Cardiovascular autonomic modulation and activity of carotid baroreceptors at altitude

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A B S T R A C T

1. To assess the effects of acute exposure to high altitude on baroreceptor function in man we evaluated the effects of baroreceptor activation on R–R interval and blood pressure control at high altitude. We measured the low-frequency (LF) and high-frequency (HF) components in R–R, non-invasive blood pressure and skin blood flow, and the effect of baroreceptor modulation by 0.1-Hz sinusoidal neck suction. Ten healthy sea-level natives and three high-altitude native, long-term sea-level residents were evaluated at sea level, upon arrival at 4970 m and 1 week later.

2. Compared with sea level, acute high altitude decreased R–R and increased blood pressure in all subjects [sea-level natives: R–R from 1002 ± 45 to 775 ± 57 ms, systolic blood pressure from 130 ± 3 to 150 ± 8 mmHg; high-altitude natives: R–R from 809 ± 116 to 749 ± 47 ms, systolic blood pressure from 110 ± 12 to 125 ± 11 mmHg ($P < 0.05$ for all)]. One week later systolic blood pressure was similar to values at sea level in all subjects, whereas R–R remained elevated in sea-level natives. The low-frequency power in R–R and systolic blood pressure increased in sea-level natives [R–R-LF from 47 ± 8 to $65 \pm 10\%$ ($P < 0.05$), systolic blood pressure-LF from 1.7 ± 0.3 to 2.6 ± 0.4 ln-mmHg^2 ($P < 0.05$)], but not in high-altitude natives (R–R-LF from 32 ± 13 to $38 \pm 19\%$, systolic blood pressure-LF from 1.9 ± 0.5 to 1.7 ± 0.8 ln-mmHg^2). The R–R-HF decreased in sea-level natives but not in high-altitude natives, and no changes occurred in systolic blood pressure-HF. These changes remained evident 1 week later. Skin blood flow variability and its spectral components decreased markedly at high altitude in sea-level natives but showed no changes in high-altitude natives. Neck suction significantly increased the R–R- and systolic blood pressure-LF in all subjects at both sea level and high altitude.

3. High altitude induces sympathetic activation in sea-level natives which is partially counteracted by active baroreflex. Despite long-term acclimatization at sea level, high-altitude natives also maintain active baroreflex at high altitude but with lower sympathetic activation, indicating a persisting high-altitude adaptation which may be genetic or due to baroreflex activity not completely lost by at least 1 year's sea-level residence.

INTRODUCTION

Hypoxia (due to endogenous or atmospheric causes) not only affects ventilation, but also greatly influences autonomic cardiovascular regulation in normal as well as in pathological conditions, such as chronic cardiac and respiratory failure. Although in the latter pathological conditions the effects of chronic hypoxia superimpose and complicate other aspects of the underlying disease, exposure of healthy subjects to high altitude provides a

Key words: autonomic nervous system, baroreflex, heart rate variability, high altitude.

Abbreviations: HF, high frequency; LF, low frequency; SBF, skin blood flow.

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unique model to study specific adaptations to acute and prolonged hypoxia independently of pre-existing pathology.

Exposure to high altitude activates the sympathoadrenal system, but the time course and mechanisms of this activation are not well established. Urinary or blood catecholamine levels have shown a progressive increase in noradrenaline during the first few days at altitude [1–4]. In contrast, blood levels of adrenaline increase upon initial arrival at altitude and then decrease slightly, although urinary excretion is unchanged by altitude exposure [1–4]. These sympathoadrenal responses can influence cardiovascular (blood pressure, heart rate and vascular resistance) and metabolic (lactate, glucose and lipid) adaptations associated with a short- or long-term high-altitude stay. Increases in blood pressure, heart rate and systemic vascular resistance have been reported, and these increases were correlated with the increase in noradrenaline excretion during the first 8 days of altitude exposure [1].

Less information exists on the autonomic neural response to high-altitude exposure. Changes in heart rate variability based on 24-h Holter monitoring as an indirect index of autonomic modulation or on analysis of short-term R–R interval recordings have been reported [5,6]. These studies, however, ignored the effects of respiration on heart rate variability. The autonomic modulation of the blood pressure and of the microcirculation on high-altitude exposure has not been studied, and the relative contribution of sympathetic or vagal activity to cardiovascular function in this hypoxic environment remains unknown. Moreover, the activity of the baroreceptors in cardiovascular control at altitude has not been investigated.

We have shown that sinusoidal neck suction provides a simple, non-invasive method to selectively stimulate the carotid baroreceptors, and to observe their effects on both the heart rate and the arterial system (blood pressure and arterioles of skin microcirculation) in several different physiological or pathological conditions [7–9].

Although Himalayan natives who also reside at high altitude might have a different autonomic activation pattern [10], it is not known to what extent this is the result of long-term adaptation to the hypoxic environment or of a genetic advantage manifest as an intrinsic capability to adapt better and faster on acute altitude exposure.

We therefore evaluated the effect of high altitude on carotid baroreceptors and autonomic modulation of the heart, blood pressure and microcirculation in order to assess if the baroreflex is still active in sea-level natives acutely exposed to high altitude. We also compared the effects of acute altitude exposure in sea-level residents and in a small group of high-altitude natives resident and acclimatized to sea level.

METHODS

Ten healthy non-smoking subjects who were sea-level natives and living at sea level in Italy, Switzerland and the U.S.A., aged 45.9 ± 5.3 years (range 24–70 years), participated in the study. We also evaluated three healthy non-smoking subjects aged 24.3 ± 4.8 (19–34) years who were high-altitude natives (i.e. were born and had lived at altitudes between 3000 and 3800 m in Ladakh, a region of Indian Himalaya) but resident in New Delhi (sea level) for at least 1 year. None of the subjects was affected by either cardiovascular or pulmonary disease. The investigation conformed with the principles outlined in the Declaration of Helsinki, and the protocol was approved by the ethics committee of the University of New Mexico at Albuquerque, U.S.A. All subjects gave their informed consent. Recordings were obtained at sea level (New Delhi) and in the mountains of Zanskar (Ladakh) at 4970 m, reached 3 days after the sea-level recordings. All subjects were studied 1 day after arrival at 4970 m, and 1 week later at the same altitude. The ambient temperature at sea level was maintained at 20 °C by air conditioning. At high altitude the temperature in the study tent was kept close to 20 °C by controlling the openings of the tent. The ambient temperatures at high altitude did not differ by more than 2 °C from those observed at sea level.

After a variable period of 20–30 min of sitting and familiarization with the environment, we recorded the ECG (lead II), respiration, non-invasive blood pressure and neck chamber pressure. The respiratory signal was obtained simultaneously from ECG electrodes by means of an electrical impedance pneumograph designed in our laboratory (with a flat frequency response from 0 to 25 Hz) [11]. Non-invasive recordings of blood pressure were also obtained by a continuous non-invasive device (CBM 3000 tonometer, Colin®, San Antonio, TX, U.S.A.) and by a conventional sphygmomanometer. The Colin® tonometer had the advantage of using the radial artery rather than the smaller digital arteries, which are used to obtain non-invasive signals by other instruments. These smaller arteries are markedly constricted by acute altitude exposure and signals are usually lost or difficult to obtain.

The skin arteriolar blood flow (SBF) was evaluated by IR photoplethysmography measured on the volar surface of the right index finger. This simple device, which measures the fluctuations of a 1-mW 950 nm IR light back-scattered from the erythrocytes moving in the skin vessels, particularly the arterioles, is able to detect spontaneous fluctuations in skin arteriolar circulation. The theory and applications of this device have been extensively reviewed [12,13], and it is now accepted that the photoplethysmograph is sensitive to a combination of both volume and flow in the most superficial (in the range of 1 mm) layer of the skin. The arterioles and/or the arterio-venous anastomoses are the main deter-

minants of the SBF signal [14–17]. Using spectral analysis techniques, we have shown that spontaneous fluctuations of the skin microcirculation are closely related to the oscillations of the R–R interval and respiration, and the LF (around 0.1 Hz) oscillations of the skin circulation are modified by changes in sympathetic activity [7,18]. Others have reported that sympathetic activity also affects the LF oscillations in blood pressure [19,20].

The neck suction was applied by means of a semi-flexible plastic collar connected to a vacuum cleaner whose power was modulated by a computer-driven phase-control power unit via a digital-to-analog interface built in our laboratory [8,9]. By selecting the appropriate signal amplitude and frequency we could then obtain a sinusoidal suction with the desired characteristics. The neck pressure was continuously monitored by a piezo-resistive differential pressure transducer (part no. 286–692, RS, Corby, U.K.). The sinusoidal suction was set to oscillate from 0 to −30 mmHg neck chamber pressure. By cyclical stimulation of the carotid baro-receptors, neck suction-synchronous reflex oscillations could be observed in a large number of signals, including R–R interval, blood pressure, microcirculation and muscle sympathetic nerve activity, with only minor changes in mean values [7]. An increase in fluctuations (at the same frequency of the stimulus) from no neck suction to neck suction was considered as evidence of activity of the carotid baroreceptors on each signal. This technique presents several advantages with respect to other methods used to evaluate the baroreflex, including the non-invasiveness, the absence of side effects connected to use of drugs, and the possibility to evaluate both the response to the heart and to the blood pressure of the baroreflex. Possible disadvantages are that sinusoidal neck suction is selective and stimulates only the carotid baroreceptors; in addition, it requires a fixed rate of breathing in order to avoid superimposition of respiratory- and neck suction-induced oscillations (due to occasional slow breathing), which would have been impossible to distinguish. This methodology also differs from other techniques of neck suction, which normally use impulses rather than sinusoids.

Respiration was thus maintained at 0.25 Hz (15 breaths/min) and the sinusoidal neck suction was set at 0.10 Hz (6 cycles/min, LF stimulation). No instructions were given to maintain a given depth of breathing, in order to allow each subject to maintain adequate ventilation. Therefore, the purpose of controlling the breathing rate was essentially to avoid spurious slow breaths that would have created spurious respiratory slow fluctuations in the other signals. In our experience, this procedure has the effect of eliminating artefacts in the interpretations of spectral analysis data and also maintains constant frequency and depth of respiration during the recording, without alterations in the autonomic pattern [21]. After a 4-min recording during controlled respiration only, LF neck suction stimulation was performed for periods of 2 min. Due to an equipment failure, the second evaluation at altitude (subacute altitude exposure) could not include neck suction stimulation.

The data were digitized on line by a 12-bit analogue-to-digital converter built in our laboratory, at a sampling rate of 300 samples/s for the ECG, 90 samples/s for the blood and neck pressures, and 45 samples/s for respiration and SBF. The converter was connected to a Macintosh Powerbook 170 portable computer (Apple Inc., Cupertino, CA, U.S.A.) via RS-232 serial interface. A 'C' language programme identified all the QRS complexes in each sequence and then located the peak of each R wave. The R–R interval, systolic and diastolic pressure, neck suction, respiration and photoplethysmographic time series were obtained from these data. The respiratory time series was obtained from the respiration signal at the peak of each R wave, and was expressed in arbitrary values. For each step of the protocol, 250–350 R–R intervals (this variability was due to different heart rates present at sea level and at altitude) for the 4-min recordings and 100–150 R–R intervals for the 2-min recordings during neck suction were analysed. Premature beats were interactively identified and corrected by linear interpolation with the previous and subsequent beats. The original signals and time series were then stored for further analysis for each signal, including mean signal, signal variability (evaluated as the S.D.) and auto-regressive power spectrum analysis.

We applied power spectrum analysis to R–R interval, respiratory, systolic and diastolic blood pressure, SBF and neck suction signals, using an autoregressive model similar to previous studies [11]. In most cases a model order of 11–13 was found to be adequate. Spectral components were obtained by a decomposition method to measure the area (i.e. the power) below each spectral peak [11]. According to the autoregressive model, each peak identifies the presence of an oscillatory component; however, in order to distinguish between signal and noise, only components above 5% of total variability were considered as present. Two frequency bands were considered: the so-called low-frequency (LF) band (0.03–0.15 Hz) and the respiratory (high) frequency (HF) band (0.18–0.35 Hz). Having acquired the respiratory signal, we checked whether the respiration was in fact appropriately performed (i.e. if it remained confined in the HF band); in this way we could exclude that occasional slow breaths were indeed creating spurious respiratory LF fluctuations. In the R–R interval spectrum, the LF and HF components were evaluated as a percentage of the total power (i.e. the variance) as an index of the relative prevalence in the sympathetic or vagal contribution [20,21], whereas in the blood pressure and SBF signals the LF and HF were maintained in their absolute units, as they represent different quantities (sympathetic

modulation for the LF, mechanical component due to respiration for the HF [8,20].

The results are given as means ± S.E.M. Due to their skewed distribution the low- and high-frequency oscillations were analysed statistically only after natural logarithmic transformation. Data were analysed by repeated measures analysis of variance, to assess the different effects of altitude and recordings on each variable. Significance levels were set at $P \leqslant 0.05$. We compared, for each group, the results at sea level, after exposure to high altitude and after a week of acclimatization. The activity of the baroreflex in the two groups was studied comparing the LF induced in the cardiovascular signals by neck suction with respect to the LF during controlled breathing with no neck suction. Due to the small number of subjects in the high-altitude native group, no inter-group comparisons were made.

RESULTS

The results are summarized in Figures 1 (mean values), 2 (signal variabilities), 3 (cardiovascular fluctuations evaluated by spectral analysis) and 4 (effect of carotid baroreceptor stimulation by neck suction).

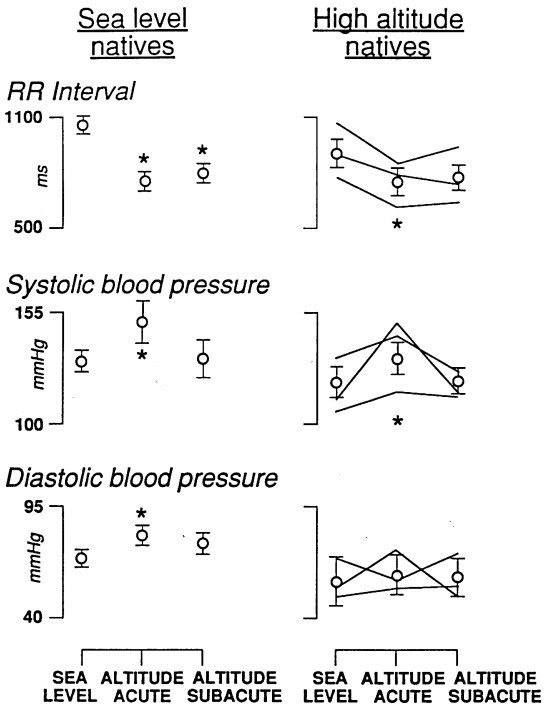

Figure 1 Mean values in R–R interval, systolic and diastolic blood pressures obtained at sea level and after acute and subacute (1 week) exposure to high altitude (4970 m) in sea-level natives and in three high-altitude natives

Individual data are plotted. Note the marked changes in sea-level natives upon arriving at altitude, which were blunted in the high-altitude natives. $^{*}P < 0.05$ versus sea level.

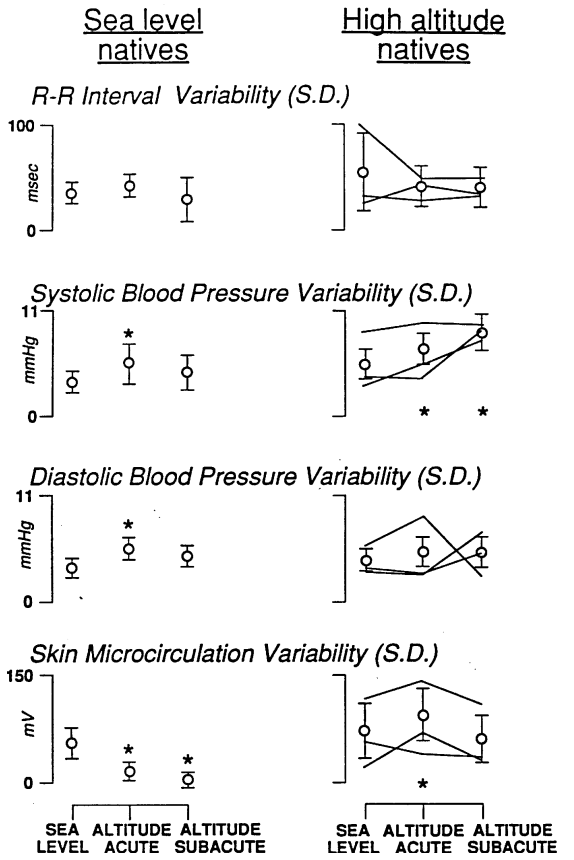

Figure 2 Trends of global variability, determined from S.D. of the mean values, in R–R interval, systolic and diastolic blood pressures and SBF obtained at sea level and after acute and subacute (1 week) exposure to high altitude (4970 m) in sea-level natives and in three high-altitude natives

Individual data are plotted. Note the striking and progressive reduction in variability of the SBF in sea-level natives, which contrasts with the opposite trend in high-altitude natives. Note also the lack of decrease in global variability of all other signals. $^{*}P < 0.05$ versus sea level.

At sea level, systolic and diastolic blood pressures were lower (but not statistically significant) in high-altitude natives (Figure 1). Signal variabilities were similar in the two groups at sea level (Figure 2). All subjects showed typical R–R interval spectra characterized by the simultaneous presence of both LF and HF. LF and HF were also present in the blood pressure and SBF signals (Figure 3). In all subjects the LF neck suction was able to markedly increase the LF components in all cardiovascular signals with respect to the spontaneous LF (Figure 4). This increase in LF was evidence of the capability of the carotid baroreceptors to respond to neck suction, and thus an indirect index of their functionality. During neck suction, only minor decreases in blood pressures and increases in mean R–R interval were observed. Figures 5, 6 and 7 show the time series and the spectra obtained in one sea-level native and in one high-altitude native.

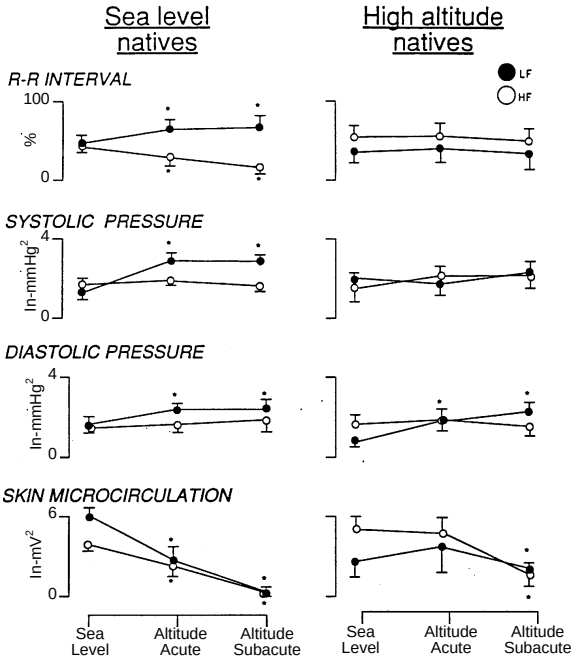

Figure 3 Percentage power of LF and HF components of R–R interval, systolic and diastolic blood pressures and SBF at sea level and after acute and subacute (1 week) exposure to high altitude (4970 m) in sea-level natives and in three high-altitude natives

Note the increases in the LF of R–R interval and blood pressures, which were either absent or less pronounced in high-altitude natives. Note also the reduction in both LF and HF of skin blood flow, due to the extreme vasoconstriction that markedly reduced global SBF variability. $^{*}P < 0.05$ versus sea level.

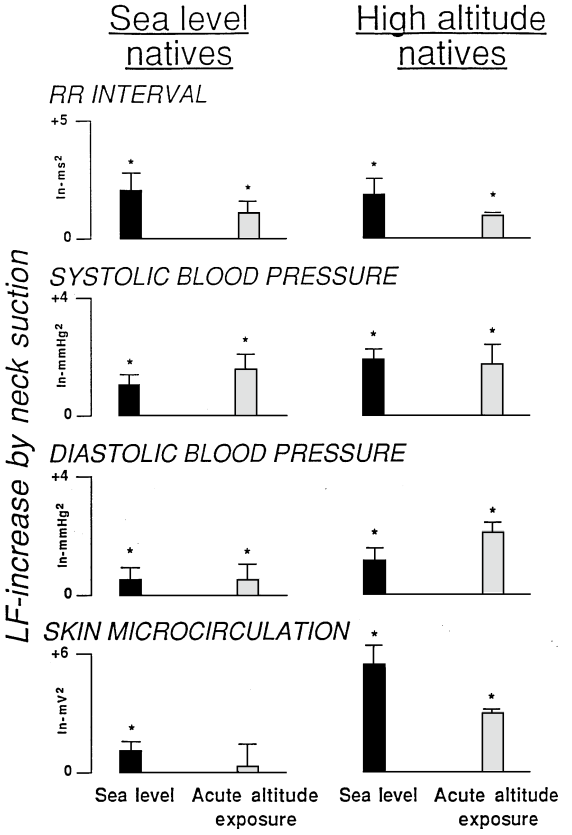

Figure 4 Effect of sinusoidal neck suction on the LF of R–R interval, systolic and diastolic blood pressures and SBF at sea level and after acute exposure to high altitude (4970 m) in sea-level natives and in three high-altitude natives

The bar graphs shows the amount of change in LF induced by neck suction. The asterisks ($^{*}P < 0.05$ or better) mark a significant increase in LF from no neck suction to neck suction. The increase in the LF was evident in all subjects both at sea level and at altitude. All signals showed the same trend; only skin blood flow at altitude showed no significant response, due to the extreme vasoconstriction that markedly reduced global SBF variability.

On acute altitude exposure the mean R–R interval decreased and the systolic and diastolic blood pressures increased in sea-level natives, whereas in high-altitude natives the R–R interval decreased, the systolic blood pressure increased slightly and the diastolic pressure did not change (Figure 1). In both groups the global variability in R–R interval remained unchanged and the variability in systolic pressure increased, whereas the variability in diastolic blood pressure increased in sea-level natives but remained unchanged in high-altitude natives (Figure 2). The variability of SBF markedly decreased in sea-level natives but increased in high-altitude natives (Figure 2). In sea-level natives, the spectrum of the R–R interval showed an increase in the percentage of LF and a relative decrease in the percentage of HF components. In high-altitude natives the predominance of HF components remained unchanged after acute exposure to altitude for R–R interval and SBF. In both groups, however, acute exposure to altitude increased the LF power in diastolic pressure, whereas only in sea-level natives did the LF power increase in systolic pressure. For SBF, the marked reduction in global variability reduced both the LF and HF power in sea-level natives, whereas limited changes were observed in high-altitude natives (Figure 3). Examples of the spectra obtained at high altitude are shown in Figures 6 (sea-level native) and 7 (high-altitude native).

In the sea-level native subjects, the frequency of breathing during spontaneous breathing at sea level was 0.24 ± 0.02 Hz and it increased only slightly after acute exposure to high altitude (to 0.27 ± 0.01 Hz, P not significant). Therefore, in both conditions, the frequency of spontaneous breathing did not significantly differ from that of controlled breathing (0.25 Hz).

On acute exposure to altitude, carotid baroreceptor stimulation by neck suction increased the LF power in R–R interval, systolic and diastolic blood pressure, in both groups, but the increase in LF did not reach statistical significance for SBF. The extent of the increase

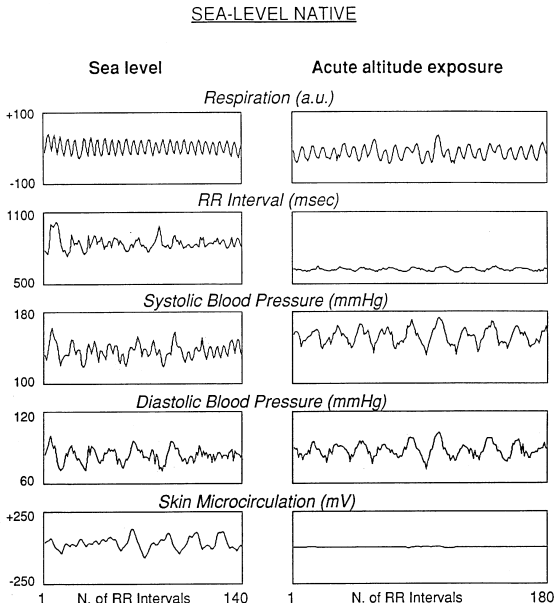

Figure 5 Time series from a sea-level resident obtained at sea level and during acute exposure to high altitude

Notice the almost exclusive presence of LF oscillations at high altitude, in all cardiovascular signals except the microcirculation, which is characterized by an extreme reduction in the variability, as a result of vasoconstriction. a.u., arbitrary units.

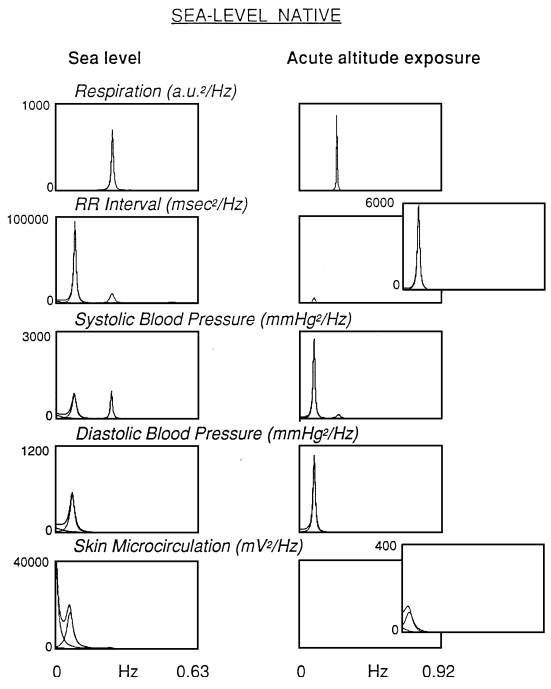

Figure 6 Autoregressive power spectra from the time series of Figure 5

At high altitude the R–R interval and skin microcirculation spectra have been reproduced at the same scale as the corresponding spectra at sea level, and magnified in order to compare the spectral distribution despite the changes in total power.

was slightly but not significantly lower than that observed at sea level (Figure 4).

After 1 week at altitude the mean R–R interval was still lower compared with sea level in sea-level natives, whereas no difference was evident in high-altitude natives. The systolic and diastolic blood pressures were not significantly different from sea level in sea-level natives, and remained unchanged in high-altitude natives (Figure 1). The heart rate variability did not differ significantly compared with sea level in both groups, nor did variability in diastolic blood pressure, whereas the variability in systolic blood pressures decreased in sea-level natives but remained unchanged in high-altitude natives. Conversely, the variability in SBF was further reduced in sea-level natives, whereas in high-altitude natives the values returned to those at sea level (Figure 2).

The R–R interval power spectrum continued to show a relative predominance in LF components in sea-level natives, whereas in high-altitude natives it remained similar to that at sea level, with a predominance in HF components, after 1 week at altitude (Figure 3). The systolic and diastolic blood pressure spectra remained similar to those seen on acute exposure to altitude in the two groups. In the skin microcirculation a further

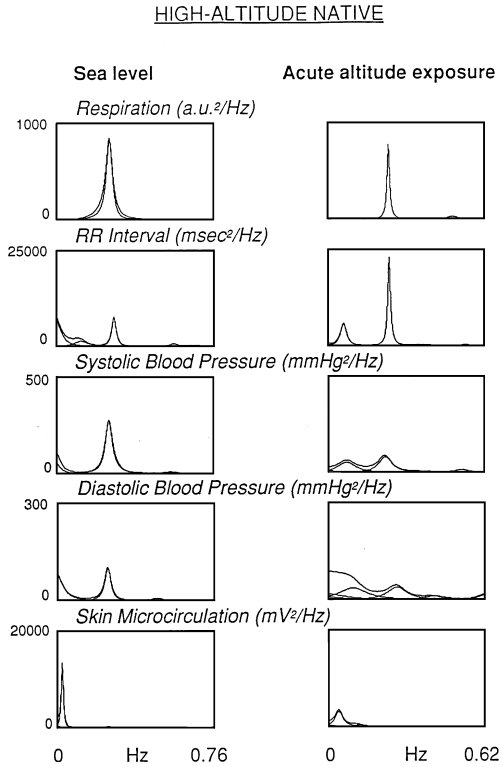

Figure 7 Autoregressive power spectra from a high-altitude native at sea level and during acute exposure to high altitude

Notice the persistence in the HF components (respiratory sinus arrhythmia) in the R–R interval, the moderate increase in LF components of systolic and diastolic blood pressure, and the lack of change in the area (i.e. the power) of the low-frequency oscillations in the skin microcirculation.

reduction in both the LF and HF components was observed in both groups, but values remained higher (though not significantly different from sea-level subjects) in high-altitude natives (Figure 3).

DISCUSSION

After exposure to high altitude we have found evidence of overall sympathetic activation to the heart and blood vessels. Sympathetic activation was evident in sea-level natives by a decrease in R–R interval, an increase in blood pressure, in the LF components of R–R interval, systolic and diastolic blood pressure, and from a marked decrease in SBF variability. Differences in ambient temperature typically affect this parameter, but the changes observed could not be ascribed to the small changes in temperature observed at sea level compared with at high altitude, and a difference in temperature would have affected sea-level and high-altitude natives to a similar extent. After 1 week, however, there was a further reduction in LF and HF spectral components in SBF, confirming vasoconstriction in both groups. This vasoconstriction could be due to the progressive increase in circulating noradrenaline levels reported during the first days of high-altitude exposure [1].

Results of previous studies [1,5–6,10] are in general agreement with our observation. Studies performed with spectral analysis also report a similar relative increase in sympathetic activity, and, at least in relative terms, a decrease in parasympathetic activity [5,6]. Nevertheless, the absence of respiratory signals from these previous investigations, and the presence of frequent altitude-induced respiratory abnormalities, limit the value of these observations. Our study shows the effects of high altitude on autonomic modulation not only of the R–R interval, but also blood pressure and the microcirculation, having measured and standardized the effects of respiration on cardiovascular variability by both measuring and controlling its rate and not fixing the depth to maintain adequate ventilation according to the need of each subject. It should also be noted that the frequency of controlled breathing was not different from that of spontaneous mean respiratory rate; therefore, the main effect of controlled breathing was that of regularization, and it did not induce major alterations of spontaneous breathing pattern.

The presence of higher blood pressure and higher mean heart rate (reduced R–R interval) without major changes in global R–R interval variability is at first sight contradictory. In general, it is assumed that an increase in sympathetic tone is associated with both increased heart rate and decreased heart rate variability [20]. This may not be a strict rule, however, as it has been observed that in normal subjects, in the morning hours, an increase in heart rate is associated with an increase in heart rate variability [22]. The changes observed in spectral components are in the direction of an increase in the LF in both R–R interval and blood pressure, at least in sea-level natives. There is now increasing evidence [23] that these fluctuations can be determined by the activity of the arterial baroreceptors, although the latter are certainly not the only cause of such fluctuations [19,20,24,25].

In our study neck suction was used with a sinusoidal function to cyclically stimulate and de-stimulate the carotid baroreceptors and to observe the effects on the R–R interval and blood pressure [8]. Despite evidence of sympathetic activation, the carotid baroreceptors, as assessed by the neck suction technique, were active on R–R interval and blood pressure, even during acute exposure to high altitude. As a result of the arteriolar vasoconstriction, however, the changes induced by neck suction were not significant in the SBF signal. The present data thus suggest that the arterial baroreflex (or at least of the carotid baroreceptors) is counteracting the increase in sympathetic tone which is related to increased circulating catecholamines [1]. However, the increase in blood pressure in our sea-level native subjects suggests that the baroreflex activity is not sufficient to completely counteract the catecholamine-induced increase in vasoconstriction.

During subacute exposure, although technical failure prevented complete observations, the presence of 0.1-Hz rhythm in all signals remained an indirect indication that the arterial baroreflex was still active, in agreement with the hypothesis that the baroreflex can be a major – although not exclusive [24,25] – determinant of heart rate variability [9] and of LF [23].

Although sympathetic activation was clearly evident in sea-level natives, it was markedly blunted in high-altitude natives living at sea level when acutely returned to their high-altitude birthplace. Although the very small number of subjects studied suggests caution in the interpretation of the data, the reduced sympathetic activation in high-altitude natives was strikingly evident from the limited increase in R–R interval, the lack of increase in diastolic blood pressure and the trend of skin microcirculation which showed an increase in variability during acute altitude exposure. These results, if confirmed in a larger group of subjects, imply that a different autonomic pattern is present in high-altitude Himalayan natives. The explanation for these differences remains speculative. Our three high-altitude natives were living in New Delhi at sea level before joining our expedition: their different autonomic behaviour on re-exposure to altitude may represent an autonomic 'memory' from previous acclimatization, perhaps based on genetic differences of Himalayan natives.

This difference brings us to the question of whether sympathetic activation is deleterious or helpful during acute and subacute exposure to hypoxia. Although sympathetic activation obviously increases oxygen con-

sumption, it might represent a natural response by non-adapted subjects to withstand the effects of hypoxia. Treatment of sea-level natives with β-blockers at altitude decreased heart rate and parasympathetic indices [6], but no information is available on whether reducing sympathetic activity at altitude in those subjects produced some deleterious or helpful effect. On the other hand, the present study clearly suggests that subjects better adapted to high-altitude hypoxia show less or no sympathetic activation, implying that sympathetic activation might not be a necessary consequence of exposure to hypoxia.

Limitations of the study

A field study typically lacks many of the controlled conditions present in the laboratory. Furthermore, high-altitude research (and so the present study) is typically limited to a small number of subjects. However, studies of subjects transiently exposed to the mountain environment give insights, albeit under less rigorously controlled conditions, that cannot be gleaned from laboratory studies. Altitude is not just synonymous with a decrease in barometric pressure and consequent hypoxia as achieved in an altitude chamber, but many additional ambient factors such as temperature (which was in the present study kept close to 20 °C during all the recordings), humidity, sensory stimulation and radiation also play important roles. Thus both laboratory experiments and clinical field studies have their place in defining cardiovascular adaptations to hypoxia. In the present field study, these limitations were partially compensated for by collecting a large number of signals, by carefully controlling the rate of breathing and by limiting the effects of different environmental temperatures. Field studies also offer the opportunity to compare different ethnic groups in the same mountain environment. We have shown here that Himalayan altitude natives, irrespective of their place of abode, have different autonomic responses on re-exposure to their native environment.

The subject age was slightly different between the two groups. Age might determine changes in the cardiovascular and autonomic nervous system, but it seems unlikely that age *per se* was responsible for all the different trends observed in the two groups, also because the younger and older sea-level natives did not seem to behave differently.

Conclusions

In conclusion, our study indicated the presence of increased sympathetic activity, as shown by spectral analysis of cardiovascular signals during high-altitude exposure, and demonstrated that the carotid baroreceptors are active, thus counteracting the increase in blood pressure and peripheral vasoconstriction. Although the response to stimulation of carotid baroreceptors is preserved during exposure to altitude, this capability is not sufficient to avoid an increase in blood pressure. The lesser degree of sympathetic activation observed in high-altitude Himalayan natives despite long-term acclimatization at sea level, if confirmed in a larger group, suggests the possibility of genetic adaptation or 'memory' to previous altitude exposure.

The present findings might have a practical impact on our understanding and management of mountain sickness: manoeuvres or conditions that steadily reduce the catecholamine levels and/or increase the baroreflex, and vagal activity at altitude in general, might ultimately improve the possibility of adaptation. Lessons learned from studies carried out on normal subjects at high altitude have relevance to the care of patients with cardiovascular disease (particularly coronary artery disease), in which the importance of sympathetic activation and baroreflex dysfunction is now recognized [26]. Further studies might establish whether strategies able to reduce the altitude-induced hypoxia and consequent sympathetic activation might reduce the deleterious effects of hypoxia due to pathological conditions.

REFERENCES

1 Mazzeo, R. S., Wolfel, E. E., Butterfield, G. E. and Reeves, J. T. (1994) Sympathetic response during 21 days at high altitude (4,300 m) as determined by urinary and arterial catecholamines. Metabolism **43**, 1226–1232
2 Mazzeo, R. S., Bender, P. R., Brooks, G. A. et al. (1991) Arterial catecholamine responses during exercise with acute and chronic high-altitude exposure. Am. J. Physiol. **261**, 419–424
3 Pace, N., Grinswold, R. L. and Grunbaum, B. W. (1964) Increase in urinary norepinephrine excretion during 14 days sojourn at 3800 meters elevation. Fed. Proc. **23**, 521
4 Surks, M. I., Beckwitt, H. J. and Chidsey, C. A. (1967) Changes in plasma thyroxine concentration and metabolism catecholamine excretion and basal oxygen consumption during acute exposure to high altitude. J. Clin. Endocrinol. **27**, 789–799
5 Ponchia, A., Noventa, D., Bertaglia, A. et al. (1994) Cardiovascular neural regulation during and after prolonged high altitude exposure. Eur. Heart J. **15**, 1463–1469
6 Hughson, R. L., Yamamoto, Y., McCullough, R. E., Sutton, J. R. and Reeves, J. T. (1994) Sympathetic and parasympathetic indicators of heart rate control at altitude studied by spectral analysis. J. Appl. Physiol. **77**, 2537–2542
7 Bernardi, L., Hayoz, D., Wenzel, R. et al. (1997) Synchronous and baroreceptor-sensitive oscillations in skin microcirculation. Evidence for central autonomic control. Am. J. Physiol. **273**, 1867–1878
8 Bernardi, L., Bianchini, B., Spadacini, G. et al. (1995) Demonstrable cardiac reinnervation after human heart transplantation by carotid baroreflex modulation of R–R interval. Circulation **92**, 2895–2903
9 Piepoli, M., Sleight, P., Leuzzi, S. et al. (1997) Origin of respiratory sinus arrhythmia in conscious humans: an important role for the arterial carotid baroreceptors. Circulation **95**, 1813–1821
10 Zhuang, J., Droma, T., Sutton, J. R. et al. (1993) Autonomic regulation of heart rate response to exercise in Tibetan and Han residents of Lhasa (3658 m). J. Appl. Physiol. **75**, 1968–1973

11 Bernardi, L., Salvucci, F., Suardi, R. et al. (1990) Evidence for an intrinsic mechanism regulating heart rate variability in the transplanted and the intact heart during submaximal dynamic exercise? Cardiovasc. Res. **24**, 969–981
12 Challoner, A. V. J. (1979) Photoelectric plethysmography for estimating cutaneous blood flow. In Non-invasive Physiologic Measurements (Rolfe, P., ed.), pp. 125–151, Academic Press, London
13 Bernardi, L. and Leuzzi, S. (1994) Laser Doppler velocimetry and photoplethysmography: hardware and measuring principles. In Handbooks of Skin Engineering, vol. 2 (Berardesca, E., Elsner, P. and Maibach, H., eds.), pp. 31–56, CRC Press, Boca Raton, FL, U.S.A.
14 Uretzky, G. and Palti, Y. (1971) A method for comparing transmitted and reflected light photoelectric plethysmography. J. Appl. Physiol. **31**, 32–35
15 Nielsen, P. E., Lønsmann-Poulsen, H. and Gyntelberg, F. (1973) Skin blood pressure measured by a photoelectric probe and external counterpressure. Scand. J. Clin. Lab. Invest. **31** (Suppl. 128), 137–411
16 Holstein, P., Nielsen, P. E. and Barras, J. P. (1979) Blood flow cessation at external pressure in the skin of normal human limbs. Microvasc. Res. **17**, 71–79
17 Holstein, P., Nielsen, P. E., Lund, P., Gyntelberg, F. and Lønsmann-Poulsen, H. (1980) Skin perfusion pressure on the legs measured as the external pressure required for skin reddening after blanching: a photo-electric technique compared to isotope washout. Scand. J. Clin. Lab. Invest. **40**, 35–43
18 Bernardi, L., Rossi, M., Fratino, P., Finardi, G., Mevio, E. and Orlandi, C. (1989) Relationship between changes in human skin blood flow and autonomic tone. Microvasc. Res. **37**, 16–27
19 Pagani, M., Lombardi, F., Guzzetti, S. et al. (1986) Power spectral analysis of heart rate and arterial pressure variabilities as a marker of sympatho-vagal interaction in man and conscious dog. Circ. Res. **59**, 178–193
20 Malliani, A., Pagani, M., Lombardi, F. and Cerutti, S. (1991) Cardiovascular neural regulation explored in the frequency domain. Circulation **84**, 482–492
21 Radaelli, A., Bernardi, L., Valle, F. et al. (1994) Cardiovascular autonomic modulation in essential hypertension: effect of tilting. Hypertension **24**, 556–563
22 Bernardi, L., Ricordi, L., Lazzari, P. L. et al. (1992) Impaired circadian modulation of sympatho-vagal activity in diabetes: a possible explanation for altered temporal onset of cardiovascular disease. Circulation **86**, 1443–1452
23 Bernardi, L., Leuzzi, S., Radaelli, A., Passino, C., Johnston, J. A. and Sleight, P. (1994) Low-frequency spontaneous fluctuations of R–R interval and blood pressure in conscious humans: a baroreceptor or central phenomenon? Clin. Sci. **87**, 649–654
24 Passino, C., Sleight, P., Valle, F., Spadacini, G., Leuzzi, S. and Bernardi, L. (1997) Lack of peripheral modulation of cardiovascular central oscillatory autonomic activity during apnea in humans. Am. J. Physiol. **272**, 123–129
25 Pagani, M., Montano, N., Porta, A. et al. (1997) Relationship between spectral components of cardiovascular variabilities and direct measures of muscle sympathetic nerve activity in humans. Circulation **95**, 1441–1448
26 La Rovere, M. T., Bigger, Jr, J. T., Marcus, F. I., Mortara, A. and Schwartz, P. J., for the ATRAMI investigators (1998) Baroreflex sensitivity and heart-rate variability in prediction of total cardiac mortality after myocardial infarction. Lancet **351**, 478–484

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