

Prolonged hypobaric hypoxemia attenuates vasopressin secretion and renal response to osmostimulation in men

MORTEN H. BESTLE,^{1,2} NIELS V. OLSEN,³ TROELS D. POULSEN,² ROBERT ROACH,⁴ NIELS FOGH-ANDERSEN,⁵ AND PETER BIE⁶

¹Departments of Clinical Physiology and ⁵Clinical Chemistry, Herlev Hospital, University of Copenhagen, DK-2730 Herlev; ²Department of Anesthesia and Intensive Care, Gentofte Hospital, University of Copenhagen, DK-2900 Hellerup; ³Department of Neuroanesthesia, and ⁴Copenhagen Muscle Research Centre, Copenhagen University Hospital, DK-2100 Copenhagen; and ⁶Department of Medical Biology, University of Southern Denmark, Odense, DK-5000 Odense, Denmark

Received 11 September 2001; accepted in final form 10 December 2001

Bestle, Morten H., Niels V. Olsen, Troels D. Poulsen, Robert Roach, Niels Fogh-Andersen, and Peter Bie.

Prolonged hypobaric hypoxemia attenuates vasopressin secretion and renal response to osmostimulation in men. *J Appl Physiol* 92: 1911–1922, 2002. First published January 11, 2002; 10.1152/japplphysiol.00936.2001.—Effects of hypobaric hypoxemia on endocrine and renal parameters of body fluid homeostasis were investigated in eight normal men during a sojourn of 8 days at an altitude of 4,559 m. Endocrine and renal responses to an osmotic stimulus (5% hypertonic saline, 3.6 ml/kg over 1 h) were investigated at sea level and on day 6 at altitude. Several days of hypobaric hypoxemia reduced body weight (-2.1 ± 0.4 kg), increased plasma osmolality ($+5.3 \pm 1.4$ mosmol/kgH₂O), elevated blood pressure ($+12 \pm 1$ mmHg), reduced creatinine clearance (122 ± 6 to 96 ± 10 ml/min), inhibited the renin system (19.5 ± 2.0 to 10.9 ± 0.9 mU/l) and plasma vasopressin (1.14 ± 0.16 to 0.38 ± 0.06 pg/ml), and doubled circulating levels of norepinephrine (103 ± 16 to 191 ± 35 pg/ml) and endothelin-1 (3.0 ± 0.2 to 6.3 ± 0.6 pg/ml), whereas urodilatin excretion rate decreased from day 2 (all changes $P < 0.05$ compared with sea level). Plasma arginine vasopressin response and the antidiuretic response to hypertonic saline loading were unchanged, but the natriuretic response was attenuated. In conclusion, chronic hypobaric hypoxemia 1) elevates the set point of plasma osmolality-to-plasma vasopressin relationship, possibly because of concurrent hypertension, thereby causing hypovolemia and hyperosmolality, and 2) blunts the natriuretic response to hypertonic volume expansion, possibly because of elevated circulating levels of norepinephrine and endothelin, reduced urodilatin synthesis, or attenuated inhibition of the renin system.

endothelin-1; hypertonic saline; sodium excretion; urodilatin

WATER AND SODIUM BALANCE ARE often affected by high altitude. During acute hypoxemia (hours), renal excretion rates of sodium and water usually increase (15, 16, 21, 51) but thereafter stabilize at an unchanged or lower level compared with normoxemia (1, 25, 30).

After several days of hypoxemia, total body water can be reduced by 1–3 liters (11, 18, 22), possibly due to a greater loss of insensible water because of lower humidity and increased respiration (17) or to a reduced intake of water and sodium because of poor thirst and appetite (22). Maintenance of water intake at altitude, however, does not prevent the decrease in total body water (11), indicating that renal mechanisms are probably involved.

Provided that oral water intake is adequate, water balance is mainly regulated through changes in plasma osmolality and plasma concentration of arginine vasopressin. Results obtained after 3–4 days at 3,000 m (36) and after several days at 5,400 and 6,300 m (4) showing that plasma vasopressin was reduced or unchanged despite an increase in plasma osmolality indicated that the relationship between plasma osmolality and plasma vasopressin was changed in the direction of reduced sensitivity. The expected increase in plasma vasopressin after infusion of hypertonic saline has been reported to be absent after 3–4 days at 3,000 m but not during short-term normobaric hypoxemia induced by breathing of hypoxic gas (36). A reduced vasopressin response to hypertonic saline infusion was also found in humans adapted to altitude living (35, 38). Previous results, therefore, suggest that sustained hypoxemia modulates vasopressin secretion perhaps because of a reduced sensitivity to changes in plasma osmolality. Vasopressin secretion is mainly regulated by osmoreceptors in hypothalamus (42), but changes in cardiovascular hemodynamics may also play a role. Moreover, plasma cortisol concentration, known to increase at high altitude (11, 15, 28, 40), may inhibit vasopressin secretion (31, 32).

Although body fluid homeostasis involves a complex interaction between several physiological systems, most earlier high-altitude studies have focused on the effect of hypoxemia on individual components in water

Address for reprint requests and other correspondence: M. H. Bestle, Dept. of Anesthesia and Intensive Care, Gentofte Hospital, Univ. of Copenhagen, DK-2900 Hellerup, Denmark (E-mail: mbestle@dadlnet.dk).

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked "advertisement" in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

and sodium homeostatic mechanisms, and, moreover, time-dependent changes mostly have remained uninvestigated. The aim of the present study was to describe the time-dependent, altitude-induced changes in important endocrine and renal parameters involved in the regulation of sodium and water homeostasis during a sojourn of 8 days at 4,559 m. Furthermore, we tested the hypothesis that hypoxemia attenuates the vasopressin response to an osmotic stimulus as presented by an infusion of hypertonic saline at *day 6* at altitude.

METHODS

Eight male subjects (aged 22–32 yr, mean of 26 yr; weight 65.7–80.2 kg, mean of 72.3 kg; height 174–187 cm, mean of 182 cm) participated in the experiment after they had given their written, informed consent. They were all sea level residents and were recruited among students. All subjects had a negative history of cardiovascular and kidney disease and were healthy as indicated by a physical examination and normal plasma concentrations of hemoglobin, creatinine, sodium, and potassium. The protocol was approved by the regional scientific ethical committee of Copenhagen, Denmark (j.no. KA95129).

Subjects were investigated at sea level (Herlev Hospital, University of Copenhagen) and during a sojourn of 8 days at high altitude (Capanna Regina Margherita, Mt. Rosa, Italy, 4,559 m, equivalent to a barometric pressure of ~435 Torr). Transportation to the Regina Margherita hut took place by helicopter. Sea level experiments were performed at least 2 wk before or 6 wk after altitude exposure.

At sea level, subjects were instructed to restrict to their normal diet and to avoid salty and spicy meals for 3 days before each study day. In the same period, they were instructed to avoid strenuous physical activity and alcohol- and caffeine-containing drinks, to abstain from smoking, and to have at least 6 h of sleep per night. Furthermore, they were instructed to fast for 8 h before the experiment and, for sea level experiments, use passive transportation to the laboratory to minimize physical activity on the study day. At altitude, all subjects ate the same diet provided by the staff at the Capanna Margherita, but food and fluid intake were ad libitum except during fasting for 8 h before investigation. All other instructions were identical to those at sea level.

Protocol 1: adaptation to altitude. At sea level, subjects were studied on two occasions separated by an interval of 7 days. At high altitude, subjects were studied on *days 1, 2, and 3* and again on *days 6, 7, and 8*. Subjects awoke at 7:00 AM for voiding and measuring of body weight but thereafter rested in supine position. Symptoms of acute mountain sickness (AMS) were determined according to the Lake Louise AMS scoring system (41). A venous cannula was placed in a cubital vein. After at least 10 min of rest, blood pressure and heart rate were measured manually by a sphygmomanometer. Peripheral oxygen saturation was measured by a pulse oximeter (Nellcor model N-3000, Pleasanton, CA) with the sensor attached to an ear lobe. Thereafter, 25 ml of venous blood were sampled for determination of osmolality and concentrations of hemoglobin, albumin, sodium, potassium, creatinine, renin, aldosterone, atrial natriuretic peptide (ANP), vasopressin, epinephrine, and norepinephrine. On each study day, a 24-h urine collection was performed, starting at 6:00 PM on the day of arrival (*day 0*), for measurements of urine volume, urine osmolality, and concentrations of sodium, potassium, creatinine, albumin, and urodilatin.

At sea level, subjects arrived at the laboratory at 8:00 AM on two different occasions, and the same procedure as at altitude was carried out. Sea level measurements are, therefore, expressed as mean values of two measurements made on two different days. However, plasma concentrations of epinephrine and norepinephrine, peripheral oxygen saturation, and AMS score were only determined once at sea level.

As part of another protocol (23), investigation of glucose homeostasis, involving a 2-h, euglycemic, hyperinsulinemic clamp, was performed on *days 2* and *7* after the samples of *protocol 1* had been obtained.

Protocol 2: hypertonic load. All subjects were studied on three occasions: 1) a time-control experiment at sea level (Herlev Hospital, University of Copenhagen), 2) a hypertonic saline load at sea level at the hospital, and 3) a hypertonic saline load on *day 6* at high altitude (4,559 m). On the study day, subjects awoke at 7:00 AM, and, after voiding, a water load of 500 ml was ingested. Water diuresis was obtained by replenishing renal and extrarenal losses with water. Urine volume was measured after each time subjects voided, and thereafter the same amount of water plus 1 ml/kg body wt (to replace extrarenal losses) was ingested. After subjects were weighed, they were confined to a resting supine position except for briefly standing during voiding every 60 min. Venous cannulae were placed in the cubital vein of both arms for infusion of hypertonic saline and withdrawal of blood samples, respectively. After 60 min of equilibration, five consecutive periods of 60 min each, including one baseline period (*period 1*), one infusion period (*period 2*), and three recovery periods (*periods 3–5*), were carried out. During the 60-min infusion period, subjects received an intravenous infusion of hypertonic saline (5%, ~856 mM) at an infusion rate of 3.6 ml·kg body wt⁻¹·h⁻¹. During the time control experiment, no infusion was given. Urine volume was determined for each period, and urine samples were obtained for measurements of urine osmolality and concentrations of sodium, potassium, creatinine, albumin, and urodilatin. Blood samples of 30 ml were obtained every 60 min at the end of the first four periods for measurements of osmolality and concentrations of sodium, potassium, creatinine, renin, aldosterone, ANP, arginine vasopressin, endothelin-1, epinephrine, and norepinephrine. Blood pressure and heart rate were measured manually by a sphygmomanometer every 60 min in the middle of each period. Body weight, after subjects were allowed to void, was measured immediately when subjects arrived to the laboratory and at the end of the last period.

Analyses. Urine concentrations of sodium, potassium, and creatinine were measured with a Vitros 250 (Ortho Clinical Diagnostics, Rochester, NY). Plasma concentrations of sodium, potassium, creatinine, and albumin were determined with a Vitros 950 (Ortho Clinical Diagnostics). Hemoglobin concentrations were measured in heparinized blood (OSM-3, Radiometer, Denmark). Osmolality in plasma and urine was measured by freezing-point depression (Gonotec Osmomat 030D, Berlin, Germany). Concentrations of albumin in urine were measured by enzyme immunoassay (10). Plasma concentrations of hormones and urine concentrations of urodilatin were measured by radioimmunoassays. Blood samples for hormone measurements were obtained in precooled polyethylene tubes containing EDTA and aprotinin (Novo Nordisk, Bagsvaerd, Denmark). All blood samples were centrifuged immediately at 4°C, and plasma was stored immediately on dry ice and thereafter at –18°C until measurement. Plasma and urine samples were acidified with acetic acid, and peptides were extracted with the use of C₁₈ Sep-Pak cartridges (Waters, Millipore, Bedford, MA), as previously described (8). ANP immunoreactivity was determined by using an antibody

purchased from Peninsula Laboratories (Merseyside, UK) by a procedure previously described (46). Intra-assay variation coefficients at an ANP concentration of 20 pg/ml were ~5%. Extraction recovery of unlabeled ANP was 70%. Vasopressin immunoreactivity was determined with the use of a specific antibody (kindly provided by Dr. Joergen Warberg) (20) according to a procedure described by Emmeluth et al. (9). There was negligible cross reactivity with oxytocin, vasotocin, and angiotensin II. Detection limit was <0.2 pg/ml. Extraction recovery of unlabeled vasopressin added to plasma was 85%. Intra-assay variation coefficient was 8% at a vasopressin concentration of 1.2 pg/ml. Endothelin-1 immunoreactivity in plasma was determined with the use of an antibody purchased from Peninsula Laboratories by a procedure previously described (8). Detection limit was <0.9 pg/ml. Extraction recovery of unlabeled endothelin-1 added to plasma was 93%. Intra-assay variation coefficients at an endothelin-1 concentration of 6 pg/ml were ~10%. Plasma renin concentration was measured by a two-site, two-monoclonal antibody immunoradiometric assay with plastic beads for solid phase (Nichols Institute, San Juan Capistrano, CA). The bound antibody was directed against the active site of renin, whereas the other antibody was iodinated. Limit of detection was 2 mU/l. One milli-international unit per liter obtained by the assay is equivalent to 0.6 µg/l of active renin (49). Intra- and interassay coefficients of variation were 4 and 5%, respectively. Aldosterone concentration in plasma was measured by radioimmunoassay in unextracted EDTA/aprotinin plasma (Diagnostic Products, Los Angeles, CA). Detection limit was 41 pmol/l. Intra- and interassay coefficients of variation were 10 and 15%, respectively. Urodilatin immunoreactivity in urine was determined by a specific antibody (S1969, kindly supplied by Biomedica, Vienna, Austria), as previously described (3). Cross-reactivity with human ANP-(99–126), human ANP-(4–28), rat ANP, endothelin-1, vasopressin, and angiotensin II was <0.001%. Detection limit was 0.5 pg/ml urine, and extraction recovery of unlabeled urodilatin was 94% for *protocol 1* and 70% for *protocol 2*. Intra-assay coefficients of variation were 13.4%, with a concentration of 6 pg/ml of urine. Results of radioimmunoassays were not corrected for incomplete recovery. For catecholamine measurements, blood was sampled in pre-cooled polyethylene tubes containing EGTA and glutathione. Concentrations of epinephrine and norepinephrine in plasma were measured by a radioenzymatic assay in *protocol 1*, as previously described (6), and by high-pressure liquid chromatography in *protocol 2*.

Missing values. In *protocol 2*, one subject had difficulty in emptying the bladder during voiding, which was reflected by irregular values of urinary excretion rates of creatinine. This subject was therefore excluded from the calculations of renal clearances and urinary excretion rates.

Statistical analysis. Results are presented as means ± SE. Data from *protocol 1* were subjected to a one-way ANOVA for repeated measures. In cases of $P < 0.05$, differences between

sea level and altitude values were evaluated by Newman-Keuls test. Data from *protocol 2* were subjected to a two-way ANOVA for repeated measures within series and between series. In cases of $P < 0.05$, differences between baseline and infusion periods, between time control and infusion series, and between sea level and altitude series were evaluated by Newman-Keuls test. $P < 0.05$ was considered to indicate significance. In cases of variance inhomogeneity, data were subjected to logarithmic transformation before statistical evaluation.

RESULTS

Adaptation to altitude. After arrival at high altitude, all subjects rapidly developed symptoms of AMS. On *day 1*, all subjects had three or more points on AMS score, but symptoms thereafter subsided and AMS score declined toward zero (Table 1). Oxygen saturation declined to $76 \pm 4\%$ on *day 1* but thereafter increased gradually (Table 1). Body weight declined at altitude in all subjects and was significantly reduced on *day 8* by 2.4 ± 0.4 kg compared with body weight at sea level (Table 1).

Heart rate and blood pressure increased from *day 1* at altitude (Fig. 1, A and B). Mean arterial blood pressure increased from 84 ± 2 mmHg at sea level to between 91 ± 4 and 96 ± 3 mmHg at high altitude. Systolic pressure increased from *day 1*, whereas diastolic pressure was not significantly elevated until *day 3*. As a result, pulse pressure increased on *day 1* but was not different from sea level on *days 2–8*. Norepinephrine tended to increase at altitude (ANOVA: $P < 0.09$), but significant changes could not be detected because of considerable variations between subjects (Fig. 1C). Epinephrine concentrations in plasma gradually decreased at high altitude (Fig. 1C).

Plasma osmolality increased on *days 3–8* at altitude from 291 ± 1 mosmol/kgH₂O at sea level to a maximum of 296 ± 1 mosmol/kgH₂O on *days 6* and *7* (Fig. 2). Plasma vasopressin concentrations decreased from 1.1 ± 0.2 pg/ml at sea level to a minimum of 0.4 ± 0.1 pg/ml on *day 7* (Fig. 2).

Hypoxemia caused marked decreases (~50%) in plasma concentrations of renin and aldosterone, whereas ANP concentrations remained unchanged (Fig. 3). Urodilatin excretion rate declined on *days 2–8* compared with that at sea level (Fig. 3). However, on *day 6*, urodilatin excretion rate increased and was significantly higher than rates on *days 2, 3, 7, and 8*, coinciding with the hypertonic saline infusion given on *day 6* (*protocol 2*). Hemoglobin concentrations increased from *day 1* at altitude (Table 2). Plasma so-

Table 1. Body weight, AMS, and O₂ saturation

	Sea Level	Days at High Altitude					
		1	2	3	6	7	8
AMS, point	0.0 ± 0.0	7.6 ± 0.9*	5.1 ± 1.1*†	2.4 ± 0.7†	0.3 ± 0.2†	0.3 ± 0.2†	0.4 ± 0.4†
O ₂ saturation, %	96 ± 0	76 ± 4*	78 ± 3*	83 ± 4*†	85 ± 2*†	85 ± 3*†	87 ± 2*†
Body weight, kg	72.3 ± 1.9	73.2 ± 2.1	72.5 ± 2.2	71.7 ± 1.9†	71.3 ± 1.9†	71.3 ± 1.9†	70.1 ± 2.0*†

Values are means ± SE ($n = 8$). AMS, acute mountain sickness score. *Significantly different from sea level ($P < 0.05$). †Significantly different from *day 1* at altitude ($P < 0.05$).

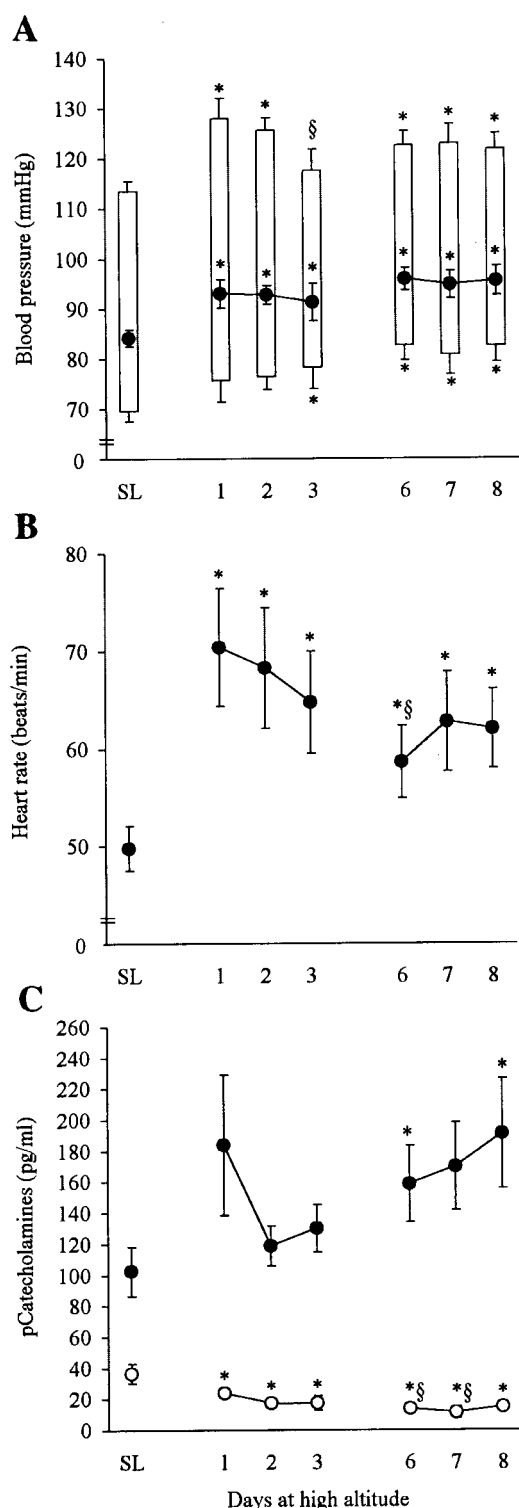


Fig. 1. Effects of high altitude on blood pressure (A), heart rate (B), and catecholamine concentrations in plasma (pCatecholamines; C). A: open bars, systolic and diastolic pressures; ●, mean arterial pressure. C: ●, norepinephrine; ○, epinephrine. Values are means \pm SE ($n = 8$). SL, sea level. *Significant deviation from SL ($P < 0.05$). §Significant deviation from day 1 ($P < 0.05$).

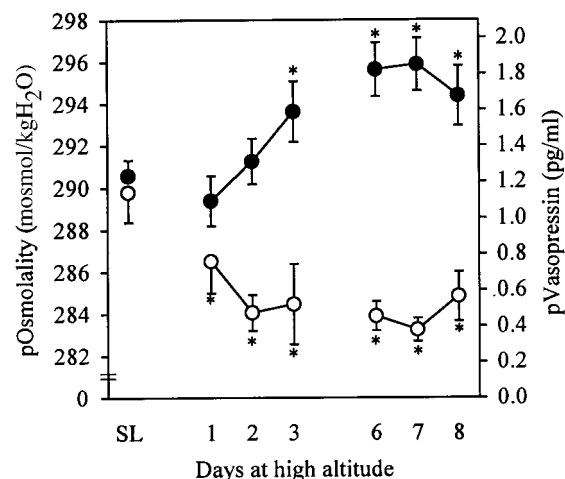


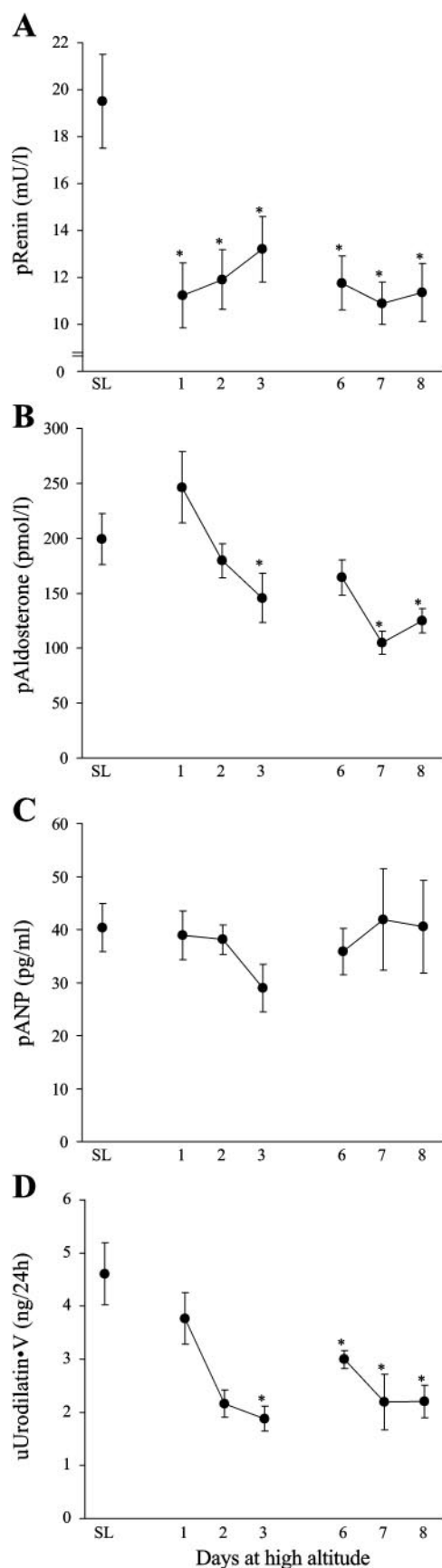
Fig. 2. Effects of high altitude on plasma osmolality (pOsmolality, ●) and plasma vasopressin concentration (pVasopressin, ○). Values are means \pm SE ($n = 8$). *Significant deviation from SL ($P < 0.05$).

dium concentrations decreased on days 1–3 but thereafter returned to a level not different from that at sea level. The same pattern was observed for plasma potassium concentration. Plasma creatinine concentrations were elevated on days 2–8 at altitude. Plasma albumin concentrations remained unchanged.

Urine flow rates increased on days 1 and 2, whereas sodium excretion rates remained unchanged in the presence of a reduction in potassium excretion (Table 3). On days 6 and 7, urinary excretions of sodium and water were elevated because of increased water and sodium intake during the investigation on day 6 (protocol 2). Albumin excretion rates more than doubled on days 1 and 2. Creatinine excretion rate remained unchanged, but because of the increase in plasma creatinine, renal clearance of creatinine decreased to a minimum of 96 ± 10 ml/min on day 7 (Table 3). Thus creatinine clearance, as determined by 24-h urine collections, was unchanged during the first 2 days at altitude but thereafter decreased significantly by 16–24%.

Hypertonic load. Heart rate and mean arterial blood pressure were increased by 24 and 30%, respectively, on day 6 compared with measurements at sea level (Table 4). Hypertonic saline loading had no effect on these increased heart rates and blood pressures.

Baseline values of plasma renin concentration were reduced by 28% at altitude. Hypertonic loading at sea level suppressed renin by 31%, but at altitude renin concentration remained unchanged (Fig. 4A). Baseline aldosterone concentrations were unchanged at altitude but were suppressed by saline loading in both environments by ~50% (Fig. 4B). Concentrations of endothelin-1 more than doubled at altitude, but in both environments they were unaffected by saline loading (Fig. 4C). Plasma ANP concentrations were unchanged at altitude compared with sea level but increased by 40–50% during saline infusions (Fig. 4D). Plasma concentration of norepinephrine was significantly increased on day 6 but remained unchanged during infusions (Fig. 4E).



Baseline values of plasma osmolality increased on *day 6* at altitude compared with sea level (from 293 ± 1 to 298 ± 1 mosmol/kgH₂O; Fig. 5A). In response to saline infusion, osmolality increased by very similar amounts (~ 5 mosmol/kgH₂O) in both environments but thereafter declined toward baseline level. Baseline vasopressin level was suppressed to 0.2 ± 0.1 pg/ml due to the sustained water loading inherent to the protocol but increased after hypertonic infusion to 0.6 pg/ml (Fig. 5B). Because of technical problems, vasopressin samples obtained at sea level did not provide meaningful results. Plasma sodium concentration increased and plasma albumin concentration decreased expectedly in response to saline loading in both environments (Table 5).

Free water clearance and urine excretion rate were high in the baseline periods due to water intake but decreased by $\sim 60\%$ after saline infusion (Figs. 5C and 6A). Responses at sea level and altitude were similar. Sodium excretion rates increased in response to hypertonic loading in both environments, but natriuretic response was significantly smaller at altitude (65 vs. 141%; Fig. 6B). Urodilatin excretion rate unexpectedly decreased after hypertonic infusion in both environments (Fig. 6C). However, measurement of urodilatin in the 24-h urine collections revealed that, on *day 6*, urodilatin excretion rates were significantly higher than on *days 2, 3, 7, and 8*. On *day 6*, when use of water diuresis during the four consecutive 1-h clearance periods is expected to have increased the precision of renal clearance measurements, creatinine clearance was consistently reduced by 24–36% compared with that at sea level (Table 6). Creatinine clearance was unaffected by saline infusion in both environments.

DISCUSSION

The present study demonstrates that hypoxemia modulates the normal relationship between plasma osmolality and plasma vasopressin so that the plasma osmolality-to-plasma vasopressin function curve is displaced to the right. However, the vasopressin response and the renal antidiuretic response to an osmotic stimulus are not attenuated at altitude in contrast to the acute natriuretic response to hypertonic sodium loading, which unexpectedly was reduced.

In agreement with previous investigations (13, 19, 29, 39, 53), we found that heart rate and blood pressure increased within 24 h and remained elevated during all 8 days at altitude. Recently, we reported that systolic blood pressure, heart rate, and plasma epinephrine increased on *day 1* at altitude but declined again on *day 5*, whereas diastolic and mean blood pressures continued to rise in parallel with plasma norepinephrine (19). Most probably, elevated blood pressure and

Fig. 3. Effects of high altitude on plasma concentrations of renin (pRenin; A), aldosterone (pAldosterone; B), atrial natriuretic peptide (pANP; C), and urinary excretion rate of urodilation (uUrodilatin·V; D). Values are means \pm SE ($n = 8$). *Significant deviation from SL ($P < 0.05$).

Table 2. *Effects of hypoxemia on hemoglobin and plasma concentrations of Na⁺, K⁺, creatinine, and albumin*

	Sea Level	Days at High Altitude					
		1	2	3	6	7	8
Hemoglobin, mmol/l	8.9 ± 0.2	9.6 ± 0.2*	9.6 ± 0.2*	9.8 ± 0.2*	9.4 ± 0.3*	9.4 ± 0.2	9.7 ± 0.3*
pNa ⁺ , mmol/l	140 ± 0	138 ± 1*	139 ± 1*	139 ± 0*	141 ± 0†	141 ± 1†	141 ± 1†
pK ⁺ , mmol/l	4.0 ± 0.1	3.8 ± 0.1*	3.9 ± 0.1	4.1 ± 0.1†	3.9 ± 0.0	4.0 ± 0.1†	4.3 ± 0.1*†
pCreatinine, μmol/l	88 ± 2	90 ± 3	95 ± 3*	99 ± 3*†	99 ± 4*†	101 ± 4*†	98 ± 4*†
pAlbumin, μmol/l	623 ± 10	623 ± 7	610 ± 9	607 ± 7	588 ± 15	584 ± 16	614 ± 14

Values are means ± SE ($n = 8$). pNa⁺, plasma Na⁺ concentration; pK⁺, plasma K⁺ concentration; pCreatinine, plasma creatinine concentration; pAlbumin, plasma albumin concentration. *Significantly different from sea level ($P < 0.05$). †Significantly different from day 1 at altitude ($P < 0.05$).

heart rate in hypoxemia is a result of increased peripheral resistance secondary to increased plasma levels of norepinephrine (19, 25, 29, 30, 36) and to augmented sympathetic nerve activity in hypoxemia (43, 44).

The prompt increase in hemoglobin concentration at altitude, which occurred long before the erythropoietic effect of hypoxemia could become significant, indicates the presence of hemoconcentration. Concomitantly with an increase in plasma osmolality of 5 mosmol/kgH₂O, body weight declined 2–3 kg during the 8 days at altitude, indicating a loss of body water. Previously, use of direct measurements in animals and humans have shown that hypoxemia decreases total body water, intracellular body water compartment, and plasma volume in the presence of an increase of the extracellular compartment (11, 18, 22). In part, hemoconcentration in hypoxemia may result from an increase in capillary permeability (14), which in renal glomerular capillaries results in a marked albuminuria (14), as also found in the present study (Table 3). Taken together, the results of the present study indicate that hemoconcentration occurred at altitude because of body water reduction and possibly also because of increased capillary permeability.

Creatinine clearance was significantly reduced after 3 days at altitude, suggesting a decrease in glomerular filtration rate (GFR). With the use of more ideal markers, we demonstrated previously that altitude hypoxemia for 24–48 h (at 4,350 m) increases renal vascular resistance and slightly decreases effective plasma flow without significant changes in GFR (29). The present results are in line with this but also suggest that more

sustained hypoxemia at this altitude eventually may decrease GFR. This might be caused by increased circulating levels of norepinephrine and endothelin and/or increased activity of renal sympathetic nerves, as indicated by direct measurements in animals (24, 47, 48). In agreement with previous studies (5, 27), we found that plasma endothelin-1 on day 6 was doubled compared with that at sea level. In high doses, endothelin-1 is known to be a very potent vasoactive peptide that causes systemic and renal vasoconstriction (50). However, short-term infusion of lower doses of endothelin-1, causing a doubling of the plasma concentration as we observed in the present study, did not increase blood pressure in humans (52) and dogs (45) and may even produce a natriuretic response (45) perhaps secondary to renal medullary vasodilation (12). It is, therefore, unknown to what extent long-term hypoxemia-induced increases in endothelin-1 are involved in changes of systemic and renal hemodynamics.

In agreement with previous investigations (1, 17, 29, 30), we found that hypoxemia reduced plasma concentrations of renin from day 1 and aldosterone from day 3. In vitro investigations have demonstrated that hypoxia directly inhibits aldosterone secretion from bovine adrenocortical cells (33), but the mechanism responsible for hypoxia-induced depression of renin secretion still remains unknown. Remarkably, reductions in resting values of renin and aldosterone occur despite an activation of the sympathetic nervous system, which under normal conditions is known to increase renin secretion via β -adrenergic stimulation. However, exercise-induced activation of the renin-aldosterone

Table 3. *Effects of hypoxemia on renal variables*

	Sea Level	Days at High Altitude					
		1	2	3	6	7	8
V, ml/24 h	1,884 ± 239	4,202 ± 677*	3,159 ± 360*	2,131 ± 298†	4,032 ± 565*	3,080 ± 255*	2,121 ± 236†
uNa ⁺ ·V, mmol/24 h	151 ± 16	186 ± 31	154 ± 26	126 ± 25†	308 ± 31*§	350 ± 32*†	260 ± 35*
uK ⁺ ·V, mmol/24 h	66 ± 9	47 ± 5	40 ± 4*	38 ± 4*	56 ± 6	52 ± 6	59 ± 4
Creatinine clearance, ml/min	122 ± 6	127 ± 5	124 ± 11	107 ± 7†	107 ± 8§	96 ± 10*†	110 ± 5
uCreatinine·V, mmol/24 h	15 ± 0	16 ± 1	17 ± 2	15 ± 1	15 ± 1	14 ± 1	15 ± 1
uAlbumin·V, mg/24 h	4.6 ± 0.9	11.3 ± 1.9*	9.2 ± 2.4*	8.2 ± 1.7	5.0 ± 0.5§	5.4 ± 2.0†	11.9 ± 2.4*
uOsmolality, mosmol/kgH ₂ O	578 ± 66	261 ± 71*	248 ± 35*	292 ± 41*	264 ± 48*	494 ± 29†	467 ± 50†
uOsmolality·V, mosM/24 h	940 ± 70	830 ± 65	727 ± 64	568 ± 61*†	1,138 ± 231	1,260 ± 62	936 ± 79

Values are means ± SE ($n = 8$). V, urine excretion rate; uNa⁺·V, Na⁺ excretion rate; uK⁺·V, K⁺ excretion rate; uCreatinine·V, creatinine excretion rate; uAlbumin·V, albumin excretion rate; uOsmolality, urine osmolality; uOsmolality·V, osmolar excretion rate. *Significantly different from sea level ($P < 0.05$). †Significantly different from day 1 at altitude ($P < 0.05$).

Table 4. *Effects of saline infusion on heart rate and blood pressure*

		Time After Infusion, min			
	Baseline	60	120	180	240
<i>Heart rate, beats/min</i>					
SL-C	47 ± 3	45 ± 3	45 ± 3	44 ± 2	45 ± 3
SL-S	47 ± 3	47 ± 2	46 ± 2	45 ± 2	45 ± 2
HA-S	61 ± 3*	58 ± 5*	55 ± 3*	58 ± 5*	58 ± 4*
<i>Mean arterial pressure, mmHg</i>					
SL-C	82 ± 3	81 ± 4	83 ± 2	85 ± 3	86 ± 2
SL-S	79 ± 2	82 ± 2	82 ± 3	80 ± 3	83 ± 2
HA-S	98 ± 4*	100 ± 3*	95 ± 3*	100 ± 4*	98 ± 4*
<i>Systolic blood pressure, mmHg</i>					
SL-C	111 ± 3	111 ± 4	111 ± 2	112 ± 3	115 ± 2
SL-S	110 ± 2	115 ± 3	114 ± 3	113 ± 3	116 ± 2
HA-S	124 ± 4*	126 ± 5*	122 ± 3*	123 ± 4*	126 ± 3*
<i>Diastolic blood pressure, mmHg</i>					
SL-C	68 ± 3	67 ± 4	69 ± 2	71 ± 3	72 ± 3
SL-S	64 ± 2	65 ± 2	66 ± 3	64 ± 2	67 ± 3
HA-S	85 ± 4*	86 ± 2*	81 ± 3*	88 ± 4*	84 ± 4*

Values are means ± SE ($n = 8$). SL-C/S, sea level control/saline infusion; HA-S, high-altitude saline infusion. *Significant difference between SL-S and HA-S ($P < 0.05$).

system is maintained in hypoxemia (30). Inhibition of the renin-aldosterone system during acclimatization to hypoxemia may protect against excessive retention of water and sodium and formation of edema, which characterize severe AMS and high-altitude pulmonary edema. The present results confirm that hypoxemia is associated with marked reductions in plasma renin and aldosterone concentrations.

Our results also demonstrate that hypoxemia modulates the normal relationship between plasma osmolality and plasma vasopressin concentration because vasopressin declined despite an increase in plasma osmolality during the 8 days at altitude. Therefore, the function curve of osmolality (abscissa) and vasopressin (ordinate) in plasma was shifted to the right, i.e., a given vasopressin concentration requires a higher osmolality compared with sea level. Vasopressin secretion is mainly regulated by osmoreceptors in the hypothalamus, but stimulation of baroreceptors in the arterial and venous systems by increases in blood pressure or volume may also change the relationship between osmolality and vasopressin (42). It is possible that vasopressin secretion during hypoxemia in this study was suppressed by the increase in systemic blood pressure. However, reduced sensitivity to changes in plasma osmolality or the expected elevation in plasma cortisol concentration may have contributed as well.

To test whether hypothalamic sensitivity to an osmotic load is reduced in hypoxemia, we investigated the endocrine and renal response to an infusion of hypertonic saline. Intravenous administration of hypertonic saline increases plasma osmolality and induces net transfer of water from the intra- to the extracellular space. Hypertonic saline infusion, therefore, provides an osmotic as well as a volume stimulus

to the body. During the time control experiment, no infusion was given, and the effects of hypertonic infusion can, therefore, be determined by comparison of the two responses at a given point in time. Hypertonic loading increased plasma osmolality by 5 mosmol/kgH₂O in both environments, indicating that the osmotic stimulus was the same, although baseline values of plasma osmolality were different. Because of sustained water loading, plasma vasopressin concentration was suppressed to 0.2 ± 0.1 pg/ml, but afterward saline infusion increased by a factor of 3 on day 6 at altitude. Unfortunately, in the present study, vasopressin could not be measured from sea level samples, but in another study from our laboratory (2) the vasopressin response to hypertonic saline loading at sea level (the same amount of sodium but given as 2.5% saline over 20 min) was equivalent to the response we found at altitude in the present study. Moreover, the antidiuretic response and the decline in free water clearance were the same in the two environments, indicating similar renal effects. Therefore, provided that renal sensitivity to vasopressin is not changed by hypoxemia, the results demonstrate that vasopressin secretion in response to an osmotic stimulus is not attenuated on day 6 at altitude. This is in contrast to the results reported by Ramirez et al. (36) showing that the expected increase in plasma vasopressin after infusion of hypertonic saline was abolished after 3–4 days at 3,000 m but not during short-term hypoxemia induced by breathing of hypoxic gas. A reduced vasopressin response to hypertonic saline infusion was also found in humans adapted to altitude living (35, 38). Results suggest that either the hypobaric condition or the longer period of hypoxemia was responsible for the suppression of the vasopressin response. Noteworthy

Table 5. *Effects of saline infusion on pNa^+ , pK^+ , $pCreatinine$ and $pAlbumin$*

Time After Infusion, min				
	Baseline	60	120	180
<i>pNa⁺, mmol/l</i>				
SL-C	140 ± 1	139 ± 1	138 ± 1*	138 ± 1*
SL-S	140 ± 1	142 ± 1*†	142 ± 1†	141 ± 1†
HA-S	141 ± 1	143 ± 0*	141 ± 1	140 ± 1
<i>pK⁺, mmol/l</i>				
SL-C	4.1 ± 0.1	4.1 ± 0.1	3.9 ± 0.1*	3.8 ± 0.1*
SL-S	4.3 ± 0.1	4.1 ± 0.1	4.2 ± 0.1†	4.1 ± 0.0†
HA-S	4.0 ± 0.1‡	3.8 ± 0.1*‡	3.9 ± 0.1‡	3.9 ± 0.1‡
<i>pCreatinine, μmol/l</i>				
SL-C	86 ± 3	82 ± 3*	82 ± 3*	81 ± 2*
SL-S	84 ± 3†	82 ± 3	82 ± 3	81 ± 3
HA-S	98 ± 3‡	97 ± 3‡	97 ± 3‡	95 ± 2*‡
<i>pAlbumin, μmol/l</i>				
SL-C	645 ± 19	647 ± 20	638 ± 19	640 ± 19
SL-S	635 ± 8	570 ± 6*†	580 ± 6*†	586 ± 9*†
HA-S	632 ± 13	560 ± 12*	567 ± 17*	569 ± 16*

Values are means ± SE ($n = 8$). *Significant difference from baseline; †significantly different between SL-C and SL-S; ‡significantly different between SL-S and HA-S ($P < 0.05$).

was the fact that the lack of vasopressin response had previously been associated with elevated levels of plasma cortisol (36, 37). Increased plasma cortisol levels have been suggested to be involved in hypoxemia-

induced inhibition of vasopressin secretion in dogs (34). In conscious dogs, sustained elevation of plasma cortisol decreased plasma vasopressin concentration (32) and attenuated the vasopressin response to hypertonic saline infusion (33) independent of changes in blood pressure and blood volume. In humans, plasma cortisol concentrations increase during the first days of hypoxemia (11, 15, 28, 40) but thereafter return to sea level values (11). In agreement with this, measurements in our laboratory's subjects (23) showed that plasma cortisol level tended to increase on *day 2* (from 201 ± 24 to 334 ± 75 nmol/l, $P = 0.078$) but not on *day 7* (232 ± 28 nmol/l) at 4,559 m. It is therefore possible that, in our study, increases in plasma cortisol during the first days in hypoxemia were involved in the inhibition of basal vasopressin secretion, but thereafter other mechanisms may have been involved. Possibly, the vasopressin response to hypertonic saline infusion was unchanged because plasma cortisol concentration on *day 6* had returned to that at sea level. Narrowing of the pulse pressure is known to be a codeterminant of vasopressin release. We found pulse pressure to be significantly increased on *day 1* but thereafter unchanged compared with sea level. All in all, the results suggest that the increase in arterial blood pressure was the main cause of the depression of basal vasopressin secretion. An increased pulse pressure and elevated cortisol levels, during the first days at altitude, might have augmented this effect, whereas hyperosmolality, on the other hand, may have had the opposite effect. Moreover, the possibility cannot be excluded that the effects of hypoxemia on baroreflex sensibility confounded the results. Hence, the net effect of these alterations may have been balanced so that basal vasopressin secretion in sustained hypoxemia was reduced.

Consistent with previous studies (1, 26, 29), sodium excretion rate remained unchanged at high altitude. However, the natriuretic response to hypertonic saline infusion was attenuated on *day 6* at high altitude. Increased activity of renal sympathetic nerves is expected to increase the reabsorption of sodium, whereas suppression of the renin-aldosterone system, as seen in hypoxemia, has a natriuretic effect. Hypoxemia suppressed baseline concentrations of plasma renin. This suggests that inhibition of angiotensin II-mediated reabsorption of sodium may have been involved in the maintenance of baseline natriuresis at altitude in the presence of the reduction of GFR. Plasma renin concentration declined expectedly in response to hypertonic loading at sea level but not at altitude perhaps

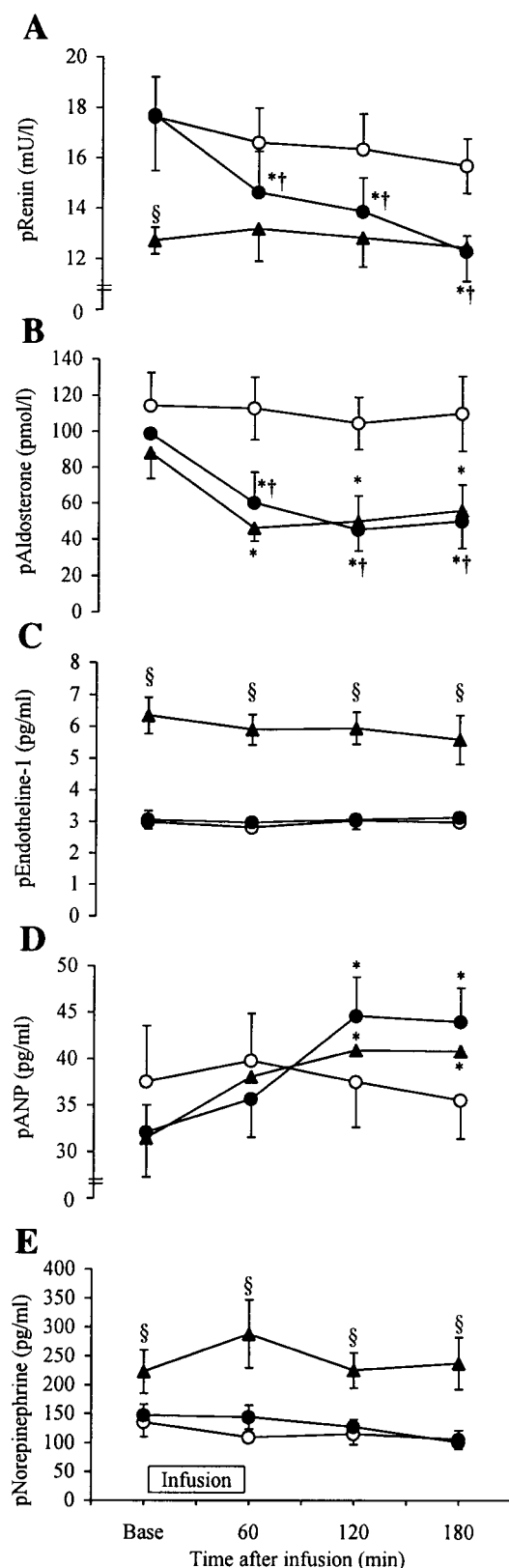


Fig. 4. Effects of saline infusion on pRenin (A), pAldosterone (B), plasma concentration of endothelin-1 (pEndothelin-1; C), pANP (D), and plasma concentration of norepinephrine (pNorepinephrine; E). \circ , Time control at sea level; \bullet , saline infusion at sea level; \blacktriangle , saline infusion at high altitude. Values are means \pm SE ($n = 8$). *Significant deviations from baseline ($P < 0.05$). †Significant difference between time control and saline infusion at SL ($P < 0.05$). §Significant difference between saline infusion at SL and at high altitude ($P < 0.05$).

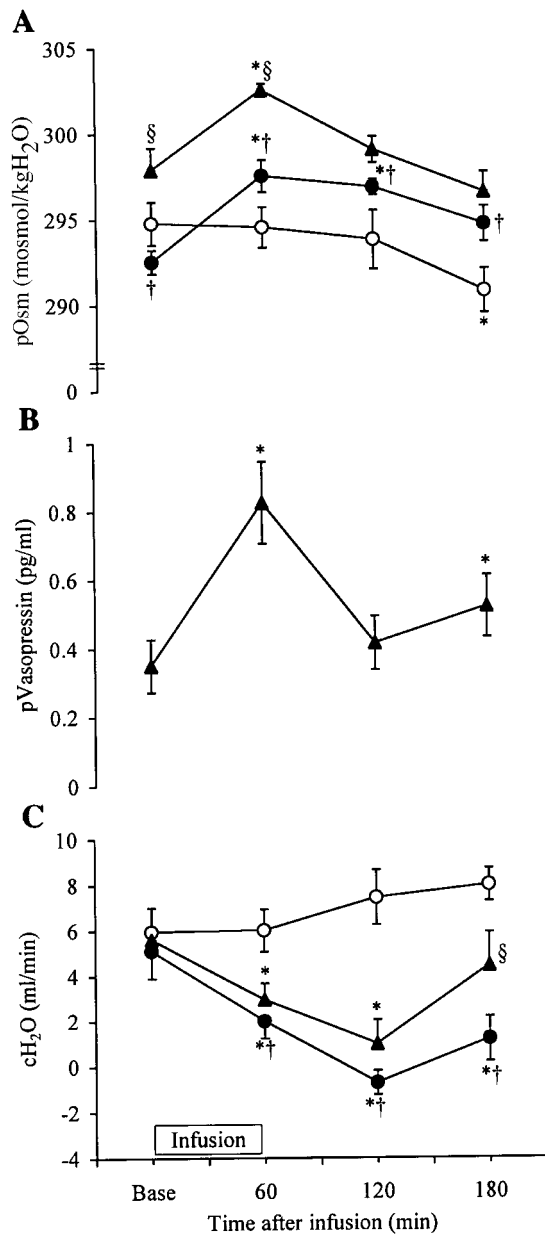


Fig. 5. Effects of saline infusion on plasma osmolality (pOsm; A), plasma vasopressin concentration (pVasopressin; B), and free water clearance (cH₂O; C). Values are means \pm SE ($n = 8$). \circ , Time control at SL; \bullet , saline infusion at sea level; \blacktriangle , saline infusion at high altitude. *Significant deviations from baseline ($P < 0.05$). †Significant difference between time control and saline infusion at SL ($P < 0.05$). §Significant difference between saline infusion at SL and at high altitude ($P < 0.05$).

because of marked inhibition by hypoxemia. A lack of additional inhibition of angiotensin II-mediated sodium reabsorption may be part of the attenuated natriuretic response to sodium loading at altitude. Effects of aldosterone and ANP probably were not involved because the responses to sodium loading remained unchanged at altitude. It cannot, however, be excluded that a diminished sodium intake due to AMS induced loss of appetite, blunted an increase in sodium excretion during the first days of hypoxemia, and contrib-

uted to attenuation of the natriuretic response to hypertonic infusion.

The natriuretic peptide, urodilatin, has been suggested to be a possible mediator of renal sodium excretion (7). With the use of a newly developed assay (3), we observed that 24-h excretion rates of urodilatin were attenuated at altitude. Thus urodilatin was probably

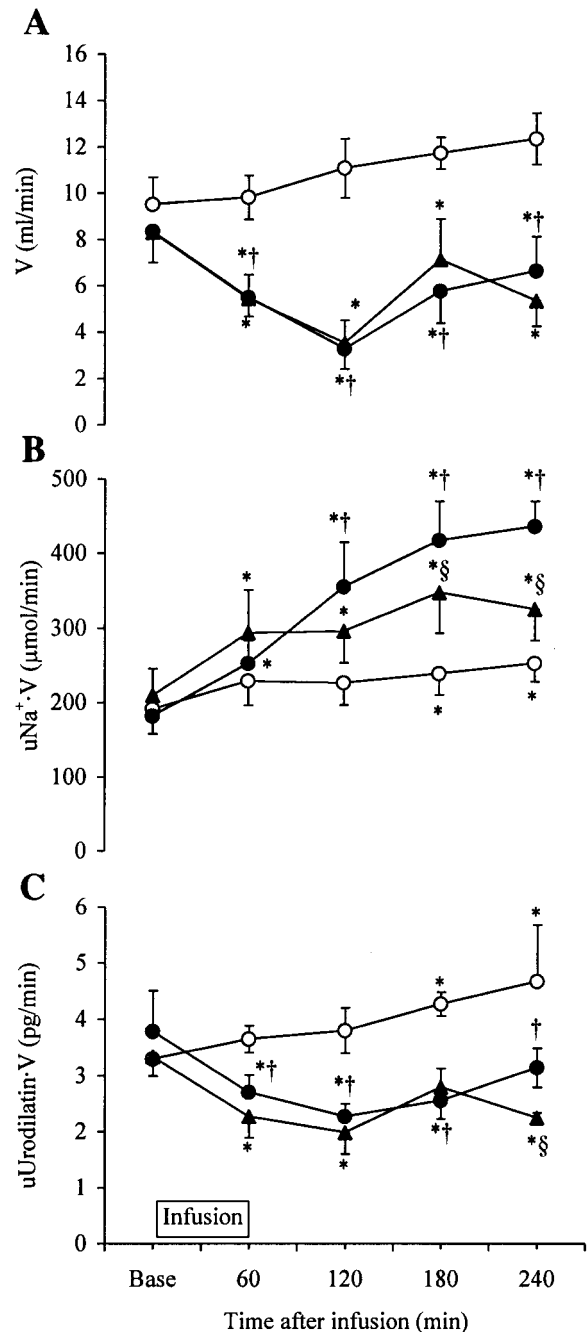


Fig. 6. Effects of saline infusion on urine excretion rate (V; A), sodium excretion rate (uNa⁺·V; B), and uRodilatin·V (C). Values are means \pm SE ($n = 8$). \circ , Time control at SL; \bullet , saline infusion at SL; \blacktriangle , saline infusion at high altitude. *Significant deviations from baseline ($P < 0.05$). †Significant difference between time control and saline infusion at SL ($P < 0.05$). §Significant difference between saline infusion at SL and at high altitude ($P < 0.05$).

Table 6. *Effects of saline infusion on renal variables*

		Time After Infusion, min			
	Baseline	60	120	180	240
<i>Creatinine clearance, ml/min</i>					
SL-C	137 ± 6	135 ± 6	128 ± 7	139 ± 6	
SL-S	142 ± 5	134 ± 5	137 ± 4	137 ± 7	
HA-S	90 ± 11‡	96 ± 7‡	88 ± 11‡	105 ± 9‡	
<i>uCreatinine·V, μmol/min</i>					
SL-C	11.7 ± 0.5	11.0 ± 0.4	10.4 ± 0.4	11.2 ± 0.5	10.8 ± 0.4
SL-S	12.0 ± 0.4	11.2 ± 0.2	11.4 ± 0.5	12.3 ± 1.4	12.8 ± 1.3
HA-S	8.9 ± 1.1‡	9.4 ± 0.7‡	8.6 ± 1.0§	10.0 ± 0.8‡	10.0 ± 0.6‡
<i>Osmolar clearance, ml/min</i>					
SL-C	3.6 ± 0.3	3.8 ± 0.2	3.6 ± 0.2	3.7 ± 0.2	
SL-S	3.2 ± 0.2	3.5 ± 0.3	4.0 ± 0.5*	4.6 ± 0.4*	
HA-S	2.7 ± 0.4	2.5 ± 0.3‡	2.5 ± 0.4‡	2.7 ± 0.3‡	
<i>uOsmolality, mosmol/kgH₂O</i>					
SL-C	118 ± 12	121 ± 12	104 ± 12	94 ± 8	88 ± 6
SL-S	192 ± 53	271 ± 35†	500 ± 69*†	404 ± 83*†	390 ± 88*†
HA-S	111 ± 18	158 ± 30	336 ± 89*‡	190 ± 59‡	271 ± 79*‡
<i>uOsmolality·V, μosM/min</i>					
SL-C	1,048 ± 74	1,121 ± 64	1,065 ± 51	1,080 ± 50	1,050 ± 55
SL-S	950 ± 69	1,039 ± 90	1,179 ± 139	1,336 ± 125*	1,404 ± 81*†
HA-S	805 ± 110	750 ± 82‡	759 ± 113‡	793 ± 103‡	939 ± 117‡
<i>uK⁺·V, μmol/min</i>					
SL-C	124 ± 7	138 ± 8	124 ± 10	106 ± 7	94 ± 6
SL-S	175 ± 41	232 ± 30†	332 ± 41*†	307 ± 48*†	271 ± 45†
HA-S	58 ± 15‡	44 ± 12‡	42 ± 4‡	42 ± 7‡	48 ± 8‡
<i>uAlbumin·V, μg/min</i>					
SL-C	3.4 ± 0.3	3.2 ± 0.3	3.6 ± 0.6	3.1 ± 0.3	3.0 ± 0.3
SL-S	2.3 ± 0.5	3.0 ± 1.0	5.4 ± 1.8	2.6 ± 1.0	3.3 ± 0.6
HA-S	4.1 ± 1.1	2.4 ± 0.6	3.1 ± 0.7	5.6 ± 2.4	3.2 ± 0.7

Values are means ± SE (*n* = 8). uOsmolality, urine osmolality; uOsmolality·V, osmolar excretion rate. *Significant difference from baseline; †significantly different between SL-C and SL-S; ‡significantly different between SL-S and HA-S (*P* < 0.05).

not responsible for maintaining natriuresis in hypoxemia, but it is possible that depression of the baseline values was involved in the attenuated natriuretic response to sodium loading. Sodium loading on *day 6* increased the 24-h renal excretion rate of urodilatin compared with at *days 2, 3, 7, and 8* (*P* < 0.05). However, in the four 1-h observation periods during and after hypertonic sodium loading, urodilatin excretion did not increase, indicating that urodilatin-induced sodium excretion, in the present situation, could be a slow-acting system, effective only several hours after hypertonic load. This attenuation of 24-h urodilatin excretion and the blunted urodilatin response to hypertonic loading were also observed during prolonged bed rest (2). It is, therefore, possible that the body fluid volume, which is reduced during bed rest and possibly also during sustained hypoxemia, is important for the urodilatin response to acute sodium loading. Also, it cannot be excluded that an increase in urodilatin excretion rate was not detected because urodilatin concentrations in urine, due to sustained water diuresis, were close to the detection limit of the assay.

Thus sodium excretion was unchanged at altitude despite reductions in creatinine clearance due, per-

haps, to hypoxemia-induced inhibition of the renin-aldosterone system. The natriuretic response to hypertonic sodium loading was attenuated at altitude, possibly due to lack of additional inhibition of the renin-aldosterone system and maybe also because of a reduction in baseline urodilatin synthesis.

The results demonstrate that several days of hypoxemia lead to hypovolemia, hyperosmolality, elevated blood pressure, reduced GFR and urodilatin synthesis, inhibition of the renin-angiotensin-aldosterone system in the presence of reduced plasma levels of vasopressin, and increased levels of circulating norepinephrine and endothelin-1. Hypovolemia and hyperosmolality may, at least in part, be secondary to hypertension-induced suppression of vasopressin secretion. The vasopressin response and the renal antidiuretic response to hypertonic saline loading were preserved in sustained hypoxemia, but the natriuretic response was attenuated.

In conclusion, chronic hypobaric hypoxemia 1) elevates the set point of the plasma osmolality-to-plasma vasopressin relationship, possibly due to the concurrent hypertension, thereby causing hypovolemia and hyperosmolality, and 2) blunts the natriuretic response to hypertonic volume expansion possibly due to ele-

vated circulating levels of norepinephrine and endothelin, reduced GFR and urodilatin synthesis, or attenuated inhibition of the renin system.

Perspectives. The present study demonstrates that hypobaric hypoxemia induces hypovolemia and hyperosmolality due to suppression of vasopressin release. Although the antidiuretic response to saline loading was unchanged at altitude, it is possible, however, that reduced renal sensitivity to vasopressin is also involved in the loss of body water. This is indicated by previous studies where hypoxemia-induced diuresis was observed concomitantly with unchanged or increased plasma levels of vasopressin (15, 51). This question should be addressed by comparison of the renal responses to vasopressin infusion at sea level and at high altitude.

We confirmed that hypoxemia induces hypertension concomitantly with elevated levels of plasma norepinephrine. Moreover, we found a 100% increase of the plasma endothelin-1 concentration. Recently, we reported that prolonged hypoxemia elicited an augmented vasoactive response to an adrenergic activation such as that elicited by local cooling (19). Thoracic fluid index increased with cooling, suggesting an increase in pulmonary vascular resistance that pointed to a possible mechanism behind the development of high-altitude pulmonary edema. The possible involvement of endothelins and other vasoactive agents in this response remains hypothetical.

The blunted natriuretic response to hypertonic volume expansion in hypoxemia observed in the present study could be explained by elevated circulating levels of norepinephrine and endothelin, reduced GFR and urodilatin synthesis, or absence of renin system inhibition. Further studies, however, are required to explain to what extent each of these components are involved in the altered regulation of sodium excretion in hypoxemia.

We thank Bodil Svendsen, Inge Hornung Pedersen, Barbara Sørensen, Birthe Lynderup, Trine Eidsvold, and Sigurd Kramer Hansen for expert technical assistance, and Dr. Svend Strandgaard (Department of Nephrology, Herlev Hospital) for measurement of albumin in urine.

This study was supported by grants from the Danish Heart Foundation, Novo Nordisk Foundation, Grocer Knud Oester-Joergensen and wife, Maria Oester-Joergensens Foundation for Medical Research, and The King Christian 10th Foundation.

REFERENCES

1. Bartsch PS, Shaw M, Franciolli MP, Gnadinger, and Weidmann P. Atrial natriuretic peptide in acute mountain sickness. *J Appl Physiol* 65: 1929–1937, 1988.
2. Bestle MH, Norsk P, and Bie P. Volume and osmoregulation in humans after a week of head-down bed rest. *Am J Physiol Regulatory Integrative Comp Physiol* 281: R310–R317, 2001.
3. Bestle MH, Olsen NV, Christensen P, Jensen BV, and Bie P. Cardiovascular, endocrine, and renal effects of urodilatin in normal humans. *Am J Physiol Regulatory Integrative Comp Physiol* 276: R684–R695, 1999.
4. Blume FD, Boyer SJ, Braverman LE, Cohen A, Dirkse J, and Mordes JP. Impaired osmoregulation at high altitude. Studies on Mt. Everest. *JAMA* 252: 524–526, 1984.
5. Cargill RI, Kiely DG, Clark RA, and Lipworth BJ. Hypoxaemia and release of endothelin-1. *Thorax* 50: 1308–1310, 1995.
6. Dela F, Mikines KJ, von Linstow M, Secher NH, and Galbo H. Effect of training on insulin-mediated glucose uptake in human muscle. *Am J Physiol Endocrinol Metab* 263: E1134–E1143, 1992.
7. Drummer C, Gerzer R, Heer M, Molz B, Bie P, Schlossberger M, Stadaeger C, Rocker L, Strollo F, Heyduck B, et al. Effects of an acute saline infusion on fluid and electrolyte metabolism in humans. *Am J Physiol Renal Fluid Electrolyte Physiol* 262: F744–F754, 1992.
8. Emmeluth C and Bie P. Effects, release and disposal of endothelin-1 in conscious dogs. *Acta Physiol Scand* 146: 197–204, 1992.
9. Emmeluth C, Drummer C, Gerzer R, and Bie P. Natriuresis in conscious dogs caused by increased carotid $[Na^+]$ during angiotensin II and aldosterone blockade. *Acta Physiol Scand* 151: 403–411, 1994.
10. Feldt Rasmussen B, Dinesen B, and Deckert M. Enzyme immunoassay: an improved determination of urinary albumin in diabetics with incipient nephropathy. *Scand J Clin Lab Invest* 45: 539–544, 1985.
11. Frayser R, Rennie ID, Gray GW, and Houston CS. Hormonal and electrolyte response to exposure to 17,500 ft. *J Appl Physiol* 38: 636–642, 1975.
12. Gurbanov K, Rubinstein I, Hoffman A, Abassi Z, Better OS, and Winaver J. Differential regulation of renal regional blood flow by endothelin-1. *Am J Physiol Renal Fluid Electrolyte Physiol* 271: F1166–F1172, 1996.
13. Hansen JM, Kanstrup IL, Richalet JP, and Olsen NV. High altitude-induced albuminuria in normal man is enhanced by infusion of low-dose dopamine. *Scand J Clin Lab Invest* 56: 367–372, 1996.
14. Hansen JM, Olsen NV, Feldt Rasmussen B, Kanstrup IL, Dechaux M, Dubray C, and Richalet JP. Albuminuria and overall capillary permeability of albumin in acute altitude hypoxia. *J Appl Physiol* 76: 1922–1927, 1994.
15. Heyes MP, Farber MO, Manfredi F, Robertshaw D, Weinberger M, Fineberg N, and Robertson G. Acute effects of hypoxia on renal and endocrine function in normal humans. *Am J Physiol Regulatory Integrative Comp Physiol* 243: R265–R270, 1982.
16. Hildebrandt W, Ottenbacher A, Schuster M, Swenson E, and Bartsch P. Diuretic effect of hypoxia, hypocapnia, and hyperpnea in humans: relation to hormones and O_2 chemosensitivity. *J Appl Physiol* 88: 599–610, 2000.
17. Hogan RP 3rd, Kotchen TA, Boyd AE 3rd, and Hartley LH. Effect of altitude on renin-aldosterone system and metabolism of water and electrolytes. *J Appl Physiol* 35: 385–390, 1973.
18. Jain SC, Bardhan J, Swamy YV, Krishna B, and Nayar HS. Body fluid compartments in humans during acute high-altitude exposure. *Aviat Space Environ Med* 51: 234–236, 1980.
19. Kanstrup IL, Poulsen TD, Hansen JM, Andersen LJ, Bestle MH, Christensen NJ, and Olsen NV. Blood pressure and plasma catecholamines in acute and prolonged hypoxia: effects of local hypothermia. *J Appl Physiol* 87: 2053–2058, 1999.
20. Kjaer A, Knigge U, Rouleau A, Garbarg M, and Warberg J. Dehydration-induced release of vasopressin involves activation of hypothalamic histaminergic neurons. *Endocrinology* 135: 675–681, 1994.
21. Koller EA, Buhner A, Felder L, Schopen M, and Vallotton MB. Altitude diuresis: endocrine and renal responses to acute hypoxia of acclimatized and non-acclimatized subjects. *Eur J Appl Physiol* 62: 228–234, 1991.
22. Krzywicki HJ, Consolazio CF, Johnson HL, Nielsen WC Jr, and Barnhart RA. Water metabolism in humans during acute high-altitude exposure (4,300 m). *J Appl Physiol* 30: 806–809, 1971.
23. Larsen JJ, Hansen JM, Olsen NV, Galbo H, and Dela F. The effect of altitude hypoxia on glucose homeostasis in men. *J Physiol (Lond)* 504: 241–249, 1997.
24. Malpas SC, Shweta A, Anderson WP, and Head GA. Functional response to graded increases in renal nerve activity during hypoxia in conscious rabbits. *Am J Physiol Regulatory Integrative Comp Physiol* 271: R1489–R1499, 1996.

25. **Mazzeo RS, Brooks GA, Butterfield GE, Podolin DA, Wolfel EE, and Reeves JT.** Acclimatization to high altitude increase muscle sympathetic activity both at rest and during exercise. *Am J Physiol Regulatory Integrative Comp Physiol* 269: R201–R207, 1995.
26. **Milledge JS, Beeley JM, McArthur S, and Morice AH.** Atrial natriuretic peptide, altitude and acute mountain sickness. *Clin Sci (Colch)* 77: 509–514, 1989.
27. **Morganti A, Giussani M, Sala C, Gazzano G, Marana I, Pierini A, Savoia MT, Ghio F, Cogo A, and Zanchetti A.** Effects of exposure to high altitude on plasma endothelin-1 levels in normal subjects. *J Hypertens* 13: 859–865, 1995.
28. **Okazaki S, Tamura Y, Hatano T, and Matsui N.** Hormonal disturbances of fluid-electrolyte metabolism under altitude exposure in man. *Aviat Space Environ Med* 55: 200–205, 1984.
29. **Olsen NV, Hansen JM, Kanstrup IL, Richalet JP, and Leyssac PP.** Renal hemodynamics, tubular function, and response to low-dose dopamine during acute hypoxia in humans. *J Appl Physiol* 74: 2166–2173, 1993.
30. **Olsen NV, Kanstrup IL, Richalet JP, Hansen JM, Plazen G, and Galen FX.** Effects of acute hypoxia on renal and endocrine function at rest and during graded exercise in hydrated subjects. *J Appl Physiol* 73: 2036–2043, 1992.
31. **Papanek PE and Raff H.** Physiological increases in cortisol inhibit basal vasopressin release in conscious dogs. *Am J Physiol Regulatory Integrative Comp Physiol* 266: R1744–R1751, 1994.
32. **Papanek PE and Raff H.** Chronic physiological increases in cortisol inhibit the vasopressin response to hypertonicity in conscious dogs. *Am J Physiol Regulatory Integrative Comp Physiol* 267: R1342–R1349, 1994.
33. **Raff H, Ball DL, and Goodfriend TL.** Low oxygen selectively inhibits aldosterone secretion from bovine adrenocortical cells in vitro. *Am J Physiol Endocrinol Metab* 256: E640–E644, 1989.
34. **Raff H, Shinsako J, Keil LC, and Dallman MF.** Feedback inhibition of adrenocorticotropin and vasopressin responses to hypoxia by physiological increases in endogenous plasma corticosteroids in dogs. *Endocrinology* 114: 1245–1249, 1984.
35. **Ramirez G, Bittle PA, Agosti SJ, Dietz J, and Colice GL.** Salt loading test in a population adapted to moderately high altitude living (3,000 m). *Aviat Space Environ Med* 64: 831–838, 1993.
36. **Ramirez G, Hammond M, Agosti SJ, Bittle PA, Dietz JR, and Colice GL.** Effects of hypoxemia at sea level and high altitude on sodium excretion and hormonal levels. *Aviat Space Environ Med* 63: 891–898, 1992.
37. **Ramirez G, Herrera R, Pineda D, Bittle PA, Rabb HA, and Bercu BB.** The effects of high altitude on hypothalamic-pituitary secretory dynamics in men. *Clin Endocrinol (Oxf)* 43: 11–18, 1995.
38. **Ramirez G, Pineda DO, Bittle PA, Agosti SJ, Rabb HA, and Dietz JR.** Salt excretory capacity in natives adapted to moderate high altitude living after acute mobilization to sea level. *Aviat Space Environ Med* 66: 1063–1070, 1995.
39. **Reeves JT, Mazzeo RS, Wolfel EE, and Young AJ.** Increased arterial pressure after acclimatization to 4300 m: possible role of norepinephrine. *Int J Sports Med* 13 Suppl 1: 1992.
40. **Richalet JP, Rutgers V, Bouchet P, Rymer JC, Keromes A, Duval Arnould G, and Rathat C.** Diurnal variations of acute mountain sickness, colour vision, and plasma cortisol and ACTH at high altitude. *Aviat Space Environ Med* 60: 105–111, 1989.
41. **Roach RC, Bartsch P, Hackett P, and Oelz O.** The Lake Louise acute mountain sickness scoring system. In: *Hypoxia and Molecular Medicine*, edited by Sutton JR, Houston CS, and Coates G. Burlington, VT: Queen City, 1993, p. 272–274.
42. **Robertson GL and Berl T.** Pathophysiology of water metabolism. In: *The Kidney*, edited by Brenner BM. Philadelphia, PA: Saunders, 1996, p. 873–929.
43. **Rowell LB, Johnson DG, Chase PB, Comess KA, and Seals DR.** Hypoxemia raises muscle sympathetic activity but not norepinephrine in resting humans. *J Appl Physiol* 66: 1736–1743, 1989.
44. **Saito M, Mano T, Iwase S, Koga K, Abe H, and Yamazaki Y.** Responses in muscle sympathetic activity to acute hypoxia in humans. *J Appl Physiol* 65: 1548–1552, 1988.
45. **Sandgaard NC and Bie P.** Natriuretic effect of non-pressor doses of endothelin-1 in conscious dogs. *J Physiol (Lond)* 494: 809–818, 1996.
46. **Schutten HJ, Johannessen AC, Torp-Pedersen C, Sander-Jensen K, Bie P, and Warberg J.** Central venous pressure: a physiological stimulus for secretion of atrial natriuretic peptide in humans? *Acta Physiol Scand* 131: 265–272, 1987.
47. **Shibamoto T, Uematsu H, Matsuda Y, Fujita T, Sawano F, Saeki Y, Hayashi T, and Koyama S.** Acute effect of hypobaria and hypoxia on renal nerve activity in anaesthetized rabbits. *Acta Physiol Scand* 144: 47–53, 1992.
48. **Shirai M, Matsukawa K, Nishiura N, Kawaguchi AT, and Ninomiya I.** Changes in efferent pulmonary sympathetic nerve activity during systemic hypoxia in anesthetized cats. *Am J Physiol Regulatory Integrative Comp Physiol* 269: R1404–R1409, 1995.
49. **Simon D, Hartmann DJ, Badouaille G, Caillot G, Guyenne TT, Corvol P, Pau B, and Marchand J.** Two-site direct immunoassay specific for active renin. *Clin Chem* 38: 1959–1962, 1992.
50. **Sorensen SS, Madsen JK, and Pedersen EB.** Systemic and renal effect of intravenous infusion of endothelin-1 in healthy human volunteers. *Am J Physiol Renal Fluid Electrolyte Physiol* 266: F411–F418, 1994.
51. **Swenson ER, Duncan TB, Goldberg SV, Ramirez G, Ahmad S, and Schoene RB.** Diuretic effect of acute hypoxia in humans: relationship to hypoxic ventilatory responsiveness and renal hormones. *J Appl Physiol* 78: 377–383, 1995.
52. **Vierhapper H, Wagner O, Nowotny P, and Waldhausl W.** Effect of endothelin-1 in man. *Circulation* 81: 1415–1418, 1990.
53. **Vogel JA and Harris CW.** Cardiopulmonary responses of resting man during early exposure to high altitude. *J Appl Physiol* 22: 1124–1128, 1967.