: blood sampling; C.V.: color vision test. The whole study lasted 12 days, from D-3 to D8.

Figure 2. Systemic hemodynamic parameters and exercise performance. *, **, ***: p<0.05, p<0.01, p<0.001 vs sea level pre; #, ##: p<0.05, p<0.01 SIL vs PLA. §, §§: p<0.05, p<0.01 D1 vs sea level pre for the whole group. +, ++: p<0.05, p<0.01 SIL vs PLA for pooled high altitude with treatment values.

Figure 3. Echocardiographic evaluation of pulmonary hemodynamics. PAP : pulmonary artery pressure. *, **, ***: p<0.05, p<0.01, p<0.001 vs sea level pre; #, ##: p<0.05, p<0.01 SIL vs PLA. §, §§: p<0.05, p<0.01 D1 vs sea level pre for the whole group. +, ++: p<0.05, p<0.01 SIL vs PLA for pooled high altitude with treatment values.

Figure 4. Serum cyclic GMP. *: p<0.05 vs sea level pre; #: p<0.05 SIL vs PLA. §§: p<0.05 D1 vs sea level pre for the whole group. +: p<0.05 SIL vs PLA for pooled high altitude with treatment values.

TABLE 1. Clinical symptoms and color vision.

		Sea level pre	D1	D2	D3	D5	Sea level post
Lake Louise score, a.u.	PLA	0.1 ± 0.1	2.8 ± 1.6 *	1.8 ± 1.4 *	1.3 ± 0.8 *	0.9 ± 1.1	0.3 ± 0.4
	SIL	0.1 ± 0.1	1.0 ± 0.7 *, §§	1.9 ± 1.3 *	1.3 ± 0.5 *	0.3 ± 0.3	0.0 ± 0.0
Headache score, a.u.	PLA	0.0 ± 0.1	1.2 ± 0.6 *	0.8 ± 0.5 *	0.7 ± 0.4 *	0.3 ± 0.4	0.0 ± 0.0
	SIL	0.0 ± 0.1	0.7 ± 0.5 *, §§	1.1 ± 0.5 *	0.9 ± 0.2 *	0.2 ± 0.3	0.0 ± 0.0
Fatigue score, a.u.	PLA	0.1 ± 0.1	0.6 ± 0.5 *	0.6 ± 0.7 *	0.6 ± 0.5 *	0.4 ± 0.6	0.3 ± 0.4
	SIL	0.0 ± 0.1	0.2 ± 0.3 §	0.4 ± 0.5	0.3 ± 0.3	0.1 ± 0.1	0.0 ± 0.0
Sleep score, a.u.	PLA	0.6 ± 0.5	2.0 ± 1.1 *	1.0 ± 0.9	0.7 ± 1.0	0.4 ± 0.5	0.5 ± 0.8
	SIL	0.3 ± 0.4	1.6 ± 0.5 *, §	0.9 ± 0.9	0.5 ± 0.8	0.6 ± 1.2	0.3 ± 0.8
Color confusion index, a.u.	PLA	1.21 ± 0.18	1.51 ± 0.39 *	/	1.26 ± 0.27	1.26 ± 0.24	1.27 ± 0.24
	SIL	1.28 ± 0.32	1.51 ± 0.39 *	/	1.31 ± 0.13	1.34 ± 0.21	1.22 ± 0.46

Sea level pre: basal normoxic condition, D1 to D5: first to fifth day at 4,350m, Sea-level post: return to normoxic conditions. * : p<0.05 vs Sea level pre. a.u.: arbitrary units. §, §§: p<0.05, p<0.01 D1 vs Sea level pre for the whole group

TABLE 2. Echocardiographic parameters.

		Sea level pre	D1	D2-D3	D5-D6	Sea level post
left ventricle diameter in diastole, mm	PLA	52.2 ± 3.8	52.8 ± 5.7	49.1 ± 4.0 *	49.4 ± 3.8 *	51.4 ± 4.2
	SIL	50.9 ± 2.4	48.1 ± 1.5 *,§§	47.4 ± 2.1*	48.9 ± 1.6*	50.5 ± 0.8
left ventricle diameter in systole, mm	PLA	30.7 ± 4.8	32.6 ± 3.8	27.5 ± 3.8 *	28.2 ± 2.5	30.8 ± 3.9
	SIL	29.6 ± 1.9	28.9 ± 1.8	26.3 ± 2.8 *	28.4 ± 2.4	30.7 ± 2.2
shortening fraction, %	PLA	40.6 ± 4.6	39.0 ± 3.6	44.0 ± 4.2 *	43.1 ± 3.1	40.0 ± 5.2
	SIL	40.5 ± 3.6	40.1 ± 4.3	46.0 ± 5.2 *	42.3 ± 3.9	39.8 ± 4.6
mitral E/A wave ratio	PLA	2.0 ± 0.4	1.4 ± 0.3 *,§	1.4 ± 0.2 *	1.4 ± 0.1 *	1.8 ± 0.4
	SIL	1.8 ± 0.3	1.6 ± 0.3	1.3 ± 0.2 *	1.4 ± 0.2 *	1.5 ± 0.4
left atrium diameter, mm	PLA	36.1 ± 3.6	33.2 ± 3.4 *,§	33.4 ± 3.4 *	32.4 ± 5.0 *	32.2 ± 4.9
	SIL	36.7 ± 3.4	35.0 ± 3.8	31.4 ± 5.2 *	32.3 ± 3.9 *	30.1 ± 4.5 *
cardiac output, l/min	PLA	5.09 ± 0.55	6.45 ± 1.30	6.86 ± 1.97 *	6.16 ± 1.38	5.10 ± 0.96
	SIL	4.75 ± 0.59	6.15 ± 0.77 §	6.05 ± 0.69	5.19 ± 0.50	4.42 ± 0.65

Sea level pre: basal normoxic condition, D1 to D6: first to sixth day at 4,350m, Sea-level post: return to normoxic conditions. * : p<0.05 vs sea level;

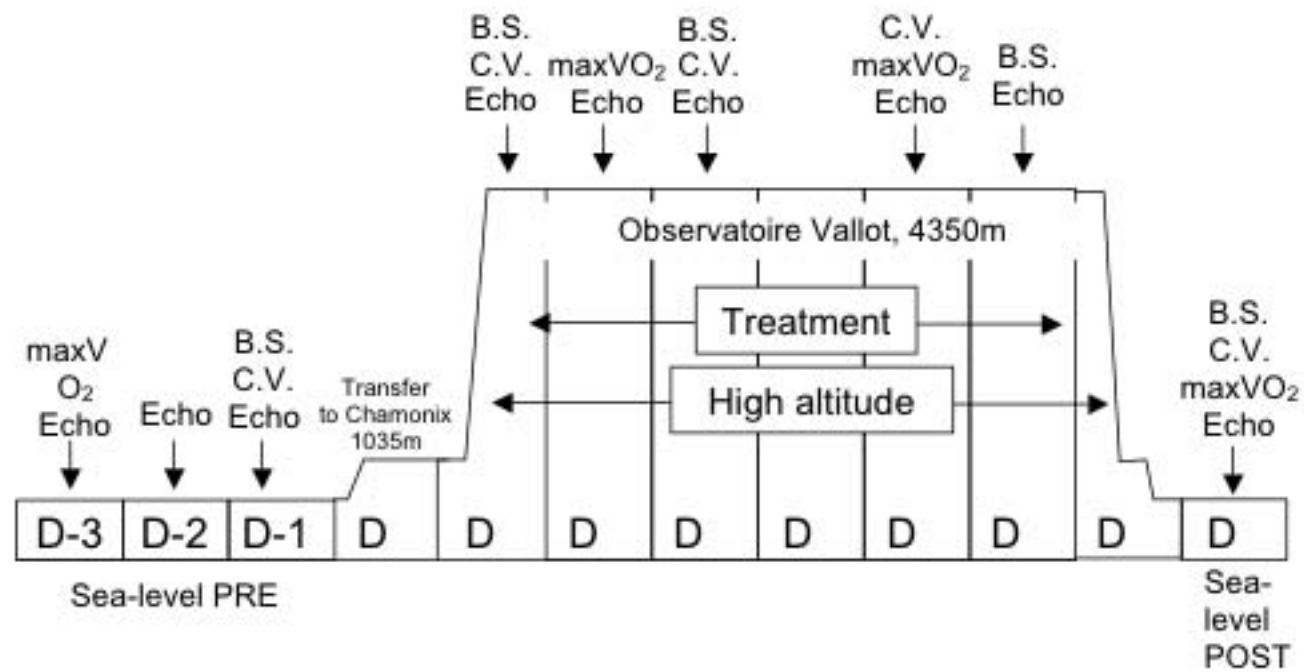
#, : p<0.05, SIL vs PLA. §, §§: p<0.05, p<0.01 D1 vs sea level pre for the whole group.

TABLE 3. Exercise and gas exchange data.

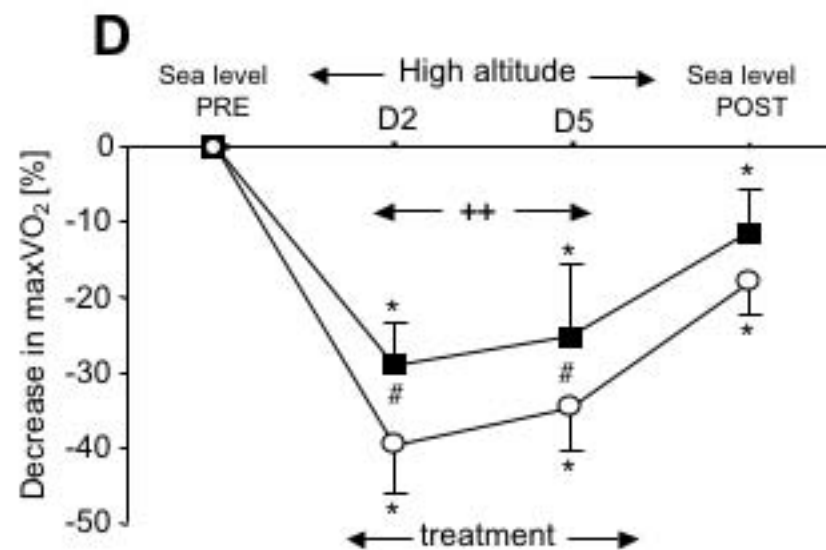
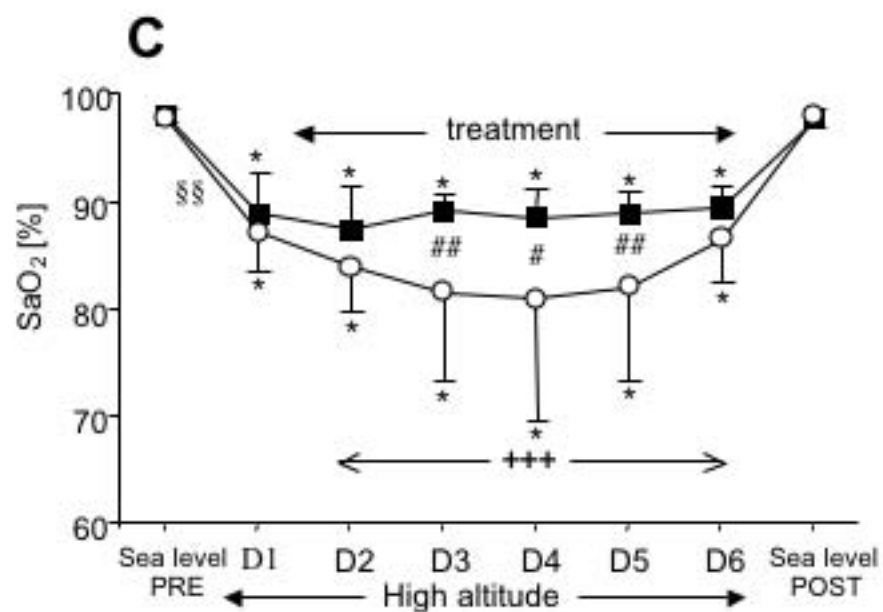
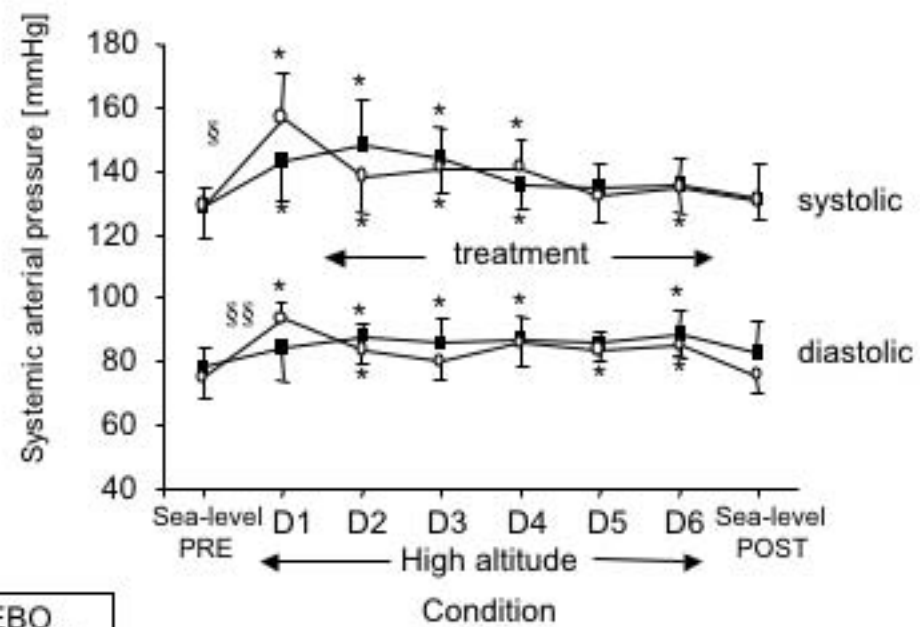
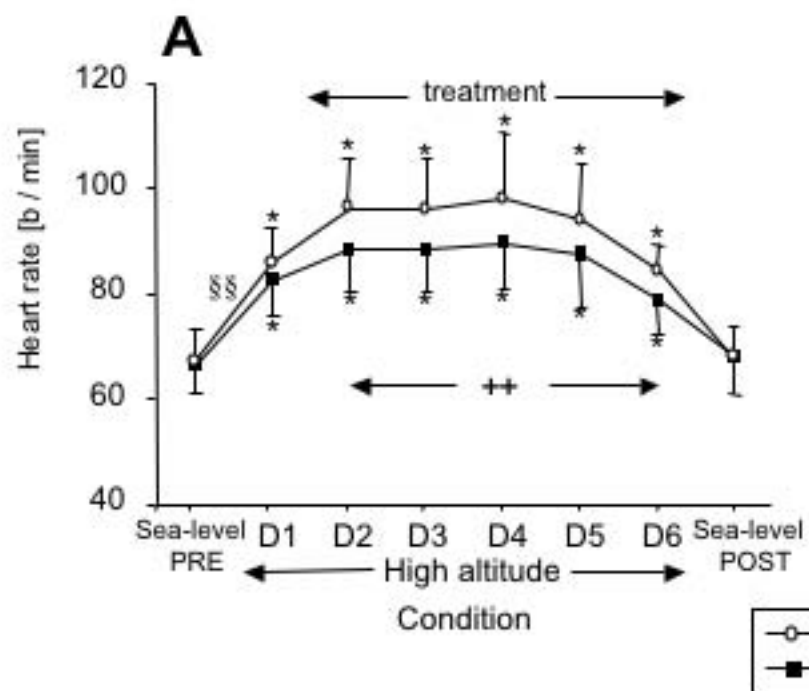
			Sea level pre	D2	D5	Sea level post
PaO ₂ , mmHg	rest	PLA	95.8 ± 5.3	47.7 ± 9.1 *	46.0 ± 7.2 *	90.1 ± 5.1 *
		SIL	94.7 ± 4.5	49.4 ± 4.1 *	52.2 ± 2.6 *,#	88.3 ± 8.7 *
	smax exercise	PLA	89.1 ± 7.0	39.9 ± 3.9 *	39.5 ± 3.0 *	90.5 ± 8.9 *
		SIL	91.4 ± 4.1	41.8 ± 2.6 *	44.8 ± 0.9 *,##	98.2 ± 3.2 *
PA-PaO ₂ , mmHg	rest	PLA	17.4 ± 5.4	12.0 ± 2.8	14.4 ± 5.2	30.4 ± 5.4 *
		SIL	18.2 ± 3.2	8.6 ± 2.0 *,#	8.4 ± 2.9 *,#	30.8 ± 5.1 *
	smax exercise	PLA	23.2 ± 5.2	23.8 ± 4.3	27.8 ± 6.0	32.1 ± 5.2
		SIL	22.9 ± 5.3	21.0 ± 3.8	22.0 ± 2.7 #	23.2 ± 3.8 #
PaCO ₂ , mmHg	rest	PLA	37.8 ± 2.1	26.5 ± 2.8 *	26.3 ± 3.0 *	35.3 ± 1.9
		SIL	38.2 ± 3.5	28.4 ± 1.7 *	24.6 ± 1.6 *	33.3 ± 2.5 *
	smax exercise	PLA	37.7 ± 3.4	26.3 ± 2.4 *	24.2 ± 1.2 *	33.5 ± 3.5 *
		SIL	38.7 ± 2.3	26.6 ± 1.6 *	24.0 ± 0.9 *	32.4 ± 1.9 *
pHa	rest	PLA	7.44 ± 0.02	7.50 ± 0.04 *	7.49 ± 0.01 *	7.44 ± 0.02
		SIL	7.43 ± 0.01	7.49 ± 0.02 *	7.52 ± 0.02 *	7.44 ± 0.02
	smax exercise	PLA	7.35 ± 0.01	7.43 ± 0.04 *	7.46 ± 0.04 *	7.37 ± 0.03
		SIL	7.37 ± 0.01	7.43 ± 0.04 *	7.47 ± 0.03 *	7.36 ± 0.04

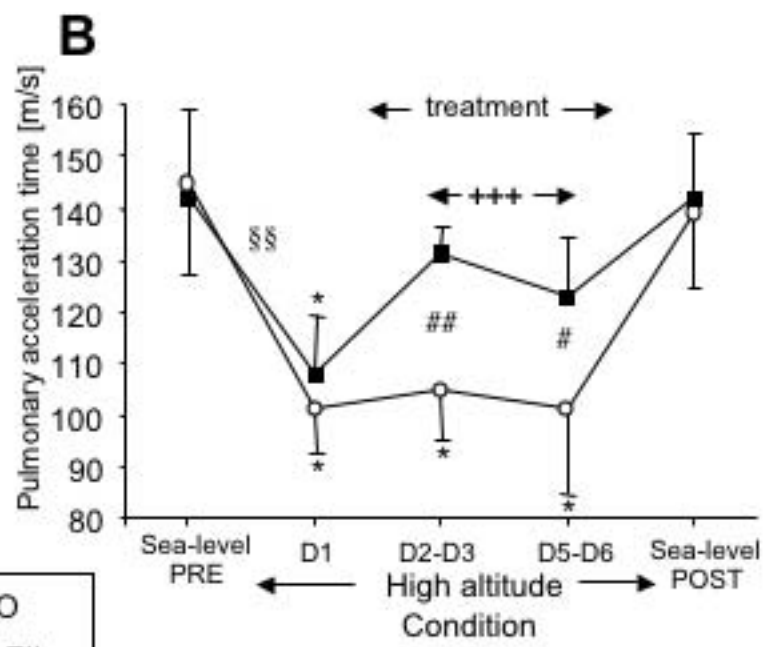
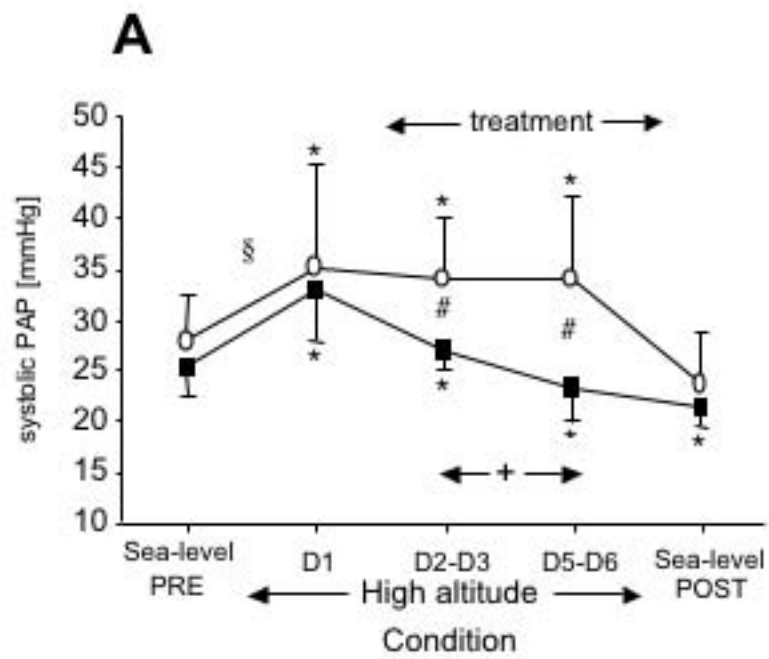
Cardiac output, l/min	rest	PLA	4.7 ± 0.9	7.7 ± 2.0 *	6.3 ± 1.8 *	5.4 ± 0.8
		SIL	5.4 ± 0.8	7.1 ± 1.4 *	6.7 ± 1.4 *	5.2 ± 0.8
	smax exercise	PLA	15.6 ± 1.8	17.5 ± 2.0	14.6 ± 2.7	15.0 ± 1.3
		SIL	15.6 ± 2.4	17.3 ± 4.6	14.9 ± 4.1	16.0 ± 1.9
Heart rate, b/min	smax exercise	PLA	173 ± 12	170 ± 15 *	158 ± 12 *	167 ± 15
		SIL	171 ± 11	163 ± 11 *	151 ± 11 *	161 ± 6
	max exercise	PLA	192 ± 14	184 ± 12 *	175 ± 13 *	187 ± 15 *
		SIL	194 ± 6	181 ± 9 *	173 ± 8 *	187 ± 6 *
Δ IFT index, %	rest	PLA	0	17.7 ± 9.1 *	22.5 ± 17.1 *	2.8 ± 16.3
		SIL	0	15.5 ± 11.5 *	2.8 ± 11.3 #	-3.7 ± 14.1

Sea level pre: basal normoxic condition, D2, D5: second and fifth day at 4,350m, Sea-level post: return to normoxic conditions. PaO₂, PaCO₂, pHa: arterialized blood gases; PA-PaO₂: alveolar-arterial difference in PO₂; Δ IFT index: variation of intrathoracic fluid from Sea level pre. *: p<0.05 vs sea level pre; #, ##: p<0.05, p<0.01 SIL vs PLA

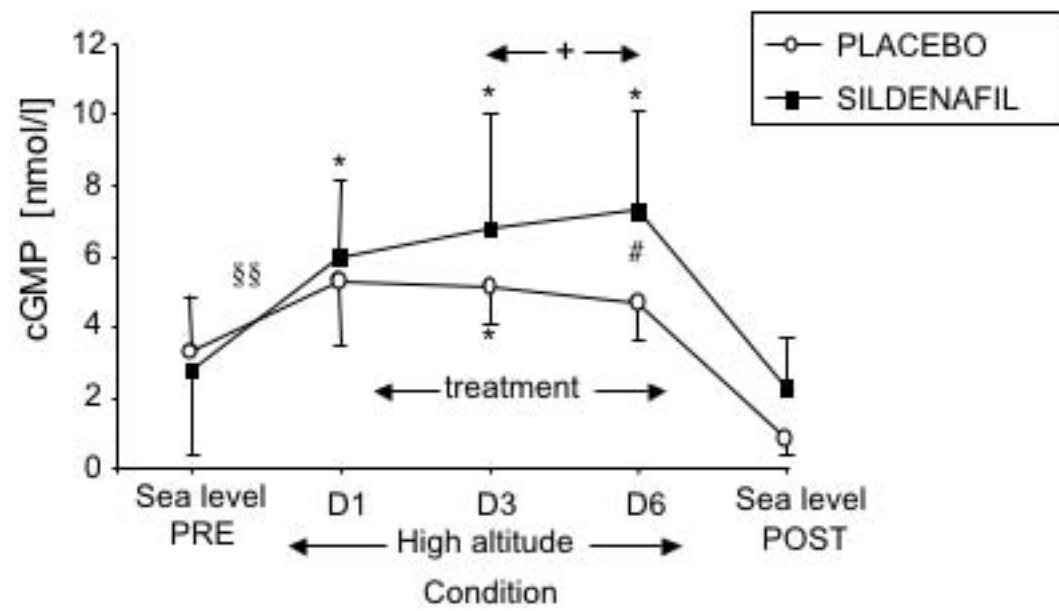


Richalet et al., figure 1.





Richalet et al., figure 3



Richalet et al., figure 4

Sildenafil inhibits the altitude-induced hypoxemia and pulmonary hypertension.

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Online data supplement

Methods

Subjects

The two groups of subjects were similar in age, years (29 ± 3 for PLA and 31 ± 6 for SIL, ns), body weight, kg (79 ± 6 for PLA and 78 ± 14 for SIL, ns) and height, cm (180 ± 6 for PLA and 181 ± 7 for SIL, ns). Subjects were healthy and had regular physical activity but were not trained athletes. They all had a previous experience of high altitude exposure but, at the beginning of the study, had not been to high altitude ($> 3,000\text{m}$) for six months. Before inclusion in the study, a clinical history and physical examination, a complete 12-lead ECG and an echocardiography were performed to exclude any significant cardiac abnormality. All subjects showed ECG and echo values within normal limits. They were not taking any medication.

Procedure

The altitudes facilities allow a comfortable environment with an ambient temperature kept between 15°C at night and 20°C during daytime.

Clinical evaluation.

The Lake Louise consensus questionnaire was used to evaluate the symptoms of AMS (headache, gastro-intestinal symptoms, dizziness, fatigue). In this questionnaire, each symptom was coded from 0 (absence) to 3 (extreme, debilitating). The Lake Louise score was calculated as the sum of the four items (maximum of 12).

Echocardiography.

Complete 2-dimensional, TM-echography and Doppler parameters for left cardiac function were recorded following classical procedures: aorta, left atrium, left ventricle diastolic and systolic diameters, mitral flow velocities and cardiac output. Tricuspid regurgitation (TR) was detected

by color-flow Doppler on 4-chamber apical or modified parasternal views. TR peak velocity was measured by continuous-wave Doppler. Systolic (sPAP) pulmonary arterial pressures was calculated by adding the right atrial pressure to the right atrium-right ventricle gradient. Pressure gradients between the right atrium and the right ventricle were then calculated by the Bernoulli equation: $\Delta P = 4 \cdot v^2$. A pulse-wave Doppler recording of systolic pulmonary flow was also analyzed in a transverse parasternal view with the sample volume positioned just below the pulmonary valve plane. Right atrium pressure was estimated by the inferior vena cava diameter and its variation during respiration. All echocardiographic data were digitalized and stored in the computer of the echographic device. Images were analyzed independently by two experienced echocardiographers in a blinded manner (before opening the code). When a discrepancy of more than 15% was found for an echographic parameter, an additional analysis was performed by both echocardiographers.

Maximal exercise test

After a 3-min warm-up at 60 watts, the workload was increased by 30 watts every two minutes until the imposed pedalling rate of 60 cycle/min could not be maintained despite strong encouragement. Ventilatory and metabolic parameters were measured breath-by-breath and then averaged every 15 seconds throughout the test, using a CPX-D Medical Graphics apparatus (Minneapolis, USA). The ventilatory threshold was determined visually at the change of slope of the end-tidal PCO_2 vs time curve.

Blood gases.

Arterialized blood gases were measured from a capillary sample obtained at rest and at the ventilatory threshold on the other ear, also previously dilated by a capsaicin cream.

Trans-thoracic impedencemetry

The position of the electrodes was carefully marked with a permanent marker during the experiments at sea-level and served as a reference for the recordings during and after high altitude exposure.

Color vision.

A color confusion index (CCI) was obtained by summing the differences between adjacent color caps according to Bowman. A test completed with no error shows a CCI value of 1.