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REGIONAL CEREBRAL BLOOD FLOW DURING ACUTE HYPOXIA IN INDIVIDUALS SUSCEPTIBLE TO ACUTE MOUNTAIN SICKNESS

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Abstract

Individuals susceptible to high altitude pulmonary edema show altered pulmonary vascular responses within minutes of exposure to hypoxia. We hypothesized that a similar acute-phase vulnerability to hypoxia may exist in the brain of individuals susceptible to acute mountain sickness (AMS). In established AMS and high-altitude cerebral edema, there is a propensity for vasogenic white matter edema. We therefore hypothesized that increased cerebral blood flow (CBF) during acute hypoxia would also be disproportionately greater in white matter (WM) than grey matter (GM) in AMS-susceptible subjects.

We quantified regional CBF using arterial spin labeling MRI during 30-minutes hypoxia ($F_{I}O_2=0.125$) in 2 groups: AMS-susceptible (AMS-S, n=6) who invariably experienced AMS at altitude, and AMS-resistant (AMS-R, n=6) who never experienced AMS despite multiple rapid ascents.

SaO₂ during hypoxia did not differ between groups (AMS-S=87±4%, AMSR=89±3%, p=0.3). Steady-state whole-brain CBF increased in hypoxia (p<0.005), but did not differ between groups (Normoxia: AMS-S=42.7±14.0ml/100g/min, AMS-R=41.7±10.1ml/100g/min, Hypoxia: AMS-S=47.8±19.5ml/100g/min, AMS-R=48.2±10.1ml/100g/min, p=0.65), and cerebral oxygen delivery remained constant. The percent change in CBF did not differ between brain regions or between groups (although absolute CBF change was greater in GM): (GM: AMS-S=6.1±7.7ml/100g/min (10±11%), AMS-R=8.3±5.7ml/100g/min (17±11%) p=0.57; WM: AMS-S=4.3±5.1ml/100g/min (12±15%), AMS-R=4.8±2.9ml/100g/min (16±9%), p= 0.82).

Conclusion: CBF increases in acute hypoxia, but is not different between WM and GM, irrespective of AMS susceptibility. Acute phase differences in regional CBF during acute hypoxia are not a primary feature of susceptibility to AMS.

Keywords

acute mountain sickness; cerebral blood flow; hypoxia; cerebral oxygen delivery; magnetic resonance imaging; arterial spin labeling

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1. Introduction

Acute Mountain Sickness (AMS) is the most common of the high altitude illnesses and is characterized by headache, nausea, vomiting, anorexia, dizziness, lethargy, fatigue, and sleep disturbance experienced during rapid ascent to a higher altitude. Symptoms are highly variable between individuals, and individual susceptibility appears to play a significant role (Hackett et al. 2001; Roach et al. 2001; Basnyat et al. 2003).

In high altitude pulmonary edema (HAPE), a previous history of HAPE is an important risk factor (Bartsch et al. 2005). In these individuals, an altered pulmonary vascular response is evident as early as 10 minutes after exposure to hypoxia (Hopkins et al. 2005), long before the time required for the development of symptoms. We hypothesized that a similar vulnerability may exist in the brain, such that in a susceptible population of subjects who consistently developed acute mountain sickness on exposure to high altitude (AMS-susceptible) the early cerebral blood flow response to acute hypoxia would be augmented compared to subjects who rarely if ever developed symptoms, despite repeated rapid high altitude exposures (AMS-resistant). In severe AMS with edema, or in high altitude cerebral edema (HACE) which is believed to share a common pathophysiology (Hackett et al. 2001), previous MRI studies have shown the edema is primarily localized to white matter (Hackett 1999; Schoonman et al. 2007). Consequently, disturbances to normal cerebral blood flow, particularly in white matter, may be present in subjects predisposed to AMS.

Therefore the purpose of this study was to evaluate the acute cerebral response to hypoxia looking specifically at differences in cerebral blood flow in grey matter and white matter in relation to AMS susceptibility. We used a sensitive magnetic resonance imaging (MRI) technique, pulsed arterial spin labeling (ASL) (Wong et al. 1997; Wong et al. 1998), to quantify regional cerebral blood flow. The high spatial resolution (~3–4mm) allows the quantification of the blood flow response regionally across the brain and the ability of ASL to make dynamic measurements at high temporal resolution (~2–4 seconds) allows us to examine the effects of acute hypoxia and the return to normoxia during a continuous scanning session. We hypothesized that the acute cerebral blood flow increase in response to hypoxia, particularly in white matter, would be greater in AMS-susceptible than AMS resistant subjects.

2. Methods

2.1 Subject selection

The Human Subjects Research Protection Program of the University of California San Diego approved this study. All subjects gave written informed consent. Healthy non-smoking sea-level resident subjects were recruited based a history of repeated ascents above 2,500 m and self reporting of susceptibility to symptoms of AMS. Subjects completed a questionnaire detailing their high altitude exposure above 2,500 m and 4,200 m and completed a Lake Louise Consensus AMS Scoring questionnaire with questions regarding symptoms relevant to AMS experienced during their average prior high altitude visits (headache, nausea, fatigue, dizziness, insomnia, confusion, ataxia, edema, malaise). This was used to retrospectively assign a total AMS score reflecting severity (Roach et al. 1993). Subjects were selected based on frequency and severity of symptoms (or absence of symptoms), as well as ability to undergo MRI examination. To remove any potential confounding effects of recent altitude exposure, during the month prior to the experiments all subjects slept below 1800 m and did not sojourn above 2,400 m.

2.2 Experimental Design

To maximize sensitivity to small changes in cerebral blood flow, data were obtained within a single experimental session. Subjects lay in a supine position in the MRI scanner (watching a movie or listening to music) while wearing a face mask equipped with a non-rebreathing valve (Hans Rudolph 8930/2700, Kansas City, MO). Baseline cerebral blood flow data were obtained while resting in the scanner for 10 minutes breathing room air. Then the inspired port of the Hans Rudolph valve was connected to a Douglas bag containing a gas mixture of 12.5% O₂ in nitrogen N₂ (equivalent to an inspired partial pressure of O₂ of approximately 90 Torr or 3800 m equivalent altitude) using a remotely controlled pneumatically activated three-way valve (Hans Rudolph 4285/8500, Hans Rudolph Inc. Kansas City, MO) located on the inspired limb of the circuit. Data were collected continuously during 30 minutes of normobaric hypoxia, before switching back to room air for an additional 30 minutes of normoxia. Since all subjects had extensive high altitude experience and could potentially identify the symptoms of acute hypoxia it was not possible to entirely blind subjects to the F_IO₂ they were breathing. They were, however, unaware of the timing of the change in F_IO₂. Heart rate, respiration rate, oxygen saturation (SaO₂) and end-tidal pCO₂ were continuously monitored during the studies using an MRI compatible physiological monitoring system (3150 MRI patient monitor, and 3155 remote display, In Vivo Research Inc.).

2.3 MRI Measurements

All MR Images were acquired at 3 Tesla (Excite II, General Electric, Milwaukee, WI). Cerebral blood flow data were acquired using a pulsed arterial spin labeling (ASL) technique with a PICORE tagging scheme with QUIPSS II modification to control the bolus width (Wong et al. 1998). ASL data were acquired in 5 axial slices through the superior cerebrum using a dual-echo spiral readout with: TE₁ = 9.1 ms, TE₂ = 30 ms, TR = 4 s, Flip angle = 90 degrees, 20 cm tag width, 1 cm gap between the tag and the proximal imaging slice, TI₁ = 700 ms, TI₂ = 1400 ms, resolution ~4×4 mm in-plane, 8 mm thick. A small diffusion gradient was applied to remove flow signal contamination from large vessels (b-value = 2 s/mm²) and thus data acquired represent a true measure of cerebral perfusion. Data were continuously acquired over the 70-minute paradigm of 10 minutes normoxia, 30 minutes 12.5% hypoxia, 30 minutes normoxia (1050 4-second repetitions). Blood flow data were post-processed using the surround subtraction technique described by Liu and Wong (Liu et al. 2005). We simultaneously acquired additional measurements of proton density of the cerebrospinal fluid and a correction for coil sensitivity profile allowing us to quantify the cerebral blood flow in absolute units (Buxton et al. 1998; Wegener et al. 2007). A T1 weighted anatomical dataset using a 3D Fast Spoiled GRASS (FSPGR) sequence was also acquired and used to generate the masks to study regional perfusion (TE = 3.1 ms, TR = 7.9 ms, TI = 450 ms. BW 31.25 kHz, FOV 25cm × 25cm × 15cm, matrix 256 × 256 × 124 ~1 × 1 × 1.2 mm resolution). From the FSPGR images we used the FMRIB automated segmentation software (FMRIB Software Library, FMRIB, Oxford, UK) to generate partial volume maps of 3 cerebral tissues (grey matter, white matter and cerebrospinal fluid). Using the default threshold of 0.7, we defined 3 regions of interest (masks): global brain (i.e. all cerebral tissues in the cerebrum), grey matter (encompassing all pixels with >0.7 probability of containing grey matter) and white matter (encompassing all pixels with >0.7 probability of containing white matter). These 3 masks were applied to the ASL data (Figure 1). hypoxic (and normoxic) periods were averaged to examine steady-state differences in cerebral blood flow between brain regions, and between AMS-groups. The two periods for steady-state blood flow measurements were baseline normoxia (measured from the mean CBF signal in the first 10 minutes and last 10 minutes of the 70-minute paradigm), and hypoxia (measured during the last 15 minutes of the 30-minute hypoxia stimulus). In addition time-varying (transient) changes in cerebral blood flow were plotted for global brain, grey matter and white matter (Figures 3–5). Signal to noise in the ASL data was calculated for each

5-minute acquisition from the ratio of the mean signal in the grey or white matter regions of interest to the standard deviation of the background noise.

2.4 Cerebral oxygen delivery

Cerebral oxygen delivery rate, the product of cerebral blood flow and arterial oxygen content ($\text{CBF} \times \text{CaO}_2$), was estimated from the measured CBF and the arterial O_2 content equation: $\text{CaO}_2 = \text{Hb} \times 1.34 \times \text{SaO}_2 + \text{PaO}_2 \times 0.003 \text{ ml O}_2/100 \text{ ml blood}$ (where CaO_2 is arterial O_2 content, PaO_2 is arterial O_2 tension, SaO_2 is arterial O_2 saturation and Hb is arterial hemoglobin concentration). We assumed that dissolved O_2 in plasma was negligible, and there is no change in arterial Hb concentration during the 30 minute hypoxia exposure. Thus the fractional change in O_2 delivery rate during acute hypoxia was calculated from the fractional change in $\text{CBF} \times \text{SaO}_2$ product, which for simplicity is referred to as “cerebral oxygen delivery” in the manuscript.

2.5 Statistical Analysis

Data were analyzed using ANOVA (Statview 4.1, SAS Institute Inc. Cary, NC) with one grouping factor (subject group, 2 levels - AMS-susceptible, AMS-resistant) and one repeated measure (time 14 levels - 5 minute intervals 0–70 minutes). Steady-state data were analyzed with two grouping factors (subject group, 2 levels - AMS-susceptible, AMS-resistant; brain region, 2 levels - grey matter, white matter) and one repeated measure (FIO_2 , 2 levels - normoxia, hypoxia). All data are expressed as means $\pm$ SD. Significance was accepted at $p < 0.05$, 2 tailed.

3. Results

3.1 Subject characteristics

Thirty-three subjects completed questionnaires. Of these, twelve subjects met criteria for either the AMS-susceptible or AMS-resistant study group and completed the study. Subject characteristics are given in Table 1. The groups were similar with respect to age. On average, the AMS-resistant group had more visits to altitudes and ascended to significantly higher overall elevations. The AMS-susceptible group reported more frequent altitude-related symptoms and a higher mean Lake Louise Score, compared with the AMS-resistant group who reported almost never experiencing any altitude-related symptoms on prior altitude visits. All subjects tolerated the MRI and acute hypoxia well. None reported any cerebral symptoms following the acute hypoxic stimulus (in keeping with the short duration of hypoxic exposure).

3.2 Arterial Oxygen saturation, heart rate and end-tidal pCO_2

Heart rate and arterial oxygen saturation data are summarized in Figure 2. During the 30 minutes of hypoxic exposure, heart rate increased significantly ($p < 0.0001$) but was not statistically different between groups ($p = 0.55$). Oxygen saturation decreased significantly ($p < 0.0001$), averaging $87 \pm 4\%$ but the change in saturation was also not significantly different between groups ($p = 0.31$). End-tidal pCO_2 averaged 39 ± 7 Torr during hypoxia, but this also was not significantly different between groups ($p = 0.56$) (data not shown). Respiratory rate during hypoxia averaged $12.4 \pm 3.2 \text{ min}^{-1}$, but this also was not significantly different from normoxia ($12.5 \pm 4.4 \text{ min}^{-1}$, $p = 0.29$) nor significantly different between groups ($p = 0.25$) (data not shown).

3.3 Steady State Cerebral Blood Flow

Steady-state cerebral blood flow during hypoxia was compared between cerebral regions (white matter vs. grey matter) and between AMS groups (AMS-R vs. AMS-S). Data are summarized in Table 2.

Comparing steady-state blood flow between gray matter and white matter; The absolute change in blood flow was significantly greater for grey matter than white matter ($p < 0.01$) however the percentage change in blood flow was not significantly different between regions ($p = 0.74$).

Comparing AMS groups; Mean steady state global brain blood flow increased during hypoxia by 5.1 ± 6.4 ml/100g/min (10 ± 12 %) in the AMS-susceptible subjects, and by 6.5 ± 4.0 ml/100g/min (17 ± 10 %) in the AMS-resistant subjects. In grey matter, mean steady state flow increased by 6.1 ± 7.7 ml/100g/min (10 ± 11 %) and by 8.3 ± 5.7 ml/100g/min (17 ± 11 %) (in the AMS-susceptible and AMS-resistant subjects respectively). In white matter, mean steady state blood flow increased during hypoxia by 4.3 ± 5.1 ml/100g/min (12 ± 15 %), and by 4.8 ± 2.9 ml/100g/min (16 ± 9 %) (in the AMS-sensitive and AMS-resistant subjects respectively). For both absolute and percentage measurements, grey matter and white matter blood flow change was not significantly different between AMS groups ($p = 0.66$, $p = 0.39$ for absolute and percentage differences respectively).

3.4 Dynamic Cerebral Blood Flow

To detect transient changes in cerebral blood flow that might otherwise be masked by examining the steady state changes alone, we also plotted the dynamic cerebral blood flow for different brain regions during the hypoxic stimulus (Figures 3–5). In both AMS-susceptible and AMS-resistant groups, cerebral blood flow showed a significant increase during 30-minutes of hypoxia and returned to baseline during the subsequent 30-minutes of normoxia for all 3 regions (main effect for time, $p < 0.0001$ for each of global, grey matter and white matter regions). Mean blood flow was also highly significantly different in grey matter and white matter (main effect for brain region, $p < 0.0001$). The change in blood flow with hypoxia was not significantly different between AMS-susceptible and AMS-resistant subjects for global brain, grey matter or white matter ($p = 0.75$, $p = 0.71$, $p = 0.89$ respectively). The mean signal to noise ratio (SNR) for the 5-minute cerebral blood flow measurements was 12.6 in white matter and 21.6 in grey matter.

3.5 Cerebral oxygen delivery

Cerebral oxygen delivery was maintained during 30-minutes of hypoxia with no significant fractional change in O_2 delivery in grey matter or white matter, based on the measured $SaO_2 \times CBF$ product ($p = 0.22$ for global brain, $p = 0.11$ for grey matter and $p = 0.55$ for white matter -main effect for brain region). Fractional change in cerebral oxygen delivery with hypoxia was also not significantly different between AMS-susceptible and AMS-resistant subjects ($p = 0.11$ for global brain, $p = 0.14$ for grey matter and $p = 0.27$ for white matter).

4. Discussion

4.1 Main Findings

Arterial spin labeling MRI allowed dynamic measurement of absolute regional cerebral perfusion, and to our knowledge this is the first time it has been used to quantify regional cerebral blood flow during acute hypoxia in the context of susceptibility to acute mountain sickness. The main findings of this study were 1) that acute exposure to 12.5% oxygen increased cerebral blood flow to a similar extent in both AMS-susceptible and AMS-resistant subjects. 2) This blood flow increase was observed in both white matter and grey matter regions of the brain, and was not different between susceptibility groups. Even though the absolute change in blood flow was significantly greater in grey matter than in white matter, this was in proportion to the resting blood flow for each region, so the percentage change was not significantly different between brain regions. 3) During acute hypoxia the cerebral oxygen delivery was maintained in both grey matter and white matter, irrespective of AMS susceptibility.

4.2 Subject selection

The hypoxic stimulus of 12.5% oxygen is equivalent to 3800m elevation, which is a level where many individuals will become symptomatic and a level of hypoxia consistent with eventual symptoms in our subjects. In our group of subjects, based on their prior history from extensive experience at altitude we would expect all of the AMS-S group to develop acute mountain sickness, and none of the AMS-R group to develop symptoms. In the current study we found an increase in cerebral blood flow in both grey matter and white matter following 30 minutes of hypoxia at 12.5% O₂, but no significant difference in the acute-phase response to hypoxia between AMS-susceptible and AMS-resistant subjects. We selected subjects from distinct susceptible and resistant groups so that if there are physiological differences in the brains of those who are at risk from AMS and those who are not, we expect it to be evident in our subject population. However, this may not be a sufficiently prolonged stimulus to elicit differences in cerebral blood flow between these two groups of subjects associated with actual AMS symptoms. Symptoms of AMS at this level of hypoxia peak only after 24–48 hours of exposure, and exercise may also be a factor. However, it would be important to extend our measurements and to continue the exposure for a further 48 hrs and observe if the blood flow change is sustained, or if changes do correlate with the eventual development of AMS symptoms.

4.3 Advantages of arterial spin labeling for CBF quantitation

Arterial spin labeling MRI used in our measurements offers a number of key advantages over existing techniques for probing the underlying physiology of the hypoxic cerebral response. The arterial spin labeling technique magnetically tags blood water and measures its delivery to tissue capillaries, to obtain a measure of tissue perfusion in units of ml blood/100g tissue/minute (Figure 1). These magnetically-tagged protons act as an endogenous diffusible “tracer” in an analogous manner to tagged water in ¹⁵O PET studies (Buxton 2002). For modeling the cerebral response to hypoxia these quantitative measurements of tissue perfusion are a more relevant quantity than surrogates of perfusion such as proximal artery velocity (Buxton 2004). Another strength of MRI measurements of CBF is the ability to quantify the regional flow (rather than just a global estimate of blood flow). Although this is a new application for investigating the cerebral hemodynamics in hypoxia as they relate to acute mountain sickness, the arterial spin labeling method has already been used for other investigations of cerebral hemodynamics, and been shown to be both a valid (Walsh et al. 1994; Tsekos et al. 1998; Zhou et al. 2001) and reliable (Yen et al. 2002; Liu et al. 2004; Parkes et al. 2004) measure of cerebral blood flow. In our own studies we have shown measures of baseline CBF to be highly reproducible with test-retest r^2 of 0.96 (n=10, unpublished data). In this study, data were averaged spatially over ~1,500 voxels for grey matter and ~2,000 for white matter, and the individual 4-second acquisitions were also averaged over 5-minute time periods. Thus despite the relatively low SNR per acquisition for the serially-subtracted arterial spin labeled images, the SNR for our blood flow measurements in white and grey matter regions of interest are large (12.6 and 21.6 respectively) due to this additional temporal and spatial averaging.

4.4 Limitations of arterial spin labeling in white matter

Our choice of pulse sequence parameters for these studies were based on previous optimizations of the QUIPSS II technique for quantifying cerebral blood flow in grey matter (Wong et al. 1998; Restom et al. 2006). Blood flow in white matter is significantly lower than in grey matter, and so satisfying the QUIPSS II conditions in white matter would ideally require specific optimization of the timing parameters (TI1, TI2, TR) (Wong et al. 1998). It is thus likely we are sensitive to transit delays in our white matter blood flow data. This may result in an underestimation of the absolute blood flow values (the magnitude of this error being dependent upon the actual blood flow value). Thus comparisons of quantitative values between grey matter and white matter need to be treated with some caution. However, despite choosing

timing parameters optimized primarily for grey matter, the percent changes in cerebral blood flow with hypoxia were of the same order of magnitude for both tissue types. Also, the quantitative white matter blood flow values are consistent with values in the literature (Calamante et al. 1999). We observed a large intersubject variability in resting CBF. This has been noted in prior studies in human subjects, and is sensitive to the choice of region of interest (Leontiev et al. 2007). Our use of an anatomically-defined single region of interest that encompasses all grey matter (or white matter) may be contributing to the intersubject variation in resting CBF in this study. For these studies, we were primarily interested in detecting differences between the subject groups with hypoxic exposure rather than the quantitative cerebral blood flow values in WM compared to GM. Thus with the repeated measures design of this study, where each subject acts as their own control such uncertainties are minimized.

4.5 Comparison with other measures of cerebral blood flow

The mean increase in global cerebral blood flow of 13% in response to acute hypoxia that we observed in this study is comparable to some of the prior studies in the literature. Berre (Berre et al. 1999) demonstrated a 20–23% increase in middle cerebral artery velocity with hypoxia but found no significant difference between AMS-susceptible and AMS-resistant subjects. Using ^{15}O - H_2O PET Buck (Buck et al. 1998) reported no change in quantified global cerebral blood flow after acute (20 minute) exposure to 3,000 m, but a 36% increase ($+7.2 \text{ ml}/100\text{g}/\text{min}$) at 4,500 m (i.e. a greater hypoxic stimulus than the present study), although the blood flow was not compared with AMS symptoms. Using a ^{133}Xe technique, Jensen (Jensen et al. 1990) reported a 24% increase in global cerebral blood flow at 3,475 m after 24 hrs in all subjects irrespective of development of AMS.

Other reports of blood flow change in hypoxia are more varied, differing by technique as well as duration and altitude of exposure. Cerebral blood flow has been reported to show no change following acute hypoxia using SPECT radionucleotide imaging (Pagani et al. 2000), and no change in middle cerebral artery velocity after 24 hrs hypoxia (Van Osta et al. 2005). There are also reports of a fall in middle cerebral artery velocity after 24 hours hypoxia (Poulin et al. 2002). Other studies report very variable hemodynamics with no change in acute basilar artery velocity for hypoxia as low as 8%, but at 7% O_2 , a 29% velocity decrease in climbers (presumably AMS-resistant) but a 28% increase in controls (Jansen et al. 2002). Another study reported a 20 % increase in internal cerebral artery and vertebral artery velocity but only when hypoxic duration exceeded 18 hrs (Huang et al. 1987). Several studies at 4559 m (a more hypoxic stimulus than used in the current study) have also shown varied responses ranging from 36% decrease to 45% increase in middle cerebral artery velocity with 3 hours of acute hypoxia (Baumgartner et al. 1999), or increased middle cerebral artery velocity after 24–48 hrs with a 48% increase in subjects with AMS, compared to only 27% increase in subjects without AMS (Baumgartner et al. 1994).

Although universally referred to as a measure of “cerebral blood flow”, these methods are not measuring exactly the same physiological process. Inert gas techniques (Severinghaus et al. 1966) and xenon-133 (Jensen et al. 1990) make measurements of global or broadly regional perfusion. Doppler sonography measures arterial velocity in the proximal intracranial or neck arteries (Reeves et al. 1985; Huang et al. 1987; Ter Minassian et al. 2001; Jansen et al. 2002; Poulin et al. 2002; Van Osta et al. 2005) and radionucleotide Single Photon Computed Emission Tomography - SPECT (Pagani et al. 2000) or Positron Emission Tomography - PET (Buck et al. 1998) map the quantified or relative regional cerebral perfusion (similar to our MRI measurements). Thus, fundamental differences in the experimental design of prior studies combined with disparity in the hemodynamic parameters being measured make direct comparison of the current and prior studies difficult.

4.6 Acute-phase hemodynamic response and O₂ delivery

In this study we observed that mean baseline blood flow in grey matter is two-fold higher than in white matter (57.3 ml/100g/min in grey matter compared with 31.4 ml/100g/min in white matter), and mirrors the ~2 fold greater oxygen metabolism in grey matter than white matter (Pantano et al. 1984). Global measures of blood flow will thus be dominated by grey matter flow, and have the potential to miss smaller changes in white matter blood flow. By making simultaneous regional measurements of absolute change in blood flow in white matter and in grey matter we have been able to test our hypothesis of differential flow responses to acute hypoxia in white matter and grey matter. Previous studies have observed edema of vasogenic origin in association with clinical symptoms of AMS (Hackett 1999), or late in disease progression (>6 hours), even without clinical AMS symptoms (Schoonman et al. 2007). The expected differences in regional hemodynamics in white matter were not seen despite a predilection to vasogenic cerebral edema for white matter. One possible explanation may be that changes in regional hemodynamics (and hence vulnerability to developing AMS) may be a late feature, and our acute measurements may be too early to detect this.

Prior studies have shown that cerebral oxygen delivery is maintained over several hours of exposure to hypoxia in humans (Wolff 2000) and in animals (Nioka et al. 1990). During 30 minutes of acute 12.5% hypoxia we observed no significant decrease in cerebral oxygen delivery to either white matter or grey matter regions, indicating that cerebral oxygen delivery is maintained during the acute-phase response to acute hypoxia as well.

4.7 Poikilocapnic hypoxia

For this study we chose a poikilocapnic hypoxic stimulus to more closely reflect the actual physiological stimulus of acute altitude exposure. In this setting, the potential for increased cerebral blood flow stimulated by hypoxia may be inhibited by the hypoxic ventilatory response and increased alveolar ventilation. Hyperventilation causes reduced arterial pCO₂, and thus a reduced pCO₂, which would tend to reduce cerebral blood flow. While an isocapnic hypoxic stimulus would help elucidate the potential magnitude of the cerebral blood flow responses to hypoxia alone, any influences of the acute hypocapnia on the cerebral response would be negated. The aim of this study was to look for altered cerebral physiology that may indicate a selective vulnerability to developing AMS, and thus we chose a poikilocapnic hypoxic stimulus to more closely mirror the actual physiological stimulus that eventually leads to AMS.

4.8 Statistical power of the study

An issue that needs to be addressed is whether the present study has sufficient power to justify the statistical conclusions reached. One approach is to calculate *a priori* the number of subjects that would be required to detect a statistically significant difference in future studies, based on the observed changes in cerebral blood flow with acute hypoxia that were observed in the present study (if, in fact, it were present in the population). Assuming a 2 tailed test, $p < 0.05$ and a power of 0.8, 123 subjects per group would be required based on the observed changes in grey matter and 746 subjects per group would be required based on the observed changes in white matter. This suggests that the true difference between groups is likely very small. Additionally, based on previous published data we hypothesized that the increase in cerebral blood flow in acute hypoxia would be greater in AMS-susceptible subjects than in AMS-resistant subjects, which was not the case. In fact, the changes in both grey matter and white matter cerebral blood flow were numerically less (although statistically not significant) in AMS-susceptible subjects than AMS resistant subjects further strengthening our conclusions.

4.9 Summary

In conclusion, increased cerebral blood flow was seen in grey matter and white matter in response to 30 minutes of 12.5% hypoxia, with no significant change in cerebral oxygen delivery rate. The increase in cerebral blood flow with acute hypoxia was not greater in AMS-susceptible compared to AMS-resistant, subjects. In addition the flow response in grey matter and white matter paralleled each other, with similar percentage changes. These findings, unlike those in high altitude pulmonary edema, do not demonstrate altered hemodynamics in early hypoxia as defining a specific vulnerability to developing AMS.

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Figure 1.

MRI of cerebral anatomy and blood flow. Upper row shows map of regional cerebral perfusion for 5 slices through the cerebrum (scale is ml/100g/min, right side of the brain is on the left of the image). Second row is corresponding T1-weighted anatomical image used to define the grey matter and white matter regions of interest. Rows 3 and 4 show the grey matter and white matter masks (containing pixels with >70% probability of containing grey or white matter respectively). The masks were applied to the blood flow map maps to calculate mean white matter and grey matter perfusion.

Figure 2.

Mean ($\pm$ SD) oxygen saturation (SaO₂) and Mean heart rate over 5-minute intervals during 70 minute paradigm. 30-minutes of 12.5% hypoxia is indicated by the grey bar. Mean SaO₂ decreased significantly with hypoxia and returned to baseline during post-hypoxic normoxia ($p < 0.0001$) but was not significantly different between groups ($p = 0.31$). Mean heart rate increased significantly with hypoxia and returned to baseline during post-hypoxic normoxia ($p < 0.0001$) but was also not significantly different between groups ($p = 0.55$).

Figure 3.

Mean ($\pm$ SD) absolute global cerebral blood flow in the cerebrum measured by arterial spin labeling MRI averaged over 5-minute intervals during 70 minute paradigm. 30-minutes of 12.5% hypoxia is indicated by the grey bar. Global cerebral blood flow increased significantly during hypoxia and returned to baseline during post-hypoxic normoxia ($p < 0.0001$), but was not different between groups ($p = 0.75$). Dashed line indicates mean baseline CBF during normoxia.

Figure 4.

Mean ($\pm$ SD) absolute cerebral blood flow in grey matter measured by arterial spin labeling MRI averaged over 5-minute intervals during 70 minutes minute paradigm. 30-minutes of 12.5% hypoxia is indicated by the grey bar. Grey matter cerebral blood flow increased significantly during hypoxia and returned to baseline during post-hypoxic normoxia ($p < 0.0001$), but this change was not different between groups ($p = 0.71$). Dashed line indicates mean baseline CBF during normoxia.

Figure 5.

Mean ($\pm$ SD) absolute cerebral blood flow in white matter averaged over 5-minute intervals during the 70 minute paradigm. 30-minutes of 12.5% hypoxia is indicated by the grey bar. White matter cerebral blood flow increased significantly during hypoxia and returned to baseline during post-hypoxic normoxia ($p < 0.0001$), but this change was not different between groups ($p = 0.89$). Dashed line indicates mean baseline CBF during normoxia.

Table 1

Subject Characteristics

Subject	Age	Sex	Group	Most recent Lake Louise Score	Frequency of prior symptoms (%)	Number of prior visits >2400m	Number of prior visits >4200m	Prior max elevation reached (m)
1	34	F	AMS-S	19	100	>20	10	4300
2	34	F	AMS-S	16	100	10	0	3050
3	32	M	AMS-S	9	100	>20	10	4300
4	28	M	AMS-S	7	100	10	10	4300
5	27	F	AMS-S	12	66	>20	1	4300
6	55	F	AMS-S	14	100	3	1	5940
Mean (SD)	35 (10)			13 (4)	94 (14)	14 (7)	5 (5)	4210 (680)
7	36	M	AMS-R	0	0	>20	>20	6700
8	27	F	AMS-R	4	0	>20	>20	5600
9	32	F	AMS-R	0	0	>20	>20	6400
10	23	M	AMS-R	0	0	>20	10	4300
11	27	M	AMS-R	0	0	>20	0	4300
12	42	M	AMS-R	2	0	>20	>20	5800
Mean (SD)	32 (7)			1 (2)	0 (0)	>20 (0)	15 (8)	5510 (1040)
p	0.44				-	0.06	< 0.05	< 0.05

AMS-S = subjects susceptible to AMS, AMS-R = subjects resistant to AMS. Symptom frequency are symptoms related to possible AMS (headache, nausea, fatigue, dizziness, insomnia, confusion, ataxia, edema, malaise) averaged over prior sejours >3,800m altitude. Lake Louise Score was retrospectively assigned based on subject's most recent symptoms at altitude. P value is comparison between AMS- groups. Values are mean ($\pm$ SD)

Table 2

Steady State Cerebral Blood Flow (ml/100g/min)

Brain Region	Subject Group	Normoxia	Hypoxia	P (F _I O ₂)	p (Group × F _I O ₂)
Global	AMS-S	42.7 (14.0)	47.8 (19.5)	< 0.005	0.65
	AMS-R	41.7 (10.1)	48.2 (10.1)		
Gray Matter	AMS-S	53.9 (18.5)	60.0 (24.9)	< 0.005	0.57
	AMS-R	53.6 (13.1)	61.9 (12.2)		
White Matter	AMS-S	32.0 (9.4)	36.2 (13.5)	< 0.005	0.82
	AMS-R	30.8 (6.8)	35.7 (7.7)		

AMS-S = subjects susceptible to AMS, AMS-R = subjects resistant to AMS. p (F_IO₂) = main effect for F_IO₂, p (Group × F_IO₂) = Group by F_IO₂ interaction. Values are mean (± SD)