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QUANTIFYING ALTITUDE RELATED OXIDATIVE STRESS BY CARBONYL PROTEINS

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Abstract

Introduction: In quantifying altitude related oxidative stress on a molecular level carbonylated proteins show great promise. They have been used as a biological marker to determine oxidative stress in newborns so far. In contrast to other markers this substance group is very stable and early detectable. Therefore, their use in a high altitude environment should be with good prospects.

Objective: To test the hypothesis that acute exposure to hypobaric hypoxia induces measurable oxidative stress that might be related to the severity of AMS.

Methods: Twelve subjects spent a night in a hypobaric chamber at a simulated altitude of 4000 m. One hour before altitude exposure and two hours after arrival at 4000 m a blood sample was taken and protein carbonylation was analyzed by special ELISA. Results were compared with severity of acute mountain sickness (AMS) after spending 11 hours at 4000 m quantified by the Lake Louise Score.

Results: The fast ascent to 4000 m generates a measurable oxidative stress to the organism with a wide range in individual susceptibility. This appears at altitude expressed by a significant change of carbonyl proteins (P = 0.01) as well as for the severity of AMS. Three of the subjects having no or least AMS showed a decrease in carbonyl proteins. All other subjects had an increase in carbonyl proteins. The participant with the highest increase in carbonyl proteins was the most severely ill person. Regression analysis showed a linear regression; r^2 = 0.43.

Conclusions: This is the first time a correlation between carbonyl proteins and high altitude symptomatology has been detected, making these highly interesting for further investigations at high altitude.

Key words: *high altitude, oxidative stress, protein carbonylation, carbonyl proteins, AMS*

Introduction

Altitude exposure is known to induce a significant oxidative stress [1] even without strenuous physical exertion. The burden of oxidative stress increases during the time spent at altitude and may even persist for some time upon return to sea level. The physiological and medical consequences of increased oxidative stress engendered by altitude are unclear; indeed, hypoxia is believed to be the trigger for the cascade of signalling events that ultimately leads to adaptation to altitude. These signalling events include the generation of reactive oxygen species (ROS) that may elicit important adaptive responses. If produced in excess, however, these ROS may contribute to impaired muscle function and reduced capillary perfusion at altitude or may even play a role in precipitating more serious neurological and pulmonary crisis. Altitude related illnesses like acute mountain sickness (AMS), high altitude pulmonary edema (HAPE) and high altitude cerebral edema (HACE) are possibly related to hypoxia induced free radical formation that alter blood-brain barrier permeability [2].

In quantifying this altitude related oxidative stress on a molecular level carbonyl proteins (CP) show great promise. They have been used as a biological marker to determine oxidative stress in newborns so far [3,4]. In contrast to other markers this substance group is very stable and early detectable [5]. Therefore, their use in a high altitude environment should be with good prospects.

This prospective comparative study was conducted to test the hypothesis that acute exposure to hypobaric hypoxia induces measurable oxidative stress that might be related to the severity of AMS.

Material and methods

The test was performed in the altitude and climate chamber of the German Air Force Institute of Aviation Medicine in Koenigsbrueck, Germany (134 m). This hypobaric chamber can hold six individuals for overnight stays. As we had a group of twelve male subjects (median age: 23 years, range: 18-33 years; median height: 182.5 cm, range: 169-194 cm; median weight: 76 kg, range: 55-100 kg; median body mass index: 22.5 kg/m^2, range: 19-29 kg/m^2) two identical trials were conducted. Each trial consisted of an overnight stay in the chamber. The subjects were decompressed over a period of 53 minutes to a pressure equivalent to that at an altitude of 4000 m. The subjects spent twelve hours in the chamber under these altitude conditions. At any time, the subjects could be rapidly recompressed or could leave the chamber through an airlock. An emergency physician with expertise in altitude medicine was always present. Prior to the

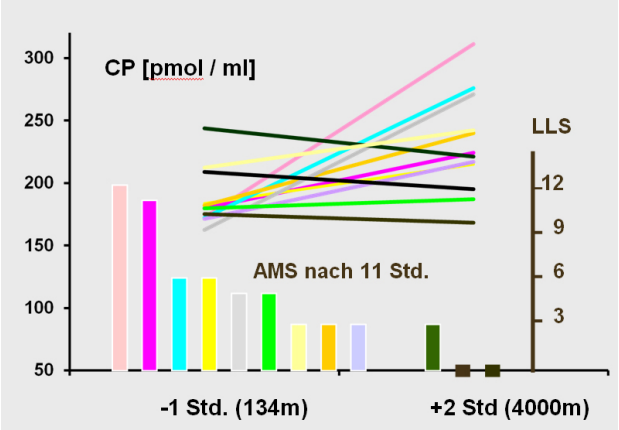

Fig. 1. *Course of carbonyl proteins (CP) while exposed to 4000 m over 2 hours; 3 subjects (dark colors) show a decrease, 9 (bright colors) an increase of CP. The beams (same color: same subject) express the Lake Louise Score after 11 hours of exposure to 4000 m; the dark colored subjects (decrease of CP) had only a minor (0-3) LLS-Score; all the subjects with CP-increase (bright colors) had an even higher LLS-Score (3-12)*

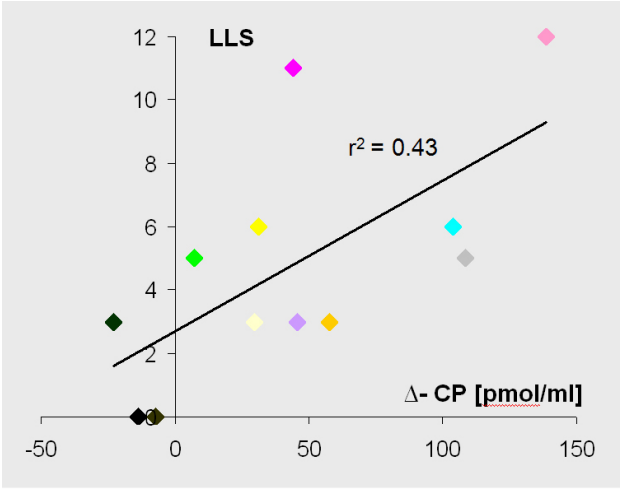

Fig. 2. *Regression analysis of carbonyl proteins (CP) and Lake Louise Score (LLS) while exposed to 4000 m over 2 hours; 3 subjects (dark colors: left side) show a decrease, 9 (bright colors: right side) an increase of CP*

tests, the subjects received an echocardiogram (ECG). Routine laboratory blood tests and a 12-lead ECG were performed immediately before the tests. All tests were normal.

This study was embedded in a larger study that had been approved by the ethics committee of the Society of Physicians of the state of Baden-Wuerttemberg, Stuttgart, Germany. All participants had given their written informed consent to take part in the study.

During the test-period including the day before all subjects received the same food that was prepared by the staff restaurant of the Institute of Aviation Medicine. This food was normal German Army diet without any special dietary preparations. During the trial, blood was collected from an indwelling venous cannula ($1^3/_4$ French) in the left forearm without the use of a tourniquet 1 hour before the decompression and 2 hours after reaching 4000 m. The blood was centrifuged and the serum was separated and stored at -20°C. The samples were transported on dry ice at a temperature of -40°C and CP was analyzed by ELISA (Immundiagnostik, Germany). The presence or absence of altitude symptoms was assessed using the Lake Louise scoring (LLS) system [6]. In accordance with the recommendations by Maggiorini et al., subjects were considered to be affected by altitude sickness when they had an LLS of 5 or greater [7]. CP-results were compared with the severity of AMS after spending 11 hours at 4000 m quantified by the LLS.

Statistical analysis

The results of the first and the second test blood sample were compared using the paired *t*-test. We accepted a *P*-value of less than 0.05 as indicator of statistical significance. Linear regression analysis was used to describe the correlation of LLS and change in hypoxia related CP.

Results

The fast ascent to 4000 m generates a measurable oxidative stress to the organism with a wide range in individual susceptibility. This appears at altitude expressed by a significant change of CP ($P = 0.01$) as well as for the severity of AMS. Three of the subjects having no or least AMS showed a decrease in CP. All other subjects had an increase in carbonyl proteins and an even higher LLS-Score of 3 to 12 (Fig.1). The participant with the highest increase in carbonyl proteins was the most severely ill person. Regression analysis showed a linear regression; $r^2 = 0.43$ (Fig. 2).

Discussion

Our results supports the findings of other authors that altitude related oxidative stress plays an important role in the acclimatization process to altitude [8-10] as well as in the development of altitude related illness [11,12].

Although the regression analysis showed only a moderate correlation it is remarkable that CP after 2 hours at altitude comparable to 4000 m behaved complete different in subjects with no or least AMS symptoms (decrease of CP) compared to those with apparent AMS (increase in CP). Despite of the unphysiological rapid ascent to altitude the course of CP seems to have prognostic relevance regarding the development of AMS symptoms in the near future. This might become very helpful in all cases of decision making if individuals could ascent further or if they need more time to acclimatize. This appears particularly if a fast ascent (car, aircraft, cable railway) is planed. This is a topic of high interest for example in military missions or rescue operations at high altitude [13].

This is the first time a correlation between carbonyl proteins and high altitude symptomatology has been detected, making these highly interesting for further investigations at high altitude.

Declaration of interest

The authors report no conflicts of interest.

References

1. Jefferson JA, Simoni J, Escudero E, et al. Increased oxidative stress following acute and chronic high altitude exposure. *High Alt Med Biol* 2004; 5(1): 61-9.
2. Askew EW. Work at high altitude and oxidative stress: antioxidant nutrients. *Toxicology* 2002; 180(2): 107-19.
3. Saker M, Soulimane Mokhtari N, Merzouk SA, et al. Oxidant and antioxidant status in mothers and their newborns according to birthweight. *Eur J Obstet Gynecol Reprod Biol* 2008; 141(2): 95-9.
4. Zitnanova I, Sumegova K, Simko M, et al. Protein carbonyls as a biomarker of foetal-neonatal hypoxic stress. *Clin Biochem* 2007; 40(8): 567-70.
5. Pantke U, Volk T, Schmutzler M, et al. Oxidized proteins as a marker of oxidative stress during coronary heart surgery. *Free Radic Biol Med* 1999; 27(9-10): 1080-6.
6. Hackett P, Oelz O. The Lake Louise consensus on the definition and quantification of altitude illness. In: Sutton J, Coates G, Houston C, Ed. Hypoxia and Mountain Medicine, Burlington: Queen City Printers, 1992; 327-30.
7. Maggiorini M, Muller A, Hofstetter D, et al. Assessment of acute mountain sickness by different score protocols in the Swiss Alps. *Aviat Space Environ Med* 1998; 69(12): 1186-92.
8. Hornbein TF. The high-altitude brain. *J Exp Biol* 2001; 204(18): 3129-32.
9. Joanny P, Steinberg J, Robach P, et al. Operation Everest III (Comex'97): the effect of simulated sever hypobaric hypoxia on lipid peroxidation and antioxidant defence systems in human blood at rest and after maximal exercise. *Resuscitation* 2001; 49(3): 307-14.
10. Neubauer JA. Invited review: Physiological and pathophysiological responses to intermittent hypoxia. *J Appl Physiol* 2001; 90(4): 1593-9.
11. Roach RC, Hackett PH. Frontiers of hypoxia research: acute mountain sickness. *J Exp Biol* 2001; 204(18): 3161-70.
12. Roach RC, Maes D, Sandoval D, et al. Exercise exacerbates acute mountain sickness at simulated high altitude. *J Appl Physiol* 2000; 88(2): 581-5.
13. Tannheimer M, Albertini N, Ulmer HV, et al. Testing individual risk of acute mountain sickness at greater altitudes. *Mil Med* 2009; 174(4): 363-9.

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