**Table S1.** Stopping rules

**Protocol withdrawal rules will be as follows:**

* Desaturation: SpO2<70% for more than 1 minute not immediately responsive to oxygen therapy
* Chest pain with evidence of ischemia on EKG (e.g., T-wave inversions, ST segment changes)
* Signs and symptoms of acute pulmonary edema:
  + Rapidly progressive, severe shortness of breath at rest
  + Severe dyspnea, or a feeling of suffocating or drowning despite return to normoxia
  + Wheezing or gasping for breath accompanied with anxiety, restlessness or a sense of apprehension
  + A cough that produces frothy sputum that may or may not be tinged with blood
* Signs and symptoms of acute cerebral edema:
  + Acute ataxia or altered mental status
  + Any focal neurologic deficit
  + Increase in optic nerve diameter >4mm compared to the baseline value (first measurement at room air) associated with neurologic deficits suggesting cerebral edema
* Change in mental status as determined by mental status exam
* Increase in systolic pulmonary artery pressure by 15 mmHg from baseline (first transthoracic echocardiogram in ambient air) associated with either TAPSE < 1.6 cm or B lines > 12
* Systemic blood pressure increase to 150/100 mmHg or increase by 40% from baseline, whichever is lower
* Syncope

##### Febrile illness (> 100.4 F on two consecutive measurements 1 hour apart)

**Table S2.** Pre-2018 Lake Louis Acute Mountain Sickness Score Questionnaire

1. HEADACHE: 5) DIFFICULTY SLEEPING:
   1. No Headache 0. Slept as well as usual
   2. Mild Headache 1. Did not sleep as well
   3. Moderate Headache 2. Woke many times
   4. Sever, incapacitating headache 3. Could not sleep at all
2. GI (STOMACH): 6) CHANGE IN MENTAL STATUS
   1. No GI symptoms 0. No change
   2. Poor appetite, mild nausea 1. Lethargy/lassitude
   3. Moderate nausea or vomiting 2. Disoriented/confused
   4. Severe N&V, incapacitating 3. Stupor/semiconscious
3. FATIGUE/WEAKNESS: 7) ATAXIA (HEEL-TOE WALKING):
   1. Not tired or weak 0. No ataxia
   2. Mild fatigue/weakness 1. Movement to maintain balance 2. Steps off-line
   3. Moderate fatigue/weakness 3. Falls down
   4. Sever F/W, incapacitating 4. Can’t stand

1. DIZZY/LIGHTHEADED:
   1. Not dizzy 8) PERIPHERAL EDEMA:
   2. Mild dizziness 0. No edema
   3. Moderate dizziness 1. One location
   4. Severe, incapacitating 2. Two or more locations

**QUESTIONS TO ASSESS MENTAL STATUS:**

1. Orientation: Ask for name, date, location, president. **INTACT? (yes / no)**
2. Attention: Ask to spell WORLD or other 5-letter word forwards and backwards. **INTACT? (yes / no)**
3. Naming: Point to a body part and ask to name it (e.g. wrist). **INTACT? (yes / no)**
4. Calculation: Ask simple calculation with money (e.g # of quarters in $2.50). **INTACT? (yes / no)**
5. Commands: Give simple command crossing midline (e.g. Touch right thumb to left ear). **INTACT (yes / no)**

**Table S3.** Maximum Lake Louise score by participant

|  |  |  |
| --- | --- | --- |
|  | **Lake Louise Score** | **Symptoms** |
| **Subject 1** | 1 | Mild headache |
| **Subject 2** | 1 | Did not sleep as well |
| **Subject 3** | 1 | Mild headache |
| **Subject 4** | 0 | No complaints |
| **Subject 5** | 2 | Woke many times/Did not sleep as well |

Lake Louise Score: A score of 3 – 5 constitutes mild Acute Mountain Sickness (AMS), and a score of 6 or more constitutes severe AMS.

**Table S4. Daily** average of right and left optic nerve sheath diameters (mm) at various FiO2 levels

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | **Day 1** | **Day 2** | **Day 3** | **Day 4** | **Day 5** | **Day 6** |
| **FiO2** | **0.21** | **0.16** | **0.15** | **0.14** | **0.13** | **0.11** |
| **Subject 1** | 4.55 | 4.7 | 4.55 | 4.55 | 4.67 | 4.57 |
| **Subject 2** | 5.0 | 4.65 | 4.9 | 4.95 | 4.9 | 5.02 |
| **Subject 3** | 5.04 | 5.0 | 4.65 | 5.15 | 5.0 | 5.23 |
| **Subject 4** | 5.95 | - | 5.85 | 5.85 | 5.65 | 5.85 |
| **Subject 5** | 6.1 | 6.1 | 6.2 | - | 6.1 | 6.15 |

**Table S5.** Total number of B lines measured twice daily at different FiO2 (%)

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **Day 1** | | **Day 2** | | **Day 3** | | **Day 4** | | **Day 5** | | **Day 6** | |
| **FiO2** | **0.21** | **0.14** | **0.16** | **0.13** | **0.15** | **0.12** | **0.14** | **0.11** | **0.13** | **0.11** | **0.11** | **0.21** |
| **Subject 1** | 0 | 1 | 1 | 2 | 1 | 0 | 2 | 0 | 2 | 0 | 2 | 0 |
| **Subject 2** | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| **Subject 3** | 1 | 4 | 1 | 1 | 0 | 0 | 1 | 2 | 3 | 2 | 3 | - |
| **Subject 4** | 2 | 1 | 2 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 0 |
| **Subject 5** | 0 | 0 | 0 | 0 | 0 | 0 | 2 | 0 | 0 | 0 | 0 | 0 |

**Table S6.** Daily kilocalories consumed

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Timepoint (FiO2 %)** | **Subject 1** | **Subject 2** | **Subject 3** | **Subject 4** | **Subject 5** |
| **Day 1 (14%)** | 2,068 | 1,720 | 1,969 | 1,987 | 2,116 |
| **Day 2 (13%)** | 2,038 | 1,495 | 1,941 | 1,977 | 2,074 |
| **Day 3 (12%)** | 2,269 | 1,738 | 1,850 | 1,914 | 2,320 |
| **Day 4 (11%)** | 2,378 | 1,737 | 1,988 | 1,965 | 2,299 |
| **Day 5 (11%)** | 2,530 | 1,759 | 1,930 | 2,017 | 2,206 |
| **Day 6 (21%)** | 2,286 | 1,711 | 1,873 | 2,005 | 2,151 |
| **Day 7 (21%)** | 2,480 | 1,743 | 1,920 | 1,999 | 2,117 |
| **Mean (SD)** | **2,293 (189.1)** | **1,700 (91.9)** | **1,924 (49.2)** | **1,981 (34.1)** | **2,183 (95.3)** |

**Table S7.** Daily urine output, total output, and fluid balance for Study Subject 5.

|  |  |  |  |
| --- | --- | --- | --- |
| **Timepoint (FiO2 %)** | **Urine (ml)** | **Total Output (ml)** | **Fluid Balance (ml)** |
| Day 1 (14-16%) | 3,115 | 3,515 | -2,035 |
| Day 2 (13-15%) | 2,475 | 2,875 | -1,035 |
| Day 3 (12-14%) | 4,785 | 5,185 | 15 |
| Day 4 (11-13%) | 2,075 | 3,875 | -255 |
| Day 5 (11%) | 2,445 | 3,245 | -165 |
| Day 6 (21%) | 2,720 | 1,870 | 850 |

**Table S8.** Right ventricular systolic pressure (mmHg) measured by transthoracic echocardiography at various FiO2.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | **Day 1** | **Day 2** | **Day 3** | **Day 4** | **Day 5** | **Day 6** |
| **FiO2** | **0.21** | **0.16** | **0.15** | **0.14** | **0.13** | **0.11** |
| **Subject 1** | 27 | - | 27 | 33 | 36 | 33 |
| **Subject 2** | 24 | 34 | 27 | 37 | - | 34 |
| **Subject 3** | 34 | 35 | 30 | 33 | 33 | 35 |
| **Subject 4** | 19 | - | 27 | 29 | 34 | 35 |
| **Subject 5** | 19 | 27 | 28 | - | - | 24 |

**Table S9.** Mean systolic (SBP) and diastolic (DBP) blood pressures (mmHg) throughout the study period breathing various levels of FiO2.

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Day**  **(FiO2)** | Subject 1  Mean SBP(SD) | Subject 1  Mean  DBP(SD) | Subject 2  Mean SBP(SD) | Subject 2  Mean DBP(SD) | Subject 3  Mean SBP(SD) | Subject 3  Mean DBP(SD) | Subject 4  Mean SBP(SD) | Subject 4  Mean DBP(SD) | Subject 5  Mean SBP(SD) | Subject 5  Mean  DBP (SD) |  |  |  |  |  |  |  |  |  |
| **Day 1 (14%)** | 122 (6.9) | 69 (11.5) | 115 (4.9) | 76 (2.9) | 134 (15.3) | 80 (6.7) | 113 (9.3) | 63 (8.5) | 115 (5.4) | 66 (6) |  |  |  |  |  |  |  |  |  |
| **Day 2 (13%)** | 120 (6.1) | 63 (5) | 117 (6.1) | 81 (3.3) | 143 (4.9) | 80 (3.9) | 129 (4.7) | 75 (3) | 116 (10.8) | 65 (1.7) |  |  |  |  |  |  |  |  |  |
| **Day 3 (12%)** | 112 (4.3) | 64 (5.6) | 117 (3.2) | 81 (5.3) | 137 (6.6) | 87 (7.5) | 133 (3.7) | 77 (6.3) | 107 (7.8) | 61 (5.3) |  |  |  |  |  |  |  |  |  |
| **Day 4 (11%)** | 123 (6.9) | 72 (5.9) | 116 (8.4) | 76 (7.9) | 137 (9.7) | 80 (9.2) | 135 (7.2) | 78 (3.8) | 118 (3.7) | 70 (2.9) |  |  |  |  |  |  |  |  |  |
| **Day 5 (11%)** | 123 (11.9) | 72 (3.3) | 116 (6.3) | 76 (2.6) | 137 (5.7) | 80 (12.7) | 135 (6.3) | 78 (6.2) | 118(27.6) | 70 (10.1) |  |  |  |  |  |  |  |  |  |
| **Day 6 (21%)** | 124 (7.5) | 75 (8.4) | 113 (16.9) | 73 (9.8) | 127 (5.2) | 79 (7.9) | 135 (8.8) | 74 (7.3) | 118 (8.3) | 74 (10.1) |  |  |  |  |  |  |  |  |  |
| **Day 7 (21%)** | 121 | 72 | 115 | 74 | 123 | 71 | 124 | 80 | 118 | 64 |  |  |  |  |  |  |  |  |  |

\*The average systolic/diastolic blood pressure measurements were taken over the study period and consist of at least five measurements, except for Day 7 where only one measurement was taken.

**Table S10.** Statistical analysis of Reticulocyte count (%) over the study period.

|  |  |  |  |
| --- | --- | --- | --- |
| **Dunn’s Multiple** | **Mean rank diff.** | **Significant?** | **Adjusted P value** |
| 0 vs. 24 | 0.7000 | No | >0.9999 |
| 0 vs. 48 | -3.0000 | No | >0.9999 |
| 0 vs. 72 | -6.0000 | No | >0.9999 |
| 0 vs. 96 | -11.10 | No | >0.9999 |
| 0 vs. 120 | -19.70 | No | 0.1725 |
| 0 vs. 144 | -22.70 | Yes \* | 0.0447 |
| 0 vs. 168 | -26.10 | Yes \* | 0.0174 |
| 24 vs. 48 | -3.700 | No | >0.9999 |
| 24 vs. 72 | -6.700 | No | >0.9999 |
| 24 vs. 96 | -11.80 | No | >0.9999 |
| 24 vs. 120 | -20.40 | No | 0.1277 |
| 24 vs. 144 | -23.40 | Yes \* | 0.0319 |
| 24 vs. 168 | -26.80 | Yes \* | 0.0124 |
| 48 vs. 72 | -3.000 | No | >0.9999 |
| 48 vs. 96 | -8.100 | No | >0.9999 |
| 48 vs. 120 | -16.70 | No | 0.5665 |
| 48 vs. 144 | -19.70 | No | 0.1725 |
| 48 vs. 168 | -23.10 | No | 0.0689 |
| 72 vs. 96 | -5.100 | No | >0.9999 |
| 72 vs. 120 | -13.70 | No | >0.9999 |
| 72 vs. 144 | -16.70 | No | 0.5665 |
| 72 vs. 168 | -20.10 | No | 0.2357 |
| 96 vs. 120 | -8.600 | No | >0.9999 |
| 96 vs. 144 | -11.60 | No | >0.9999 |
| 96 vs. 168 | -15.00 | No | >0.9999 |
| 120 vs. 144 | -3.000 | No | >0.9999 |
| 120 vs. 168 | -6.400 | No | >0.9999 |
| 144 vs. 168 | -3.400 | No | >0.9999 |

**Table S11.** Statistical analysis for Erythropoietin levels over the study period.

|  |  |  |  |
| --- | --- | --- | --- |
| **Dunn’s Multiple** | **Mean rank diff.** | **Significant?** | **Adjusted P value** |
| 0 vs. 24 | -6.800 | No | >0.9999 |
| 0 vs. 48 | -7.800 | No | >0.9999 |
| 0 vs. 72 | -14.50 | No | 0.1378 |
| 0 vs. 96 | -16.90 | Yes \* | 0.0359 |
| 0 vs. 120 | -24.20 | Yes \* | 0.0002 |
| 24 vs. 48 | -1.000 | No | >0.9999 |
| 24 vs. 72 | -7.700 | No | >0.9999 |
| 24 vs. 96 | -10.10 | No | >0.9999 |
| 24 vs. 120 | -17.40 | Yes \* | 0.0266 |
| 48 vs. 72 | -6.700 | No | >0.9999 |
| 48 vs. 96 | -9.100 | No | >0.9999 |
| 48 vs. 120 | -16.40 | Yes \* | 0.0482 |
| 72 vs. 96 | -2.400 | No | >0.9999 |
| 72 vs. 120 | -9.700 | No | >0.9999 |
| 96 vs. 120 | -7.300 | No | >0.9999 |

**Table S12.** Statistical analysis of SpO2, HR and RR over the study period.

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **SpO2 day** | | **SpO2 night** | | **HR day** | | **HR night** | | **RR day** | | **RR night** | |
|  | **τ (n)** | **p** | **τ (n)** | **p** | **τ (n)** | **p** | **τ (n)** | **p** | **τ (n)** | **p** | **τ (n)** | **p** |
| ***Subject 1*** | -0.56 (15) | 0.004\* | -0.83 (16) | <0.001\* | 0.46 (15) | 0.017\* | 0.46 (11) | 0.050 | -0.53 (15) | 0.012\* | -0.34 (11) | 0.201 |
| ***Subject 2*** | -0.46 (15) | 0.019\* | -0.49 (15) | 0.012\* | 0.30 (14) | 0.139 | 0.11 (14) | 0.584 | 0.67 (14) | 0.002\* | 0.52 (14) | 0.018\* |
| ***Subject 3*** | -0.42  (14) | 0.045\* | -0.69 (12) | 0.002\* | 0.39 (13) | 0.066 | 0.23 (12) | 0.303 | -0.22 (14) | .344 | 0 (12) | >0.99 |
| ***Subject 4*** | -0.54 (15) | 0.006\* | -0.55 (16) | 0.003\* | 0.39 (15) | 0.046\* | 0.40 (16) | 0.031\* | -0.58 (14) | 0.006\* | -0.32 (5) | 0.449 |
| ***Subject 5*** | -0.70 (15) | <0.001\* | -0.59 (16) | 0.002\* | 0.47 (15) | 0.015\* | 0.02 (16) | 0.928 | 0.77 (5) | 0.083 | / | / |

**\*Significant**

Data are presented as Kendall’s τ, together with the number of observations per subject. HR: heart rate; RR: respiratory rate; SpO2: transcutaneous peripheral saturation of oxygen

**Figure S1.** Representative study environment consisting of a tent filled with hypoxic air and furnished with a bed, chair, and small table

**A room with a bed and a monitor

Description automatically generated**

**Figure S2.** Respiratory rate (breaths per minute) was not affected by changes in FiO2.

A picture containing diagram, line, plot, font

Description automatically generated

Respiratory rate (breaths per minute) was recorded for each subject at vital sign checks every four hours throughout study protocol. Time depicted as protocol day or night with corresponding target fraction of inspired oxygen.

**Figure S3.** Serum creatinine (mg/dl) levels measured each morning during the study period.

**A graph of different colored lines

Description automatically generated**

**Figure S4.** Body weight (kg) over the study period.

**A graph of different colored lines

Description automatically generated**

**Figure S5.** Serum bicarbonate decreases with decreasing FiO2 and gradually increases to baseline after return to normoxia.



Serum bicarbonate levels (mEq/L) were measured throughout study protocol. Time depicted as protocol day when measurement was obtained with corresponding targeted fraction of inspired oxygen at time of sample. Multiple samples were obtained on days 5, 6, and 7, either at the very end or very start of the protocol day.

**Figure S6.** Urine sodium and chloride levels decrease after initiation of hypoxia and remain low through the remainder of study protocol, including after return to normoxia.



Urine sodium and urine chloride levels (mEq/L) were measured daily during the study protocol. Time depicted as protocol day when measurement was obtained with corresponding targeted fraction of inspired oxygen at time of sample.

**Figure S7.** Lactic acid levels remained within normal limits throughout the study

****

Serum lactic acid (mmol/L) was measured daily throughout study protocol. Time is depicted as protocol day when measurement was obtained along with corresponding targeted fraction of inspired oxygen. Multiple samples were obtained on days 5, 6, and 7, either at the very end or very start of the protocol day.

**Figure S8.** Beta-hydroxybutyrate levels rise with decreasing fraction of inspired oxygen, remain elevated immediately upon return to normoxia, and return to baseline five hours after return to normoxia



Serum beta-hydroxybutyrate (mg/dL) was measured daily during the study protocol. Time depicted as protocol day when measurement was obtained with corresponding targeted fraction of inspired oxygen at time of sample. Multiple samples were obtained on days 5, 6, and 7, either at the very end or very start of the protocol day.