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Pflugers Arch - Eur J Physiol (2002) 444:821–828
DOI 10.1007/s00424-002-0894-x

ORIGINAL ARTICLE

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Received: 30 January 2002 / Revised: 15 April 2002 / Accepted: 12 June 2002 / Published online: 2 August 2002

Abstract Plasma volume (PV) decreases at high altitude, but is rapidly restored upon return to sea-level (RSL). The aim of this study was (1) to describe PV recovery upon RSL with concomitant changes in major fluid regulating hormones, and (2) to test the hypothesis that PV recovery is promoted by the administration of a plasma expander. Ten male subjects were evaluated at rest and during submaximal exercise at sea-level (SL), after 7 days at 4,350 m (H7), and on RSL, on day 1 (RSL1, rest only) and day 2 (RSL2). PV (measured by carbon monoxide rebreathing), plasma renin (Ren), aldosterone (Aldo), atrial natriuretic factor (ANF) and arginine vasopressin (AVP) were measured at rest and during exercise. The subjects were divided into two groups 1 h before RSL, one group receiving PV expansion (475±219 ml) to ensure normovolemia (PVX, *n*=6), the others serving as controls (Control, *n*=4). PV decreased by 13.6% in H7 (*n*=10), but was restored in RSL2, regardless of PVX. Ren, Aldo and AVP, which were similar in both groups, were reduced in H7, but were higher in RSL2 (rest or exercise). ANF was modified neither by hypoxia nor by PVX. Total water intake was reduced in H7, but remained normal in RSL in both groups, whereas water output dropped in RSL. PVX increased urine flow rate in RSL1 compared with subjects not given PVX. The present results suggest that PV recovery during early RSL is mainly due to a decreased diuresis, promoted at least in part by changes in fluid regulating hormones. However, neither PV recovery, nor hormonal responses were altered with PVX-induced normovolemia upon RSL.

Keywords Carbon monoxide rebreathing · Gas exchange · High altitude · Hypoxia · Water intake · Water output

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Introduction

Plasma volume (PV) has been found to decrease within 1–3 days of high-altitude exposure and to remain low in chronic hypoxia [7, 11, 19, 22, 31]. This phenomenon leads to a rapid increase in hemoglobin concentration and arterial oxygen content, before erythropoiesis brings new red cells into the circulation. PV loss at high altitude is generally attributed to spontaneous hypohydration associated with an increase in diuresis during early acclimatization [10]. High altitude diuresis appears to be primarily related to the increase in peripheral chemoreceptor stimulation, and may also involve reflexes initiated by stretch receptors [9, 10, 27]. In this scheme, fluid- and sodium-regulating hormones are potent effector mechanisms for these reflexes [10]. Finally, direct effects of tissue hypoxia on renal function may promote high altitude diuresis [17]. On the other hand, a fluid shift from the intravascular to the interstitial and/or intracellular compartments may also account for PV loss in hypoxia [7]. In that alternative, the loss of plasma protein would play a major role [26].

With early (~8 h) return to sea-level (RSL), PV remains subnormal [26], then is restored within a few days [4, 14]. An expansion of PV, overriding sea-level (SL) value, has also been observed within the first week of RSL after prolonged high altitude exposure [1, 24]. These data support the idea that powerful mechanisms act to re-increase intravascular fluid volume upon early RSL. In a previous report, we found that, upon RSL after high-altitude exposure, PV expansion was associated with marked alterations in some water- and sodium-regulating hormones, i.e., renin, aldosterone and atrial natriuretic factor (ANF), suggesting a possible hormonal mediation [24].

Two events occurring during RSL may initiate PV recovery through water retention: (1) high-altitude hypovolemia, which induced appropriate hormonal changes, and (2) acute reoxygenation, i.e., suppression of hypoxia, known to be a potent stimulator of atrial natriuretic factor (ANF) secretion and an inhibitor of aldosterone release [2, 20]. The present study describes PV recovery upon early RSL after high-altitude exposure with concomitant changes in major fluid-regulating hormones and water intake/output.

To assess the role of the volemic stimulus on PV recovery, hypovolemia was abolished at the time of descent from high altitude by means of plasma volume expansion (PVX). This experiment tests the hypothesis that PV recovery is promoted by the administration of a plasma expander.

Submaximal exercise was used in this study to emphasize altitude- and RSL-induced changes in hormonal responses, i.e., renin, aldosterone, ANF and arginine vasopressin (AVP).

Materials and methods

Subjects

Ten male volunteers participated in the experiment. Each subject underwent a medical examination and was informed about the possible risks and the experimental procedures. Each volunteer gave informed consent. This study was approved by the Ethics Committee at the Necker Hospital, Paris, France. The subjects were moderately trained and were not acclimatized to altitude before the experiment. They had a mean (±SD) age of 29±7 years, height of 175±4 cm, and body weight of 69±8 kg.

Procedures

The subjects were investigated at SL in Paris and at the Observatoire Vallot on Mont-Blanc (altitude 4,350 m; barometric pressure, P_B 448 mmHg). After a night spent in Chamonix (1,000 m), the subjects were carried by helicopter at the altitude of 4,350 m. The high-altitude period lasted 7 days, during which the level of physical activity remained low. Ambient temperature into the observatory was controlled between 18°C and 23°C. The subjects were then transported back to SL where RSL experiments were performed during the first 2 days after high-altitude exposure, i.e., at day 1 (RSL1, 12 h post-altitude) and day 2 (RSL2, 24–30 h post-altitude). For each subject the investigations at SL, high altitude, and RSL2 were performed at the same time of day. For RSL1 (resting blood sampling only), all the subjects were evaluated simultaneously at 12 h post-altitude.

Maximal O_2 uptake ([Vitdot]$O_{2\ max}$) and maximal power (W_{max}, in watts) were measured at SL and after 6 days at 4,350 m by sitting bicycling on a mechanically braked cycle ergometer (Monark 864). The subjects performed an incremental exercise (30 W every 2 min) until exhaustion. Maximal exercise test was used to adjust the cycling intensity during a submaximal exercise, which was repeated three times, i.e., at SL, after 7 days (last day) of high-altitude exposure (H7), and on day 2 after RSL (RSL2). During SL and RSL experiments, the exercise intensity was kept constant at 50% of the W_{max} at SL for 30 min. At high altitude, exercise intensity was kept constant at 50% of high altitude W_{max} during the first 15 min, then increased to 50% of SL W_{max} during the second 15-min period. This protocol was used to compare normoxia and hypoxia as to the respective effects of same absolute and same relative workloads on exercise-induced hormonal responses. All the exercise procedures (maximal and submaximal exercises) were performed at a pedaling frequency of 60 rpm.

Gas exchange

Gas exchange was measured at rest and during maximal and submaximal exercises with an integrated computer system (CPX/D cardiopulmonary exercise system; Medical Graphics, Minneapolis, Minn., USA). Minute ventilation was measured by a symmetrically disposed Pitot tube flowmeter. O_2 concentration was measured by a galvanic fuel cell and CO_2 by an infrared analyzer. The characteristics of this device have been described previously [23]. Heart rate was measured continuously, as was arterial O_2 saturation (SaO_2), by a pulse oximeter (Biox II, Ohmeda).

Blood analyses

Investigations were preceded by 2 h of fasting. After a venous catheter was inserted in an antecubital vein of one arm, the subjects were confined to a resting sitting position for 1 h. After this period, resting blood sample was collected and then, resting plasma volume was determined by a carbon monoxide (CO) rebreathing technique [18]. Briefly, the subjects breathed into a closed circuit, including a 2-l rebreathing bag, and a CO_2 absorber. Extra O_2 was administrated into the circuit to compensate for O_2 consumption. Rebreathing time was 10 min to ensure that maximal hemoglobin CO saturation (COHb%) had been reached. The volume of CO administrated was 50 ml at SL and 60 ml at high altitude.

Before administration of CO and immediately after the completion of the rebreathing period, venous blood was collected in duplicate for measurement of COHb% and hemoglobin concentration ([Hb]) by spectrophotometry (CO oximeter, model 270, Ciba Corning). Hematocrit (Hct) was measured in triplicate by a microcentrifuge. Blood arterial O_2 content (CaO_2) was calculated as $[Hb]\times1.34\times SaO_2$. O_2 dissolved in plasma was neglected in the calculation.

The administrated amount of CO (nCO) was calculated as:

$$\mathrm{nCO} = (P\mathrm{B} \times V_{\mathrm{CO}} \times 1000 \times R^{-1} \times T^{-1}) \times 0.978$$

where P_B is in atmospheres, V_{CO} the volume of CO in litres, R the gas constant (0.082), T the temperature in Kelvin, and 0.978 is a correction factor for the CO remaining in the rebreathing system after equilibration with the blood.

The amount of circulating Hb (nHb) was calculated as:

$$\mathrm{nHb} = 100 \times \mathrm{nCO}/\Delta\mathrm{HbCO\%}$$

Blood volume (BV) was calculated as:

$$\mathrm{BV} = \mathrm{nHb}/[\mathrm{Hb}]$$

and the corresponding PV and red cell volume (RCV) were calculated as: PV=BV×(1–Hct) and RCV=BV×Hct, respectively.

The relative changes in PV (ΔPV, in %) were calculated from [Hb] and Hct with the following formula [Hct not corrected for F_{cell} ratio (overall hematocrit/peripheral hematocrit)] [8]:

$$\Delta PV\% = [([Hb]_{pre}/[Hb]_{post}) \times (100 - Hct_{post})/(100 - Hct_{pre}) - 1] \times 100$$

Forearm venous blood samples were collected at rest and during submaximal exercise, after 15 min (E15) and 30 min (E30) in SL, H7 and RN2. Additionally, a blood sample was drawn at rest in RSL1. [Hb] and Hct were measured immediately, whereas additional blood samples were immediately centrifuged and the separated plasma stored in liquid N_2 for later analysis.

Resting PV was determined three times by the CO rebreathing technique, at SL, at H7, and RSL2. Additionally, PV was calculated on day 1 after high-altitude exposure (RSL1) from ΔPV between RSL1 and RSL2, assuming that RCV did not change over this time period.

Plasma renin concentration ([Ren]) was measured by an immunoradiometric assay (Sanofi Diagnostics Pasteur, Marnes la Coquette, France) and plasma aldosterone concentration ([Aldo]) by a radioimmunoassay (Diagnostic Products Corporation, Los Angeles, Calif., USA). The mean reference value obtained in our laboratory from healthy subjects (20–40 years) in sitting position was 14.2 pg ml^{-1} (range 5.1–38.7) for renin and 14.3 ng/100 ml (range 6–35) for aldosterone. Plasma [ANF] was measured by a radioimmunoassay following an extraction step: briefly, blood was collected on a tube containing EDTA and trasylol (protease inhibitor). Plasma was separated and stored at –80°C until assayed. Then, 1 ml of plasma was acidified with 3 ml of a 4% acetic acid solution; 4 ml of acidified plasma was deposed on a C18 Sep Pak column (Waters Millford, Mass., USA) which had been previously activated with methanol (5 ml). The column was rinsed with 5 ml of distilled water. Elution was performed with 3 ml of the following solution: acetonitril 60% and trifluoroacetic acid 0.1% in distilled water. Eluate was dried under N_2 and thereafter assayed with a radioimmunoassay kit (Amersham International, Little Chalfont, UK). The mean reference value for [ANF] obtained in our laboratory from healthy subjects (26–50 years) in sitting position was 30.5 pg ml^{-1} (range 11.9–56.0).

Plasma [AVP] was determined by radioimmunoassay [3]. Synthetic AVP was purchased from ICN Biochemicals (USA) and was used for the preparation of standards. [^{125}I]Vasopressin[Arg^8] was purchased from Amersham Pharmacia (UK). The rabbit polyclonal antibody used in the assay has been produced in the laboratory [16].

Venous blood was drawn on dipotassium EDTA, centrifuged at 3,000 rpm for 15 min then the plasma was stored in liquid N_2 until analysis. Extraction was done with acetone according to Robertson et al. [25]. Mean yield of extraction was 96%. Diluent for the assay was 0.01 M sodium phosphate buffer, pH 7.3, containing 0.15 M NaCl, BSA 1 mg ml^{-1} and sodium azide 1 mg ml^{-1}. Each tube in the assay contained AVP (0–30 pg) or extracted fraction of plasma and antiserum at a final dilution of 1:150,000 under a total volume of 0.5 ml. After 2 days at +4°C, ^{125}I-AVP (1.0 pg) was added in each tube and after 2 days at +4°C, free and bound ^{125}I-AVP were separated using charcoal dextran. The supernatants and precipitates were counted in an automatic gamma-ray well-counter. For each plasma tested, nonspecific binding was estimated from incubation of the plasma in the absence of antibody. The smallest amount of ADH detectable with this assay was 0.56 pg/tube. Intra-assay and inter-assay variations were 12.5% and 17.5%, respectively. The antibody used cross-reacted with LVP (155%) and dDAVP (17.5%).

Plasma volume expansion

In order to restore PV acutely, simultaneously to return to SL, a plasma expander (Hesteril 6%, 6% hydroxyethyl starch, Fresenius) was infused intravenously in six subjects (PVX group), 1 h before leaving the high-altitude laboratory (day 8 at 4,350 m, 8:00 a.m.), whereas the four other subjects served as controls (control group). A plasma half life of 4 h was previously measured for this hydroxyethyl starch [29]. After infusion of Hesteril 6%, previous observations included (1) an immediate 100–140% PV expansion, (2) a stable 100% PV expansion over the first 4 h, and (3) a progressive decay in PV during the next 8 h [13]. In an attempt to form two comparable groups, individual altitude-induced PV changes were calculated, then the subjects were matched in pairs and randomly assigned to PVX or control groups. The mean volume (±SD) of plasma expander infused in the PVX group was 475±219 ml (range 250–750).

Water intake and output

Food and water were ingested ad libitum. Food and water intake was measured daily in a dietary record at SL, high altitude and return to RSL. The subjects recorded intake during each meal and snack, by using table scales to weigh individual food items and a graduate container for volume of drinks. The water content in food was derived from food tables [28]. Food tables [28] were also used for the determination of daily sodium intake. To determine water loss in urine and feces, subjects collected total urine and total feces for all days. Urine volume was measured at each voiding in a calibrated container. Feces were collected in preweighed bags and weighed. Water content in feces was estimated from data obtained previously at the same altitude [30], indicating a mean water fraction in feces of 0.76±0.03.

Statistics

Data are presented as arithmetic mean±SD. A Mann-Whitney *U*-test was used for analyses between groups (PVX and Control). Intra-individual differences were made with the paired Wilcoxon test, either in each group (PVX or Control) or in pooled data (n=10) when between-group analyses did not reveal any significant differences. A Pearson product-moment correlation test (r^2, coefficient of determination) was used to analyze the relationship between two quantitative variables. Differences were considered significant at P<0.05.

Results

Baseline SaO_2 was 97±1% at SL, 88±2% at high altitude, and 97±1% at RSL. Body mass decreased at high altitude and remained lower over the first two days at RSL in both control and PVX groups (Table 1). Water intake from food and beverages declined at 4,350 m, but was restored at RSL, independently of PVX (Table 1). Sodium intake from food and beverages did not change throughout the experiment, independently of PVX (Table 1). Urine flow rate and water output in urine and feces were unchanged after 7 days at 4,350 m, but decreased with RSL, at day 1 in the control group and at day 2 in the PVX group (Table 1). PVX increased urine flow rate and water output in urine and feces on the first day after return to SL compared with subjects not given PVX (Table 1). Hct and [Hb] increased at high altitude and remained higher at day 1 at RSL. Hct had returned to basal level during day 2 at RSL, whereas [Hb] was still elevated at that time, without any effect of PVX (Table 2). Plasma volume decreased by 13.6% (n=10) from 2.980±0.385 liters at SL to 2.575±0.499 liters at high altitude (P<0.05) (Fig. 1). PV was still lower on day 1 at RSL (2.482±0.256 l,

Table 1 Body weight, water intake, sodium intake and water output at sea-level (SL), high altitude and return to sea-level (RSL). Values are means±SD

	Control (n=4)	PVX (n=6)	All subjects (n=10)
Body weight, kg			
SL	68.3±7.8	69.2±8.9	68.8±8.0
4,350 m	67.5±8.7	67.8±7.9[a]	67.7±7.7*
RSL (day 1)	67.5±8.7	68.0±7.8	67.8±7.7*
RSL (day 2)	67.8±8.7	68.0±7.8	67.9±7.7*
Water intake from food and beverages (ml day^{-1})			
SL	2930±1009	3220±872	3104±886
4,350 m	1989±706*	2238±390[a]*	2138±517*
RSL (day 1)	3061±395	3034±591	3045±496
RSL (day 2)	2711±841	2980±764	2873±761
Sodium intake from food and beverages (mg day^{-1})			
SL	3213±1333	3686±630	3496±934
4,350 m	3874±657	3698±954[a]	3768±811
RSL (day 1)	3294±770	3240±759	3262±720
RSL (day 2)	3626±1208	3642±1017	3635±1030
Urine flow rate (ml day^{-1})			
SL	1530 ±730	2001±548	1813±635
4,350 m	1420±492	1589±551[a]	1522±507
RSL (day 1)	835±169*	1640±848**	1318±763***
RSL (day 2)	981±330*	1223±241*	1126±290*
Water output in urine and feces (ml day^{-1})			
SL	1665±653	2162±642	1963±661
4,350 m	1530±419	1708±557[a]	1637±489
RSL (day 1)	855±160*	1746±891**	1390±813***
RSL (day 2)	1139±422	1318±308*	1246±347*

*P<0.05; 4,350 m or RSL versus SL

**P<0.05; plasma volume expansion (PVX) versus Control

***Statistics not given since the difference was significant between control and PVX groups

[a] Indicates the time of PVX in six subjects, 1 h before leaving high altitude

Table 2 Hematocrit (Hct), hemoglobin (Hb) concentration, red cell volume and blood volume at SL, high altitude and RSL. Values are means±SD

	Control (n=4)	PVX (n=6)	All subjects (n=10)
Hct (%)			
SL	42.3±2.2	45.2±2.3	44.0±2.6
4,350 m	47.3±3.6*	49.3±2.7[a]*	48.5±3.1*
RSL (day 1)	47.3±2.8*	48.0±3.7*	47.7±3.2*
RSL (day 2)	43.3±3.0	45.7±2.9	44.7±3.1
[Hb] mmol l^{-1}			
SL	8.8±0.5	9.5±0.3	9.3±0.5
4,350 m	10.1±0.6*	10.7±0.7[a]*	10.5±0.7*
RSL (day 1)	10.0±0.6*	10.2±0.7*	10.1±0.6*
RSL (day 2)	9.2±0.5	9.8±0.5	9.6±0.6*
Blood volume (l)			
SL	5.554±0.452	5.158±0.637	5.316±0.579
4,350 m	5.044±0.860	4.962±1.002[a]	4.995±0.898
RSL (day 1)[b]	4.778±0.388	4.784±0.377	4.770±0.462*
RSL (day 2)	5.140±0.491	5.033±0.530	5.300±0.447
Red cell volume (l)			
SL	2.340±0.111	2.333±0.342	2.336±0.263
4,350 m	2.369±0.336	2.454±0.550[a]	2.420±0.455
RSL (day 1)	2.293±0.255	2.298±0.266	2.296±0.247
RSL (day 2)	2.293±0.255	2.298±0.266	2.296±0.247

*P<0.05; 4,350 m or RSL versus SL. There was no significant difference between PVX and control groups for any of the given variables

[a] Indicates the time of PVX in six subjects, 1 h before leaving high altitude

[b] Blood volume on RSL (day 1) was estimated from changes in [Hb] and Hct between RSL (day 1) and RSL (day 2), with the assumption that all effects were secondary to changes in plasma volume

P<0.05), but had almost recovered on day 2 at RSL (2.844±0.334 l, NS). PVX neither altered absolute PV at RSL (Fig. 1), nor PV changes between high altitude and RSL1 or RSL2 (results not shown). Blood volume was transiently decreased on day 1 at RSL, whereas red cell volume was not significantly altered through the experiment (Table 2). Mean CaO_2 (n=10) increased from 19.1±1.1 ml dl^{-1} at SL to 19.9±1.1 ml dl^{-1} at high altitude (P<0.05) and remained higher (19.7±1.3 ml dl^{-1}, P<0.05) with RSL (day 2). Finally, we observed that the re-increase in PV from high altitude to day 2 at RSL was significantly related to the concomitant decrease in urine volume, independently of PVX (Fig. 2), and to the altitude-induced increase in CaO_2, independently of PVX (Fig. 3).

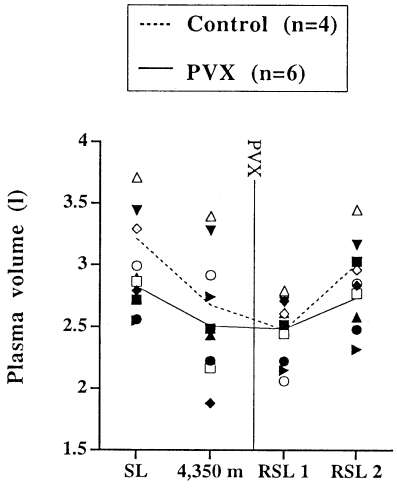

Fig. 1 Individual variations in plasma volume at sea-level (SL), after 7 days spent at the altitude of 4,350 m (*4,350 m*), and upon return to SL, on day 1 (*RSL1*) and day 2 (*RSL2*) in subjects without (*Control*, n=4) and with plasma volume expansion (*PVX*, n=6) performed 1 h before descent from high altitude. Mean values are *dotted* and *solid* lines, for control and PVX subjects, respectively

Exercise

$\dot{V}O_{2\ max}$ and W_{max} decreased from 46±3 ml min^{-1} kg^{-1} and 258±25 W at SL to 37±3 ml min^{-1} kg^{-1} and 193±22 W after 6 days at 4,350 m, respectively. Power

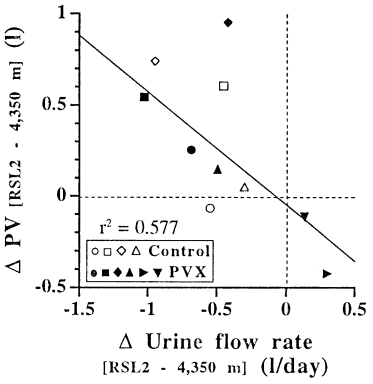

Fig. 2 Relationship between the decrease in urine flow rate and plasma volume (PV) recovery (ΔPV), from 4,350 m to RSL, day 2 (*RSL2*). Regression equation is (n=10): y=–0.001x–0.028; r^2=0.577, P<0.01

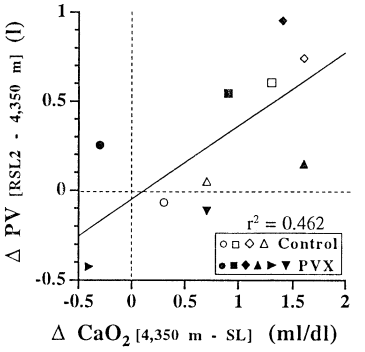

Fig. 3 Relationship between the increase in arterial oxygen content (CaO_2) from SL to 4,350 m and PV recovery (ΔPV) from 4,350 m to RSL, day 2 (*RSL2*). Regression equation is (n=10): y=0.409x –0.046; r^2=0.462

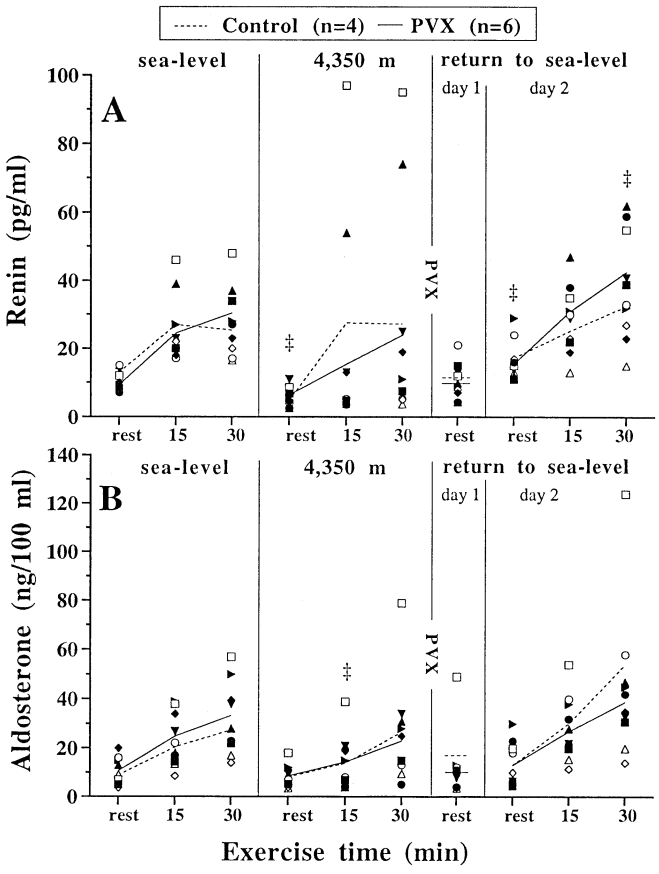

Fig. 4 Individual variations in plasma concentration of renin (**A**) and aldosterone (**B**) at rest and at 15 min and 30 min of submaximal exercise, at SL, after 7 days spent at the altitude of 4,350 m (*4,350 m*), and upon RSL, on day 1 (RSL1) and day 2 (RSL2) in subjects without (*Control*, n=4) and with plasma volume expansion (*PVX*, n=6) performed 1 h before descent from high altitude. Mean values are *dotted* and *solid* lines, for control and PVX subjects, respectively. Statistical significance is given for pooled subjects (n=10). ‡P<0.05; 4,350 m or RSL versus SL

Table 3 Power and cardiopulmonary data during a 30-min submaximal exercise. Data are mean±SD (n=10). $EX_{15\ min}$ and $EX_{30\ min}$ are measurements at 15 min and 30 min of submaximal exercise, respectively. Rate of O_2 uptake ($\dot{V}O_2$); heart rate (HR) and arterial O_2 saturation (SaO_2) were measured at SL, at an altitude of 4,350 m (day 7) and on RSL (day 2). There was no significant difference between PVX and control groups for any of the given variables

	Sea-level	High altitude	Return to sea-level
Power (watts)			
$EX_{15\ min}$	128±11	98±9	128±11
$EX_{30\ min}$	128±11	128±11	128±11
$\dot{V}O_2$ (ml min^{-1})			
$EX_{15\ min}$	1902±164	1798±206	1952±193
$EX_{30\ min}$	1980±166	2043±151	2042±153
HR (beats min^{-1})			
$EX_{15\ min}$	140±13	147±20	141±12
$EX_{30\ min}$	148±15	157±20	151±14
SaO_2 (%)			
$EX_{15\ min}$	96±0.8	82±4.9	97±0.8
$EX_{30\ min}$	96±0.7	81±3.7	97±0.6

output data and cardiopulmonary responses during 30 min of submaximal exercise were comparable in both groups (control and PVX) (Table 3). $\dot{V}O_2$ and heart rate during exercise were similar before and after high-altitude exposure (Table 3).

PVX had no subsequent effect on any of the measured hormones during RSL, neither at rest, nor at exercise (Figs. 4, 5). Compared with SL, mean resting levels of renin (n=10) decreased at 4,350 m. On day 2 at RSL, resting renin and renin response to exercise (n=10) had increased to values higher than before the high altitude sojourn (Fig. 4A). Mean aldosterone response to exercise (n=10) was lower at high altitude, and tended to be higher (P=0.10) on day 2 at RSL (Fig. 4B). Mean ANF values (n=10) were not altered by high altitude or RSL, neither at rest, nor during exercise (Fig. 5A). Mean resting AVP (n=10) was decreased at high altitude, whereas mean AVP response to exercise was found higher upon RSL (Fig. 5B). Finally, PVX had no subsequent effect on

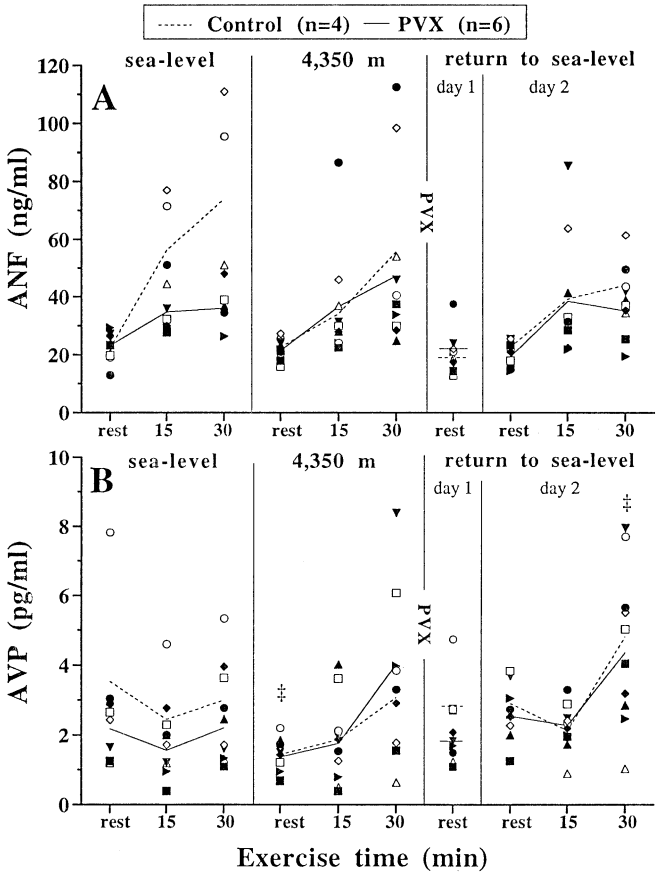

Fig. 5 Individual variations in plasma concentration of (**A**) atrial natriuretic factor (*ANF*) and (**B**) arginine vasopressin (*AVP*) at rest and at 15 min and 30 min of submaximal exercise, at SL, after 7 days spent at the altitude of 4,350 m (*4,350 m*), and upon RSL, on day 1 (RSL1) and day 2 (RSL2) in subjects without (*Control*, n=4) and with plasma volume expansion (*PVX*, n=6) performed 1 h before descent from high altitude. Mean values are *dotted* and *solid* lines, for control and PVX subjects, respectively. Statistical significance is given for pooled subjects (n=10). ‡P<0.05; 4,350 m or RSL versus SL

absolute PV during exercise or on exercise-induced PV decrease during RSL experiments (results not shown).

Discussion

The present results indicate that hypoxia-induced decrease in PV is corrected within 2 days (24–30 h) after return to SL. The re-increase in PV with descent from high altitude was related to a decline in diuresis, in accordance with the concomitant changes in fluid regulating hormones renin, aldosterone, AVP and ANF. When acute PVX-induced normovolemia was induced at the time of descent from high altitude, PV recovery was not altered, nor were the responses of fluid-regulating hormones.

The 13.6% decrease in PV observed after 7 days at 4,350 m in our subjects is in concordance with previous data obtained with the CO-rebreathing method at similar altitude [4, 18, 31]. With RSL, PV recovery after exposure to 4,300 m was found to be achieved within 4 days [14] or 5–12 days [4]. However, the present data demonstrated that PV may be restored as early as 24–30 h after altitude exposure. In Operation Everest III, we reported that after prolonged exposure to extreme altitude, PV may even be higher than the initial value at SL [24]. Such a PVX was also observed during recovery from a high-altitude expedition [1]. These data suggest that the restoration of intravascular fluid volume with RSL is controlled by powerful mechanisms. The lack of RSL-induced PVX in the present study could be attributed, at least in part, to the lower hypoxic stress and subsequent hypovolemia at 4,350 m (13.6% decrease in PV) than during extreme altitude exposure (7,600–8,848 m), where PV may be decreased by 25–26% [1, 24].

The re-increase in PV associated with descent from high altitude could be related to changes in one or several components of water balance, mainly decreased diuresis and/or fecal water, increased water intake, or reduced evaporative water loss, and to fluid shifts from the intracellular and/or interstitial compartments to the intravascular bed. The present results suggest that PV recovery with RSL would be primarily related to a decrease in diuresis (Fig. 2), whereas fecal water loss remained unchanged. Such a water retention (combined low diuresis and high water intake) during early rehabilitation from high altitude was previously observed [15]. The trend toward an increase in total water intake from 4,350 m to RSL2 (Table 1) may also account for concomitant PV recovery. However, the poor relationship between the two variables (r^2=0.09; P=0.38) leads to speculate that restoration of water intake with RSL would be of less importance.

Since we did not measure the other components of water balance, we can not exclude the possibility that PV recovery at RSL was also related to a decrease in evaporative water loss. However, this alternative is not supported by previous data, indicating that sweat loss was not found to be decreased with early RSL [15].

Prolonged hypoxia is associated with a general hypohydration, i.e., decrease in total body water, plasma volume, extracellular and intracellular water, rather than a redistribution of body fluids toward intracellular compartment [10, 11, 14]. Furthermore, Krzywicki et al. [15] reported that intracellular and extracellular water did not significantly change with rehabilitation from high altitude, supporting the idea that redistribution of body fluids would not primarily account for PV recovery upon RSL. Alternatively, it may be pointed out that these authors noted a trend toward an increase in extracellular water both at high altitude and with rehabilitation from altitude [15], whereas PV was found lower at high altitude by the same group [14]. Taken together, these observations highlight the fact that fluid shifts from the intravascular to the interstitial compartment might be of importance and even larger than those to the intracellular compartment during high altitude exposure. Therefore, that fluid shifts (from the interstitial to the intravascular bed) were

involved in PV recovery with RSL could not be excluded in the present study.

Hormonal response

At high altitude, eight of the ten subjects experienced a decrease in renin response during exercise at the same relative intensity, aldosterone response was blunted at that time, ANF remained unchanged and resting AVP was lower. Such features are consistent with numerous previous data, although conflicting results exist in the literature [21, 27]. With descent from high altitude, mean resting values of renin were above basal level, whereas resting aldosterone, ANF and AVP were not significantly higher. However, exercise data revealed that, upon RSL, renin and AVP responses were higher (aldosterone response tended to be higher) and ANF response tended to be lower for the same absolute exercise intensity. These results, which are in agreement with previous data obtained during maximal exercise [24], support the idea that the lowered diuresis observed during early RSL may be initiated by the above mentioned hormonal changes.

However, water retention upon RSL was poorly correlated to the resting hormonal levels, suggesting that other mediators may be implicated. Acute reoxygenation with RSL, which blunts the permanently high chemoreceptor activity at altitude, likely plays a role on antidiuresis and subsequent PV recovery. This mechanism would be the opposite of what happens during acute hypoxic exposure, i.e., increased peripheral chemoreflex and diuresis. The fact that the observed antidiuresis was not fully explained by the measured hormonal changes evokes other chemoreflex effectors, such as brain natriuretic peptide, digitalis-like immunoreactive substance, endothelin, adrenomedullin or urodilatin [27]. On the other hand, antidiuresis may also be explained by an increase in renal sympathetic nerve activity [12]. However, it was unlikely in the present RSL experiment, since global sympathetic activity would rather be lower with suppression of the hypoxic stress. Finally, as speculated earlier [1] and also suggested by our data (Fig. 3), a blood O_2 content-regulating mechanism could be implicated in the re-increase of PV at RSL.

The question arises, what is the role of hypovolemia on PV recovery? PVX was performed to ensure normovolemia at the time of descent from high altitude, in an attempt to accelerate PV recovery with RSL. However, this procedure failed to do so, since PVX abolished the antidiuresis experienced by the control subjects (Table 1). As a consequence, PV was still depressed at day 1 at RSL in both groups. Thus, despite the presence of a hypovolemic status with early RSL, a diuretic response to acute water load occurred, suggesting that water regulation was still efficient at that time, and likely reset to a lower PV reference. If not, one might speculate that PVX-induced normovolemia would not have blunted the process of water retention, thus leading to an earlier restoration of intravascular fluid volume at RSL.

The role of PVX on the low-pressure system has to be considered in the present work, since more than 90% of the infusion entered this part of the circulatory system. By increasing the "fullness" of the low-pressure system, PVX was probably associated with an increase in central venous pressure (CVP), which stimulated the volume receptors located in the right cardiac atria (distension), leading in turn to an increase in ANF, a decrease in AVP and subsequent diuresis [12]. Although diuresis was demonstrated by our RSL data, the suspected hormonal changes were not more significant at the time of sampling (12 h after PVX). CVP was found to be decreased at high altitude [5, 6]. However, hormonal systems do not necessarily follow CVP changes, again suggesting that stretch receptor responses and water-regulating hormones may be reset to a lower CVP at high altitude. The present data lead to speculate that, facing an acute increase in CVP (infusion) even during descent from high altitude, water regulation would remain efficient.

Nevertheless, it is not evident from our data whether hypovolemia per se was the main factor for PV recovery. Indeed, it could be supposed that the combined action of the two stimuli, i.e., sustained hypovolemia and suppression of hypoxia, would promote PV recovery upon early RSL.

In summary, the present data demonstrated that the early re-increase in PV upon RSL was primarily related to the concomitant antidiuresis. Acute normovolemia with PVX during descent from high altitude failed to accelerate the restoration of PV. Even if recovery of PV was mainly due to a decrease in urine flow rate, the responsible mechanisms remain unknown. It was not caused by simple hypohydration since urine flow rate increased after PVX. Importantly, renin and ADH was normalized very quickly upon RSL, which together with normal values of ANF argues against severe hypohydration.

Acknowledgements The authors thank the ten volunteers, as well as the Society Chiron Diagnostics, for their participation to this experiment. We thank Dr. Jean-Pierre Herry, Ecole Nationale de Ski et d'Alpinisme, for skillful assistance during the high-altitude experiments. We also want to express our gratitude for their assistance to Dr. Paget and Dr. Clergeau, from the laboratory of Sallanches Hospital.

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