THE EFFECTS OF SUSTAINED HEAVY EXERCISE ON
THE DEVELOPMENT OF PULMONARY INTERSTITIAL EDEMA
IN TRAINED MALE CYCLISTS

by

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# ABSTRACT

The transport of $O_2$ from alveolus to pulmonary capillary has not typically been thought of as the limiting step in aerobic performance. It has been demonstrated that fit athletes are able to, at high workloads, elicit a decreased arterial $O_2$ saturation to levels below 90%. This showed that healthy, fit individuals were able to exceed the capacity of the pulmonary system, and was termed exercise-induced-hypoxemia (EIH). The possible mechanisms for EIH include veno-arterial shunts, $\dot{V}_A/\dot{Q}_c$ mismatch, relative alveolar hypoventilation, decreased pulmonary transit time, and pulmonary edema. This study looked for increases in extravascular water (EW) after a 45-minute intense exercise bout as evidence of pulmonary edema. The subjects were 8 highly trained males (mean ± SD: age; $26.9 \pm 3.0$ years, height; $179.9 \pm 5.7$ cm, weight; $76.1 \pm 6.5$ kg) who performed three tests used to indicate differences pre and post exercise. The testing involved measurements to ensure normal spirometry (FVC; $6.07 \pm 1.14$ l, $FEV_1 \cdot FVC^{-1}$; $79.0 \pm 9.2$ %) and sufficient fitness ($\dot{V}O_2$ max = $63.7 \pm 2.63$ ml·min$^{-1}$·kg$^{-1}$). During intervention testing, subjects completed a 45-minute bout of maximum sustainable cycling activity, pre and post pulmonary diffusion measures, and pre and post magnetic resonance imaging. Subjects exercised at 10% below their ventilatory threshold for 45 minutes at a power output of $300 \pm 25$ watts. Diffusion for carbon monoxide ($DL_{CO}$) and lung capillary volume ($V_C$) had decreased one hour post exercise by 12% (p = 0.004) and 21% (p = 0.017), respectively, but no significant change in membrane diffusing capacity ($D_M$) was found. The magnetic resonance (MR) scans showed a 9.4% increase (p = .043) in pulmonary extravascular water after exercise, consistent with the theory that EW is

produced in well trained subjects. This study was the first to use new MR advances to

show an increase in EW following long duration heavy exercise in trained male subjects.

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## LIST OF ABBREVIATIONS AND SYMBOLS

| | |
|---|---|
| $\Delta D_M$ | change in membrane diffusing capacity |
| $\Delta EW$ | change in extravascular water |
| $\theta$ | rate of combination of hemoglobin with oxygen |
| $A\text{-}aDO_2$ | alveolar-arterial oxygen pressure difference |
| bpm | beats per minute |
| CO | carbon monoxide |
| CT | computed tomography |
| $D_L$ | lung diffusing capacity |
| DL-21% | pulmonary diffusing capacity for carbon monoxide with 21% oxygen |
| DL-90% | pulmonary diffusing capacity for carbon monoxide with 90% oxygen |
| $DL_{CO}$ | lung diffusing capacity for carbon monoxide |
| $D_M$ | pulmonary membrane diffusing capacity |
| EIH | exercise induced hypoxemia |
| EW | pulmonary extravascular water |
| $FEV_1$ | forced expiratory volume in the first second |
| FRC | functional residual capacity |
| FVC | forced vital capacity |
| MR | magnetic resonance imaging |
| MVV | maximum voluntary ventilation |
| $N_{ex}$ | number of signal averages |

| | |
|---|---|
| $p_aCO_2$ | arterial carbon dioxide pressure |
| $P_aO_2$ | arterial oxygen pressure |
| $P_AO_2$ | alveolar oxygen pressure |
| $\dot{Q}$ | cardiac output |
| $\dot{Q}_C$ | pulmonary capillary perfusion |
| RER | respiratory exchange ratio |
| rf | radio-frequency |
| $SaO_2$ | oxygen saturation with hemoglobin |
| $SaO_2min$ | minimum oxygen saturation with hemoglobin |
| SNR | signal to noise ratio |
| $T_1$ | longitudinal relaxation time |
| $T_2$ | transverse relaxation |
| TR | repetition time |
| $\dot{V}_A$ | alveolar ventilation |
| $\dot{V}_A/\dot{Q}_C$ | alveolar ventilation to pulmonary capillary perfusion ratio |
| $V_C$ | pulmonary capillary volume |
| $\dot{V}CO_2$ | carbon dioxide production |
| $\dot{V}_E$ | minute ventilation |
| VT | ventilatory threshold |
| $\dot{V}O_2$ | oxygen uptake |
| $\dot{V}O_2\,max$ | maximum oxygen uptake |
| W/D ratio | wet to dry ratio |

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# INTRODUCTION

Maximum oxygen uptake ($\dot{V}O_2$ max) has been well established as an important factor in performance involving aerobic metabolic pathways. The transport of $O_2$ from alveolus to pulmonary capillary has not typically been thought of as the limiting step in $O_2$ transport (Asmussen and Neilsen 1960; Saltin et al., 1968). Outward and easily measurable changes accompanying increased physical training typically involve the cardiovascular and musculoskeletal systems, corroborating that view. In 1984, Dempsey *et al.* demonstrated that fit athletes were able to, at high workloads, elicit a decreased arterial saturation of hemoglobin with $O_2$ ($SaO_2$) to levels below 90% associated with a reduction in arterial $O_2$ ($p_aO_2$). This discovery showed that healthy, very fit individuals were able to exceed the capacity of the pulmonary system. This suggests the pulmonary system has remained unchanged despite adaptations of the other systems. This condition, termed exercise induced hypoxemia (EIH), has been corroborated by other studies (Hopkins and McKenzie 1989) and is estimated to effect 50% of highly trained male endurance athletes (Powers et al., 1988; Powers et al., 1993; Martin et al., 1992). EIH has been implicated as a limit to the attainment of $\dot{V}O_2$ max (Lawler et al., 1988; Dodd et al., 1989; Martin and O'Kroy 1993) and maximum performance (Koskolou and McKenzie 1994), making it an important area for study surrounding elite performance.

EIH stems from an increase in the alveolar-arterial $O_2$ pressure difference (A-a$DO_2$) at intense levels of exercise (Dempsey et al., 1984; Hammond et al., 1986; Wagner et al., 1986; Asmussen et al., 1960). The increased A-a$DO_2$, accompanied by hypoxemia in well-trained athletes (Dempsey et al., 1984), suggests a decreased ability to diffuse gas across the pulmonary membrane.

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The change in A-aDO$_2$ associated with exercise can be explained by ventilation-perfusion ($\dot{V}_A/\dot{Q}_C$) inequality (Wagner et al., 1986; Hopkins et al., 1994), relative alveolar hypoventilation (Dempsey et al., 1984), right to left shunts (Wagner et al., 1986), and diffusion limitation (Wagner et al., 1986). Some athletes with high aerobic capacities, may have an inadequate ventilatory response to maximal exercise (Dempsey et al., 1984; Wagner 1992) causing a decreased alveolar O$_2$ pressure (p$_A$O$_2$). Low p$_A$O$_2$ would lower the driving force of O$_2$ transfer across the pulmonary membrane. $\dot{V}_A/\dot{Q}_C$ inequality may also contribute to EIH with matching of ventilation to perfusion becoming less uniform as exercise intensity increases (Gale et al., 1985; Hammond et al., 1986; Hopkins et al., 1994). $\dot{V}_A/\dot{Q}_C$ inequality has been shown to explain 1/3 of the widened A-aDO$_2$ at sea level (Wagner et al., 1986) and more than 60% in well-trained subjects (Hopkins et al., 1994). During maximal exercise, cardiac output ($\dot{Q}$) increases to $\sim 33$ l·min$^{-1}$ (Hopkins et al., 1994) and may reach 40 l·min$^{-1}$ (Ekblom and Hermansen 1968). Veno-arterial shunts do not have a significant impact on overall gas exchange(Wagner et al., 1986). The most significant contribution to increases in A-aDO$_2$ at sea level appears to be from diffusion limitation (Wagner et al., 1986).

A reduction in diffusing capacity of the lung (D$_L$) following exercise has been reported by numerous authors (Miles et al., 1983; Rasmussen et al., 1988; Manier et al., 1993; Hanel et al., 1994; Sheel et al., 1998; McKenzie et al., 1998). D$_L$ may be partitioned into 2 components: the pulmonary membrane diffusing capacity (D$_M$) and the pulmonary capillary volume (V$_C$). This may be used to further display the cause for post exercise D$_L$ limitation. Sheel *et al.* (1998, in press) showed that much of the change in D$_L$ post exercise was attributable to decreases in V$_C$ and that these effects remained 6 hours post exercise. The values found for D$_M$ are highly

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variable, and have not shown a significant decrease in some studies (Miles et al., 1983; Hanel et al., 1994), but reductions have been reported by others (Sheel et al., 1998).

$D_L$ limitation may be attributable to two mechanisms. The first possible mechanism is an incomplete equilibration of $p_aO_2$ with $p_AO_2$ due to a short red blood cell exposure in the pulmonary capillaries. This may occur due to the concommittant expansion of pulmonary capillary blood volume with increasing $\dot{Q}$, reaching its anatomical limit while $\dot{Q}$ continues to increase (Dempsey et al., 1984; Hopkins et al., 1994). The accumulation of pulmonary extravascular water (EW), or pulmonary edema is the second possible mechanism. EW would affect diffusion by increasing the tissue thickness that provides the barrier between alveolar and capillary spaces. The possible mechanism for the accumulation of EW includes increased capillary permeability, increased capillary surface area, increased capillary hydrostatic pressure, or a lymphatic insufficiency (West 1977). The presence of EW in athletes has been suggested by a continued $\dot{V}_A/\dot{Q}c$ mismatch found during 20 minutes of recovery from hypoxic exercise (Schaffartzik et al., 1992).

Previous studies have found acute clinical pulmonary edema in humans caused by heavy exercise at sea level (McKechnie et al., 1979) and moderate exercise at high altitude (Marshall et al., 1971). It has also been suggested that brief intense exercise in athletes with a history suggestive of lung bleeding alters blood-gas barrier function resulting in higher concentrations of red cells and protein in broncho-alveolar lavage fluid (Hopkins et al., 1997). When broncho-alveolar lavage was performed on athletes who had performed sustained submaximal exercise (one hour at 77% of $\dot{V}O_2$ max), erythrocyte content was not different from controls (Hopkins et al., 1998). This suggests that extreme levels of exercise are required to alter the blood-gas barrier integrity (Hopkins et al., 1998). There have also been studies using double indicator-

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dilution techniques (Marshall et al., 1971; Goresky et al., 1972) showing increases in EW during exercise, as well as the use of transthoracic electrical impedance during a 30 minute recovery that suggested elevated intrathoracic fluid volumes (Buono et al., 1982).

Some studies have failed to detect increases in EW (Marshall et al., 1975; Gallagher et al., 1988) possibly due to the small fluid volume (Schaffartzik et al., 1993). Also, in some athletes, there is no increase in $\dot{V}_A/\dot{Q}_c$ inequality during exercise, and pulmonary edema may not occur in such individuals (Schaffartzik et al., 1992). It is also possible that current methods lack the sensitivity to detect the small differences, as would appear to be the case in the indicator-dilution and X-ray techniques (Gallagher et al., 1988).

Experiments with the diffusion capacity of the lung for carbon monoxide ($DL_{CO}$), yield indirect evidence of a diffusion limitation relating to the pulmonary membrane (Manier et al., 1993). A more recent study using computed tomography (CT) to measure changes in EW, corroborated the presence of EW by the visual inspection of the images for linear and polygonal opacities as well as computing lung density (Caillaud et al., 1995). They found an increase in the number of opacities, and an increase in lung density ($p < 0.001$, and $p < 0.0001$, respectively). Currently, technical advances in the use of magnetic resonance imaging (MR) to quantify data has resulted in a more sensitive method of detecting changes in EW (Estilaei et al., 1998). The new MR technique has been used to measure EW in highly trained male cyclists pre and post 5 minutes of heavy exercise, but failed to find a significant increase. It has been postulated that 5 minutes of exercise was not a sufficient insult to the pulmonary tissue, and that to generate a measurable change, exercise of longer duration would be needed. This study investigated the changes in EW following a 45-minute bout of heavy exercise in well-trained cyclists.

**Hypothesis**

A 45-minute bout of heavy cycling will result in the following post-exercise changes in

elite cyclists:

1.    A decrease in $DL_{CO}$, $D_M$ and $V_C$

2.    An increase in lung water.

With a minimum difference of 0.02 $g \cdot ml^{-1}$ being considered a meaningful change in lung

water content, and a standard deviation of 0.015 $g \cdot ml^{-1}$ found in a previous study, the required

number of subjects was eight per group given a conservative power value of 0.75 and alpha set at

$\alpha = 0.05$.  As there were 3 t-tests performed on the $DL_{CO}$ data, alpha was set at $\alpha = 0.05/3$.

# METHODS

## Subjects

Eight trained male cyclists were recruited from local cycling and triathlon clubs.  To be

considered highly trained the subjects had to achieve a $\dot{V}O_2$ max $> 5 \, l \cdot min^{-1}$ or $> 60 \, ml \cdot kg^{-1} min^{-1}$. Subjects were between 18 and 30 years of age, with no known respiratory illnesses.  The

subjects were made familiar with the testing protocol, and were required to sign an informed

consent form before participation in the experiment.  All techniques had been reviewed and

approved by the Clinical Screening Committee for Experiments Involving Human Subjects.

## Protocol

The subjects performed three tests used to indicate differences pre and post exercise.  The

testing consisted of a pre-test to ensure normal spirometry and sufficient fitness, and the

intervention testing including:

• The intervention consisting of a 45-minute bout of maximum sustainable cycling activity

- Pre and post pulmonary diffusion measures

- Pre and post MR

**Pulmonary Function and $\dot{V}O_2max$ test (Pre-test)**

Subjects were asked to avoid exhaustive exercise for 24 hours, caffeine for 12 hours, and food or drink for 2 hours before testing. Height and weight was measured with the subject bare footed and wearing their cycling clothing. Using an automated ventilatory analysis system (Medical Graphics Metabolic Cart, CPX-D), subjects underwent resting pulmonary function testing including forced vital capacity and forced expiratory volume in the first second (FVC, $FEV_1$). This was followed by a 12-second maximum voluntary ventilation (MVV) test.

Subjects were allowed a 5-minute warm-up before performing a maximal exercise test. The test was performed on an electronically braked cycle ergometer (Quinton Excalibur). $\dot{V}O_2max$ was determined using a ramp protocol beginning at 0 watts, increasing 30 watts per minute to volitional fatigue. Expired gases were continuously analyzed by an automated gas analysis system (Rayfield). $O_2$ consumption ($\dot{V}O_2$), minute ventilation ($\dot{V}_E$), production of carbon dioxide ($\dot{V}CO_2$), and respiratory exchange ratio (RER) were measured and recorded every 15 seconds. Before each testing session, the gas analyzers were calibrated with room air and a known gas mixture. $SaO_2$ was measured using an ear oximeter (Ohmeda Biox 3740). The ear lobe was rubbed with a vasodilator gel (Finalgon, Boehringer Igelheim) to improve perfusion. During the test, $SaO_2$ was recorded at 15-second intervals. Ear oximetry has been shown to be a reliable measure of arterial oxyhemoglobin saturation in athletic populations (Mengelkoch et al., 1994). Heart rate was recorded at 15-second intervals using a wireless heart rate monitor (Polar Technologies, Oy) and manually transferred to the spreadsheet.

$\dot{V}O_2$ max was determined by averaging the 3 highest consecutive 15-second values. After the maximal exercise test, subjects were allowed to perform an active cool-down.

Ventilatory threshold was determined by visually inspecting graphs of the 15s interval values for $\dot{V}_E$, $\dot{V}_E/\dot{V}O_2$, $\dot{V}_E/\dot{V}CO_2$, and $\dot{V}CO_2$ versus $\dot{V}O_2$ in accordance with Coutts and McKenzie (1995). The primary criteria used to identify ventilatory threshold was an increase in $\dot{V}_E/\dot{V}O_2$ while $\dot{V}_E/\dot{V}CO_2$ remained relatively constant or decreased (Beaver et al., 1986). Points of steeper increase in $\dot{V}CO_2$ versus $\dot{V}O_2$ and then $\dot{V}_E$ vs. $\dot{V}O_2$ were used, if needed to aid in determining the threshold (Coutts and McKenzie 1995).

## Endurance Cycle Test (Intervention)

Subjects were asked to avoid exhausting exercise for 24 hours, caffeine for 12 hours, and food or drink for 2 hours before testing. The subjects were allowed to warm up for a period of 5 minutes at 50% of their maximum power output in the $\dot{V}O_2$ max test. The resistance was then increased to the predetermined power output over the next 30 seconds. The power output represented 10 % below their predicted ventilatory threshold power. This intensity was adjusted to ensure that the subject maintained a $\dot{V}O_2$ that represents 10 % below their ventilatory threshold throughout the 45-minute test. During the last 3 minutes of the test, the resistance was gradually increased towards the power output achieved at $\dot{V}O_2$ max so that the subject would exercise at that output for approximately 45 seconds. Expired gases were continuously analyzed by an automated gas analysis system (Medical Graphics Metabolic Cart, CPX-D). $\dot{V}O_2$, $\dot{V}_E$, $\dot{V}CO_2$, and RER were measured and recorded every 15 seconds. Heart rate was recorded at 15-second intervals using a wireless heart rate monitor (Polar Vantage, Oy) and manually transferred to the spreadsheet.

## Pulmonary Diffusion Measures

Before the 45-minute cycling task, a measurement of pulmonary diffusion capacity was made. Due to the sensitive nature of the diffusion measurement, the subjects were asked to remain seated for at least 30 minutes before diffusion testing to avoid disruptions to the diffusion measure (Billiet 1971). Diffusing capacity was measured by means of a single breath carbon monoxide diffusing capacity test ($DL_{CO}$, $ml \cdot min^{-1} \cdot torr^{-1}$). Using a test gas containing 21% $O_2$, 10% helium (He), 0.3% CO in a balance of nitrogen ($N_2$), the rate of disappearance of CO from alveolar gas was assessed during a 10-second breath hold. Duplicate trials were performed to ensure that values did not differ by more than 3 $ml \cdot min^{-1} \cdot torr^{-1}$ according to American Thoracic Society standards. Subjects then underwent a 5-minute washout period that consisted of breathing a mixture of 90% $O_2$, balance $N_2$, through a low resistance, non-rebreathing Hans Rudolph valve. This was followed by measuring the rate of disappearance of CO from a second test gas (10% He, 0.3% CO, in a balance of $O_2$) to allow for partitioning of the components of $DL_{CO}$. Again, duplicate trials were performed to ensure accuracy. The $DL_{CO}$ protocol was consistent with that performed by Sheel *et al.* (1996) which found a test-retest correlation for both gas measures of r=0.98 and r=0.96 respectively.

After the cycling intervention was completed, the cyclist was allowed an active cool down for 5 minutes. They then remained seated for 60 minutes at which time the diffusion protocol was repeated.

**Imaging Studies: MR**

The morning of and approximately 90 minutes after the cycling task, the subjects were imaged on a 1.5 Tesla GE Signa MR scanner (General Electric Medical Systems, Milwaukee, 5.4 level). The scanner used a specialized multi-echo pulse sequence with an echo spacing of 10 milliseconds (msec) and a repetition time (TR) of approximately 3000 msec (TR 3000, TE 10, 20, 30 . . . 160 msec). In this single slice 16-echo sequence, the $90°$ radiofrequency (rf) pulse was slice selective and the $180°$ rf pulses were non-selective composite pulses. Multiple gradient lobes (crushers) of alternate sign and incrementally decreasing amplitude were applied in the slice selection directions to eliminate stimulated echo artifacts. This sequence yielded 16 images of a single section per acquisition that provided pixel by pixel data on lung $T_2$ relaxation characteristics of a sagittal, transverse or coronal section. The relatively large number of echoes enabled multi-exponential $T_2$ analysis and accurate extrapolation to TE = 0 for water content. In water phantom studies at the magnet center, this multi-echo pulse sequence yielded a relaxation decay curve identical to that produced by a single slice Hahn echo pulse sequence at a wide range of TE values. Therefore, the multiple $180°$ pulses in the multi-echo sequence do not cause significant signal loss. The rf sensitivity of the body coil was relatively uniform throughout the thorax.

Each subject was imaged in the supine position during quiet breathing at functional residual capacity (FRC) together with a magnesium chloride doped water phantom. Initially, a coronal scout scan was performed. Two cardiac-gated versions of the 16-echo sequence were then obtained in the transverse plane at the level of the lower lobe pulmonary veins. Cardiac gating was initiated 50 msec and 250 msec after the R-wave peak for each of the two scans. The process was then repeated in the sagittal plane at the level of the aortic valve. The images were

obtained using two excitations, a 42-cm field of view, and a 256 x 128 matrix. Respiratory compensation was utilized, but flow compensation was not used.

The 16 images of MR scan data was then transferred to a Sun Sparc 10 workstation and analyzed using the PV Wave Visual Data Analysis software package (Visual Numerics, Inc. CO). For every pixel in the image, the signal intensities from the 16 echoes were fitted to a multi-exponential decay curve. This curve was extrapolated to TE = 0 to obtain the proton density for each pixel. These values were then compared to the water phantom and after correction for the doped water $T_1$ relaxation time, a water density for each pixel was calculated. Large vessels were then removed from the water density image using a water density threshold of $0.3\text{-g}\cdot\text{ml}^{-1}$. The effect of this pixel intensity cutoff was confirmed visually. Following this, the mean density and mass of left and right lungs were calculated for both the pre and post-exercise states allowing for water fraction calculation. All MR protocols and analyses were performed in accordance with validation research of Estilaei *et al.* (1998). Due to reduction in motion artifact, the most artifact free data was acquired with the 15 msec delay (end diastole) in the sagittal projection.

**Statistical Analysis**

The changes in $DL_{CO}$, $D_M$, and $V_C$ from before to after exercise were analyzed using 3 t-tests with alpha set at $\alpha = 0.018$ (0.05/3) to reduce the possibility of a type 2 error. Students t-test was also used to compare the lung water fraction found before and after the exercise trial with alpha set at $\alpha = 0.05$.

# RESULTS

## Descriptive Data

Table 1 gives the anthropometric data for age, height, and weight as well as resting

pulmonary function data. Resting pulmonary function was normal for 7 of the subjects. Subject

6 had a low value for $FEV_1/FVC$ (63%), but was included due to his level of fitness. Individual

data is located in Table 8.

*Table 1. Physical and spirometric characteristics of 8 subjects.*

|       | AGE (years) | HEIGHT (cm) | WEIGHT (kg) | FVC (l) | $FEV_1 \cdot FVC^{-1}$ (%) |
|-------|-------------|-------------|-------------|---------|---------------------------|
| MEAN  | 26.9        | 179.9       | 76.1        | 6.07    | 79.0                      |
| SD    | (3.0)       | (5.7)       | (6.5)       | (1.14)  | (9.2)                     |

## Maximum Cycle Ergometry

Table 2 shows the results from the maximum cycle ergometer test. The subjects in this

study attained $\dot{V}O_2$ max values that were comparable to other diffusion investigations involving

well-trained male subjects (Sheel et al., 1998) (McKenzie et al., 1998). Mean $SaO_2$ decreased to

$92 \pm 1.5\%$, ranging from 95 to 88% at the end of the maximal test. Individual data is found in

Table 9.

*Table 2. Maximal oxygen consumption ($\dot{V}O_2max$), peak power, minimum percent arterial oxygen saturation ($SaO_2min$), and maximum heart rate (HR) during maximal cycle ergometer test.*

|       | $\dot{V}O_2$ max ($l \cdot min^{-1}$) | $\dot{V}O_2$ max ($ml \cdot min^{-1} \cdot kg^{-1}$) | POWER (watts) | $SaO_2min$ (%) | HR (bpm) |
|-------|------------------|----------------------|---------------|----------------|----------|
| MEAN  | 4.84             | 63.7                 | 463           | 92             | 185      |
| SD    | (0.393)          | (2.63)               | (34.8)        | (1.5)          | (8.31)   |

## 45 Minute Cycling Test

Table 3 shows the results of the 45-minute cycle ergometer test. Subjects exercised at a $\dot{V}O_2$ of approximately 1 l·min$^{-1}$ below attained $\dot{V}O_2$ max values and finished at .68 l·min$^{-1}$ below $\dot{V}O_2$ max levels. End power was comparable to that in the $\dot{V}O_2$ max test with subjects attaining a mean power 38 watts below $\dot{V}O_2$ max levels. End heart rate was also comparable with subjects achieving a mean heart rate 4 bpm lower than at the end of the $\dot{V}O_2$ max . Individual data is found in Table 10.

*Table 3. 45-minute cycling test values for ventilatory threshold maximal oxygen consumption (VT $\dot{V}O_2$), ventilatory threshold power (VT POWER), end of test maximal oxygen consumption (END $\dot{V}O_2$), end of test power (END POWER), and end of test heart rate (END HR).*

|  | VT $\dot{V}O_2$ (l·min$^{-1}$) | VT POWER (watts) | END $\dot{V}O_2$ (l·min$^{-1}$) | END POWER (watts) | END HR (bpm) |
|---|---|---|---|---|---|
| MEAN | 3.7 | 300 | 4.0 | 428 | 184 |
| SD | (0.32) | (25.0) | (0.40) | (33.0) | (6.28) |

## Diffusion

Table 4 gives the values for $DL_{CO}$ at the 21% and 90% $O_2$ states, and the partitioned data yielding $D_M$ and $V_C$. $DL_{CO}$ at 21% $O_2$ was significantly decreased post exercise (p = 0.004). $V_C$ was significantly decreased post exercise (p = 0.017), while $D_M$ was not significantly decreased post exercise (p = 0.26). Individual data can be found in Table 11.

*Table 4. Lung diffusion of carbon monoxide breathing 21% oxygen (DL-21%), lung diffusion of carbon monoxide breathing 90% oxygen (DL-90%), diffusion capacity of the alveolar membrane ($D_M$), and alveolar volume ($V_C$).*

| DL-21% (ml·min$^{-1}$·torr$^{-1}$) | DL-90% (ml·min$^{-1}$·torr$^{-1}$) | $D_M$ (ml·min$^{-1}$·torr$^{-1}$) | $V_C$ (ml) |
|---|---|---|---|

|      | Pre    | Post     | Pre    | Post   | Pre     | Post     | Pre     | Post      |
|------|--------|----------|--------|--------|---------|----------|---------|-----------|
| MEAN | 34.39  | 30.15 *  | 13.81  | 11.07  | 106.80  | 117.15   | 67.75   | 53.21 *   |
| SD   | (3.33) | (2.89)   | (2.99) | (2.16) | (26.33) | (42.11)  | (23.03) | (10.21)·  |

*Significantly different from pre (p < 0.018).*

## MRI

Results for extravascular water pre and post exercise are given in table 5. They represent an average of the entire volume of the sagittal slice. There was a significant increase in pulmonary extravascular water after exercise. Individual results are located in table 12.

*Table 5. Pulmonary extravascular water (EW) before (pre) and after (post) a 45-minute bout of cycle ergometry.*

|      | EW ($g \cdot ml^{-1}$) | |
|------|--------|--------|
|      | Pre      | Post     |
| MEAN | 0.223    | 0.244 *  |
| SD   | (0.0225) | (0.0506) |

*Significantly different from pre (p = 0.043).*

Figure 1 compares the change in EW to the change in $D_M$. There was a negative

relationship with a low $R^2$ (0.21) for the linear comparison.

*Figure 1. Change in extravascular water (ΔEW) compared to change in diffusion capacity of the alveolar membrane (ΔD_M).*
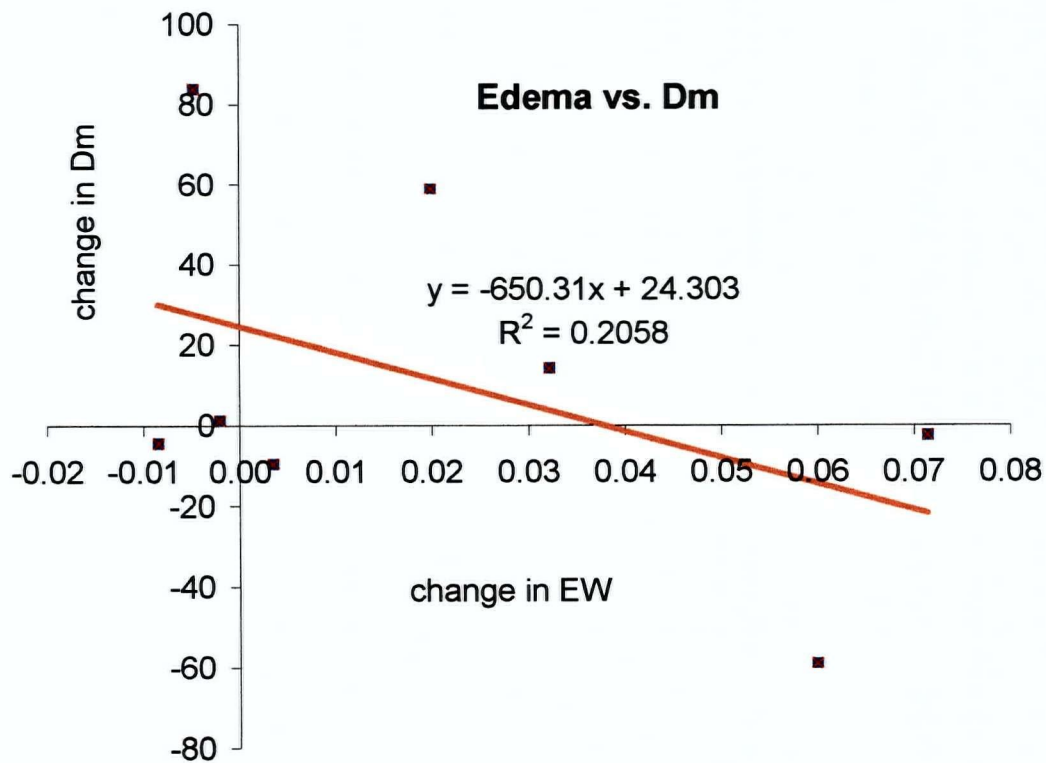
# DISCUSSION

Approximately half of well-trained endurance athletes experience EIH (Powers et al., 1988) and this may negatively effect $\dot{V}O_2$ max (Lawler et al., 1988; Dodd et al., 1989; Martin et al., 1993) and subsequently performance (Koskolou et al., 1994). This has lead to numerous investigations addressing the cause of EIH but at this time, the picture remains unclear. The possible mechanisms include veno-arterial shunts, $\dot{V}_A/\dot{Q}_C$ mismatch, relative alveolar hypoventilation, decreased pulmonary transit time, and pulmonary edema. This study confirmed an increase in EW post exercise in trained male subjects.

## Pulmonary Edema

Previous studies have used a variety of techniques to look for evidence of pulmonary edema during or after exercise in normal humans and animals. The increase in closing capacity of the lung after high levels of exercise has been interpreted as evidence of increased EW (Miles and Durbin 1982), but because these changes are not specific to edema, this can not be considered direct evidence (Gallagher et al., 1988). EW measured by indicator dilution techniques is known to increase during exercise (Marshall et al., 1971; Goresky et al., 1975), but this increase is believed to be due to vascular recruitment rather than to a real increase in EW (Vaughan et al., 1976). The relevance of these studies as to whether athletes develop EW with heavy exercise is unclear. The double-indicator dilution studies did not involve heavy exercise. Since pulmonary vascular pressures increase with exercise (Wagner et al., 1986), and increased pulmonary vascular pressures are implicated in the development of EW (Guyton and Lindsey 1959), it is necessary that subjects are selected who can generate high pulmonary pressures (i.e. athletes) and that exercise of adequate duration and intensity is used.

A study employing radiographic techniques (Gallagher et al., 1988) failed to find increased EW in maximally exercised normal males. Subjects were x-rayed before and 2 minutes following a $\dot{V}O_2$ max test and then the radiographs were analyzed by a radiologist for evidence of EW. The lack of findings may be due to large increases in pulmonary blood flow with exercise masking increases in EW, the short duration of exercise, or the fitness level of the subjects. Also, it has been demonstrated that EW increases need to be greater than 35% to be detected radiographically with certainty (Snashall et al., 1981). Since the athletes in this MR study increased EW by 9.4%, radiographic techniques would lack the sensitivity to detect this change.

Previous studies, using both MR and CT imaging, failed to detect changes in EW following intense short duration exercise. Highly aerobically trained athletes who received CT scans before and after an incremental exhaustive exercise test did not show significant alteration of pulmonary parenchyma despite a drop in $p_aO_2$ and a rise in A-a$DO_2$(Caillaud et al., 1995). Similarly, highly trained cyclists ( $\dot{V}O_2$ max = 4.5 $\pm$ 0.2 l·min$^{-1}$; peak power = 435.4 $\pm$ 19.9 Watts; mean $\pm$ SE) who received MR scans before and after a 5 minute maximal exercise bout (final 3 minutes at 100% $\dot{V}O_2$ max workload), did not show significant increases in EW despite an A-a$DO_2$ of 31.2 $\pm$ 5.0 torr (mean $\pm$ SD) at maximal exercise (McKenzie and Mayo 1996).

The presence of pulmonary edema has been shown to be tightly correlated with increases in lung density (Hedlund et al., 1983). Caillaud *et al.* (1995) used CT to demonstrate an increase of 19% in mean lung density after a triathlon. The subjects were highly trained ( $\dot{V}O_2$ max =4.8 $\pm$ 0.12 l·min$^{-1}$; mean $\pm$ SE), exercised at a high intensity, and for a long duration (120 $\pm$ 20 min; mean $\pm$ SE). In the present study, the subjects were also highly trained ( $\dot{V}O_2$ max = 4.8 $\pm$ 0.12 l·min$^{-1}$; mean $\pm$ SE), exercised at a high intensity (300 $\pm$ 8.8 watts), and for a long duration (45

minutes).  Both the study by Caillaud *et al.* (1995) and the present study found significant changes in EW.  Table 6 compares the data from these four studies, and reveals some interesting similarities.  Both the short-term, maximum intensity experiments resulted in no significant change in EW, and both the long duration, "sustainable maximum" experiments found significant changes in EW.

*Table 6.  A comparison of EW values using various exercise types and EW measures.  $\dot{V}O_2max$ ; maximum oxygen uptake before study, duration; time of exercise protocol, intensity of exercise protocol, type of measure, pre and post protocol results, and result; was there a significant change in EW.*

| Study | Unit | Caillaud *et al.* (1995) | Caillaud *et al.* (1995) | McKenzie *et al.* (1998) | Present study (1998) |
|---|---|---|---|---|---|
| $\dot{V}O_2$ max | ($l \cdot min^{-1}$) | Trained | $4.8 \pm 0.12$ | $4.5 \pm 0.2$ | $4.8 \pm 0.14$ |
| Duration | (min) | 13 | 120 | 5 | 45 |
| Intensity | (watts) | to max | Race | $435 \pm 19.9$ | $300 \pm 8.8$ |
| Measure | (Type) | CT | CT | MR | MR |
| Pre | ($g \cdot ml^{-1}$) | N/A | $0.21 \pm 0.009$ | $0.23 \pm 0.02$ | $0.22 \pm 0.008$ |
| Post | ($g \cdot ml^{-1}$) | N/A | $0.25 \pm 0.01$ | $0.22 \pm 0.01$ | $0.24 \pm 0.018$ |
| Result | ($p < 0.05$) | no | yes | no | yes |

*Values are mean $\pm$ (SE).*

This study showed a reduction in $SaO_2$ to a mean of $92 \pm 0.6\%$ (mean $\pm$ SE) at $\dot{V}O_2max$ , a comparable reduction to the $91 \pm 0.3\%$ found by McKenzie *et al.* (1998).  Since neither of the short duration high intensity studies resulted in a significant increase in EW, it is likely that the changes in EW associated with exercise are dependent on the duration of exercise as well as the

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intensity. Neither $SaO_2$ nor $A\text{-}aDO_2$ was measured during the 45-minute exercise trial, therefore it is not known whether these subjects would have experienced a further increase in $A\text{-}aDO_2$ during the exercise to exhaustion at the end of the trial. It is likely that EW represents only part of the EIH puzzle or that the change in EW with exercise of short duration is not sufficient to be measured with the current level of precision. It is likely that EW is caused by a combination of large increases in $\dot{Q}$, and relative alveolar hypoventilation resulting in low pressures on the alveolar side and higher pressures on the capillary side. Capillary wedge pressures have been shown to exceed 27 torr and pulmonary arterial pressures 40 torr (Wagner et al., 1986; Reeves et al., 1988). It is possible that this would lead to seepage of fluid into the extravascular space due to stress failure of the pulmonary capillaries. This is consistent with the findings of Hopkins, et al (1997) who found erythrocytes upon the completion of heavy exercise, by means of a bronchoalveolar lavage, although it has also been shown that there is no increase in bronchoalveolar lavage fluid erythrocyte content with submaximal exercise (Hopkins et al., 1998). Injury of the vascular endothelium would also allow fluid to penetrate the interstitium of the lung. 90 minutes post exercise it would be expected that pulmonary lymph flow would have cleared the extravascular space, but it is possible that 45-minutes of exercise resulted in an amount of fluid greater than the lymphatic system could clear in the short term.

**Pulmonary Diffusing Capacity**

$DL_{CO}$ has been shown to be reduced following endurance activities (Miles et al., 1983; Manier et al., 1991) and short-term exercise (Rasmussen et al., 1992; Hanel et al., 1994) (Sheel et al., 1998). This study confirms that $DL_{CO}$ is decreased during recovery from endurance activities showing a significant ($p = 0.004$) 12.3% reduction in $DL_{CO}$. The results of partitioning

$DL_{CO}$ into $D_M$ and $V_C$ were consistent with those of previous studies. $V_C$ decreased by 19% ($p = 0.017$), and there was no significant change in $D_M$ ($p = 0.21$). Although there was a mean increase in $D_M$, the values were highly variable with increases found in some subjects and decreases in others.

Diffusion limitations due to pulmonary edema may contribute to the hypoxemia observed in elite athletes at sea level (Younes and Burks 1985; Caillaud et al., 1993; Schaffartzik et al., 1992) and is reflected in small decreases in $D_M$ (Sheel et al., 1998) found post exercise in trained male cyclists. It would be likely that small decreases in $D_M$ would be paired to increases in EW. However, when changes in EW were compared to changes in $D_M$, there was a low negative correlation coefficient of -0.45 (figure 1). This may be due to a difficulty in obtaining a precise measure of $D_M$. This is due to the nature of the $D_M$ calculation. $D_M$ is derived from the y-intercept of two points, as seen in the relationship,

$$\frac{1}{D_L} = \frac{1}{D_M} + \frac{1}{\theta V_C}$$

resulting in an inherently large margin of error. It also may be that the measured edema is concentrated away from the alveolar-arterial tissue barrier. Previous animal studies have shown evidence of perivascular and peribronchial cuffing associated with exercise, which is consistent with that theory (Schaffartzik et al., 1993). In addition, the significant changes in $V_C$ will alter the surface area available for diffusion and $D_M$ will decrease secondary to this hemodynamic shift. Thus $D_M$ and EW are not as intimately related as previously thought.

**MR Sensitivity**

This study was unique in that EW measures were performed using a new technique that analyses the MR data via computer analysis and generates EW values that can be compared, before and after an intervention, and can select a region of interest to compare within the scan. The use of a lab setting also lead to an increased control over environmental factors, such as temperature and altitude, which may effect EW.

The improvement of the MR technique by characterizing $T_2$ distribution as a function of lung water density has lead to greater accuracy in this measure (Estilaei et al., 1998). To validate the MR measurement, porcine lung MR measures were compared with gravimetric analysis yielding a mean difference of -4.1 ± 1.7% (mean ± SE) and an excellent linear relationship of $R^2$ = 0.98 (Estilaei et al., 1998). As there was a small decrease found in the comparison of each sample, it was interpreted as a reflection of minute evaporative loss (Estilaei et al., 1998).

**Summary**

The findings of this study have demonstrated an increase in pulmonary extravascular water following intense, sustained exercise in well-trained male athletes. These data are the first to use MR techniques to document the change in EW, and to compare the change in EW to changes in $D_M$. Although $DL_{CO}$ and $V_C$ were reduced post-exercise, $D_M$ did not change significantly. The level of variability in the $D_M$ measure, the increase in EW not being localized to the gas exchange regions, and the dramatic reduction in $V_C$, are all plausible reasons for the lack of a relationship between EW and $D_M$. Increased pulmonary vascular permeability and high pulmonary capillary pressure are both possible mechanisms of increased EW.

# REFERENCE LIST

1. Asmussen, E, and M Neilsen. 1960. Alveolar-arterial gas exchange at rest and during work at different $O_2$ tensions. *Acta Physiologica Scandinavica* 50: 153-66.

2. Beaver, WL, K Wasserman, and BJ Whipp. 1986. A new method for detecting anaerobic threshold by gas exchange. *Journal of Applied Physiology* 60: 2020-2027.

3. Billiet, L. 1971. Pulmonary diffusing capacity on exercise. *Time course of the change in pulmonary diffusing capacity due to rest or exercise*. Eds MS Bern and H Ruber, 187-94.

4. Buono, MJ, HJ Wilmore, and JR FB Roby. 1982. Evidence of increased thoracic extravascular fluid following intense exercise in man (Abstract). *Physiologist* 25: 201.

5. Caillaud, C, F Anselme, J Mercier, and C Prefaut. 1993. Pulmonary gas exchange and breathing pattern during and after exercise in highly trained athletes. *European Journal of Applied Physiology* 67: 431-37.

6. Caillaud, C, O Serre-Cousine, F Anselme, X capdevilla, and C Prefaut. 1995. Computerized tomography and pulmonary diffusing capacity in highly trained athletes after performing a triathlon. *Journal of Applied Physiology* 79(4): 1226-32.

7. Coutts, KD, and DC McKenzie. 1995. Ventilatory thresholds during wheelchair exercise in individuals with spinal cord injuries. *Paraplegia* 33: 419-22.

8. Dempsey, JA. 1986. Is the lung built for exercise? *Medicine and Science In Sports and Exercise* 18(2): 143-55.

9. Dempsey, JA, PG Hanson, and KS Henderson. 1984. Exercise induced hypoxaemia in healthy human subjects at sea level. *Journal of Physiology* 355: 161-75.

10. Dodd, S L, SK Powers, D Thompson, G Landry, and J Lawler. 1989. Exercise performance following intense, short-term ventilatory work. *International Journal of Sports Medicine* 10(1): 48-52.

11. Ekblom, J, and L Hermansen. 1968. Cardiac output in athletes. *Journal of Applied Physiology* 25: 619-25.

12. Estilaei, M, A MacKay, C Roberts, and J Mayo. 1997. $^1$H NMR measurements of wet/dry ratio and $T_1$, $T_2$ distributions in lung. *Journal of Magnetic Resonance* 124(2): 410-9.

13. Estilaei, M, A MacKay, K Whittall, and J Mayo. 1998. Measurements of water content and $T_2$ relaxation times in lung using a clinical MRI scanner. *Unpublished (Submitted)* .

14. Gale, GE, J Torre-Bueno, RE Moon, HA Saltzman, and PD Wagner. 1985. Ventilatory-perfusion inequality in normal humans during exercise at sea level and simulated altitude. *Journal of Applied Physiology* 58: 978-88.

15. Gallagher, CG, W Huda, M Rigby, D Greenberg, and M Younes. 1988. Lack of radiographic evidence of interstitial pulmonary edema after maximal exercise in normal subjects. *American Review of Respiratory Disease* (137): 474-76.

16. Gledhill, N, A Groese, and J Dempsey. 1977. Ventilation to perfusion distribution during exercise in health. *Muscular exercise and the lung.* Eds Dempsey & Reeds, 325-43. Wisconsin: University of Wisconsin Press.

17. Goresky, CA, JW Warnica, JH Burgess, RFP Cronin, and BE Nadeau. 1972. Changes during exercise in man. *Federation Proceedings* (31): 307-11.

18. Goresky, CA, JW Warnica, JH Burgess, and BE Nadeau. 1975. Effect of exercise on dilution estimates of extravascular lung water and the carbon monoxide diffusing capacity in normal adults. *Circulation Research* 37: 379-89.

19. Guyton, AC, and AW Lindsey. 1959. Effect of elevated left atrial pressure and decreased plasma protein concentration on the development of pulmonary edema. *Circulation Research* 7: 649-57.

20. Hammond, MD, GE Gale, KS Kapitan, A Ries, and PD Wagner. 1986. Pulmonary gas exchange in humans during exercise at sea level. *Journal of Applied Physiology* 60: 1590-98.

21. Hanel, B, PS Clifford, and NH Secher. 1994. Restricted postexercise pulmonary diffusion capacity does not impair maximal transport for $O_2$. *Journal of Applied Physiology* 77(5): 2408-12.

22. Hanel, B, I Teunissen, A Rabol, J Warberg, and N H Secher. 1997. Restricted postexercise pulmonary diffusion capacity and central blood volume depletion. *Journal of Applied Physiology* 83(1): 11-7.

23. Hastings, AB, FC White, TM Sanders, and CM Bloor. 1982. comparative physiological responses to exercise stress. *Journal of Applied Physiology* 52: 1077-83.

24. Hayes, C, T Case, and D Ailion. 1982. Lung water quantitation by nuclear magnetic resonance imaging. *Science* 216: 1313-15.

25. Hedlund, LW, P Vock, and EL Effmann. 1983. Computed tomography of the lung. *Radiologic Clinics of North America* 21: 775-88.

26. Hopkins, SR, AS Belzberg, BR Wiggs, and DC McKenzie. 1996. Pulmonary transit time and diffusion limitation during heavy exercise in athletes. *Respiration Physiology* 103(1): 67-73.

27. Hopkins, SR, and DC McKenzie. 1989. Hypoxic ventilatory response and arterial desaturation during heavy work. *Journal of Applied Physiology* 67(3): 1119-24.

28. Hopkins, SR, DC McKenzie, RB Schoene, RW Glenny, and HT Robertson. 1994. Pulmonary gas exchange during exercise in athletes I: Ventilation - perfusion mismatch and diffusion limitation. *Journal of Applied Physiology* 77(2): 912-7 .

29. Hopkins, SR, RB Schoene, WR Henderson, RG Spragg, TR Martin, and JB West. 1997. Intense exercise impairs the integrity of the pulmonary blood-gas barrier in elite athletes. *American Journal of Respiratory and Critical Care Medicine* 155(3): 1090-4.

30. Hopkins, SR, RB Schoene, WR Henderson, RG Spragg, and JB West. 1998. Sustained submaximal exercise does not alter the integrity of the lung blood-gas barrier in elite athletes. *Journal of Applied Physiology* 84(4): 1185-9.

31. Koskolou, MD, and DC McKenzie. 1994. Arterial Hypoxemia and Performance During Intense Exercise. *European Journal of Applied Physiology* 68: 80-86.

32. Lawler, J, SK Powers, and D Thompson. 1988. Linear relationship between VO2max and VO2max decrement during exposure to acute hypoxia. *Journal of Applied Physiology* 64(4): 1486-92.

33. Manier, G, J Moinard, and H Stoicheff. 1993. Pulmonary diffusing capacity after maximal exercise. *Journal of Applied Physiology* 75(6): 2580-2585.

34. Manier, G, J Moinard, P Techoueyres, N Varene, and H Huenard. 1991. Pulmonary diffusion limitation after prolonged strenuous exercise. *Respiration Physiology* 83: 143-54.

35. Markisz, MJ, and Aquilia. 1996. Technical magnetic resonance imaging. Stanford, Conn.: Appleton & Lange.

36. Marshall, BE, LR Soma, and GR Neufeld. 1975. Lung water volume at rest and exercise in dogs. *Journal of Applied Physiology* 39: 7-8.

37. Marshall, BE, RL Teichner, T Kallos, HJ Sugerman, MQ Wyche, and KR Tantum. 1971. Effects of posture and exercise on the pulmonary extravascular water volume in man. *Journal of Applied Physiology* 31: 375-79.

38. Martin, D, M Cicale, D Huang, L Menglecoch, S Powers, and D Criswell. 1992. Incidence of Exercise-Induced Hypoxemia in Male Endurance Athletes (Abstract). *Medicine and Science in Sport and Exercise* 24: s69.

39. Martin, D, and J O'Kroy. 1993. Effects of acute hypoxia on the VO$_2$max of trained and untrained subjects. *Journal of Sports Sciences* 11: 37-42.

40. Mayo, J, A MacKay, K Whittall, E Baile, and P Pare. 1995. Measurement of lung water

content and pleural pressure gradient with magnetic resonance imaging. *Journal of Thoracic Imaging* 16: 73-81.

41.  Mayo, J, N Muller, B Forster, M Okazawa, and P Pare. 1990. Magnetic resonance imaging of hydrostatic pulmonary edema in isolated dog lungs: comparison with computed tomography. *Canadian Association of Radiologists Journal* 6: 281-86.

42.  McKechnie, JK, WP Leary, TD Noakes, JC Kallmeyer, ETM MacSearraigh, and LR Oliver. 1979. Acute pulmonary edema in two athletes during a 90km running race. *South African Medical Journal* 56: 261-65.

43.  McKenzie, DC, IL Lama, JE Potts, AW Sheel, and KD Coutts. 1998. The effects of repeat exercise on pulmonary diffusing capacity and exercise-induced hypoxemia in highly trained athletes. *Medicine and Science In Sports and Exercise* .

44.  McKenzie, DC, and J Mayo. 1996. The effects of Short term maximal exercise on the development of interstitial edema. *Physiologist* .

45.  Mengelkoch, LJ, D Martin, and J Lawler. 1994. A review of the principles of pulse oximetry and accuracy of pulse oximeter estimates during exercise. *Physical Therapy* 74(1): 40-9.

46.  Miles, DS, CE Doerr, SA Schonfeld, DE Sinks, and RW Gotshall. 1983. Changes in pulmonary diffusing capacity and closing volume after running a marathon. *Respiratory Physiology* 52: 349-59.

47.  Miles, DS, and RJ Durbin. 1982. Alterations in small airways and diffusing capacity after a 5-mile run. *Physiologist* 25: 199.

48.  Morris, A, D Blatter, and T Case. 1985. A new nuclear magnetic resonance property of lung. *Journal of Applied Physiology* 58: 759-62.

49.  Perrault, H, R Turcotte, L Kiteala, and JE Marcotte. 1991. Oxyhemoglobin saturation and lung diffusion capacity during near-maximal exercise in endurance athletes. *Medicine and Science In Sports and Exercise* 23(4): s578.

50.  Philips, P, P Allen, and S Man. 1989. Assessment of temporal changes in pulmonary edema with NMR imaging. *Journal of Applied Physiology* 66: 1197-208.

51.  Powers, SK, S Dodd, J Lawler, G Landry, M Kirtley, T McKnight, and S Grinton. 1988. Incidence of exercise-induced hypoxemia in elite endurance athletes at sea level. *European Journal of Applied Physiology* 58: 298-302.

52.  Powers, SK, S Grinton, J Lawler, D Criswell, and S Dodd. 1992. High intensity exercise training-induced metabolic alterations in respiratory muscles. *Respiration Physiology* 89(2): 169-77.

53.  Powers, SK, D Martin, and S Dodd. 1993. Exercise-induced hypoxaemia in elite

endurance athletes: Incidence, causes and impact on VO₂max. *Sports Medicine* 16(1): 14-22.

54. Powers, SK, D Martin, and S. Dodd. 1993. Exercise-induced hypoxemia in elite endurance athletes. *Sports Medicine* 16(1): 14-22.

55. Rasmussen, BD, P Elkjaer, and B Juhl. 1988. Impaired pulmonary and cardiac function after maximal exercise. *Journal of Sports Sciences* 6: 219-28.

56. Rasmussen, J, B Hanel, K Saunamaki, and NH Secher. 1992. Recovery of pulmonary diffusing capacity after maximal exercise. *Journal of Sports Sciences* 10(6): 525-31.

57. Reeves, JT, RE Moon, RF Grover, and BM Groves. 1988. Increased wedge pressure facilitates decreased lung vascular resistance during upright exercise. *Chest* 91(1): S97-99.

58. Roughton, FJW, and RE Forster. 1957. Relative importance of diffusion and chemical reaction rates in determining rate of exchange of gases in the human lung, with special reference to true diffusing capacity of pulmonary membrane and volume of blood in the lung capillaries. *Journal of Applied Physiology* 11(2): 290-302.

59. Saltin, B, B Blomqvist, JH Mitchell, RL Johnson Jr., K Wildenthal, and CB Chapman. 1968. Responses to exercise after bed rest and after training: A longitudinal study of adaptive changes in oxygen transport and body composition. *Circulation* 38[Suppl.] (7): 1-8.

60. Schaffartzik, W, JP Arcos, K Tsukimoto, O Mathieu-Costello, and PD Wagner. 1993. Pulmonary interstitial edema in the pig after heavy exercise. *Journal of Applied Physiology* 75(6): 2535-40.

61. Schaffartzik, W, DC Poole, T Derion, K Tsukimoto, MC Hogan, JP Arcos, D Bebout, and PD Wagner. 1992. VA/Q distribution during heavy exercise and recovery in humans: implications for pulmonary edema. *Journal of Applied Physiology* 72(5): 1657-67.

62. Schumacker, PT, B Guth, AJ Suggett, PD Wagner, and JB West. 1985. Effects of transfusion-induced polycythemia on O₂ transport during exercise in the dog. *Journal of Applied Physiology* 58: 749-58.

63. Sheel, AW, KD Coutts, JE Potts, and DC McKenzie. 1998. The time course of pulmonary diffusing capacity for carbon monoxide following short duration high intensity exercise. *Respiration Physiology* (in press).

64. Snashall, PD, SJ Keynes, and BM Morgan. 1981. The radiographic detection of acute pulmonary edema: a comparison of radiographic appearances, densitometry and lung water in dogs. *British Journal of Radiology* 54: 277-88.

65. Torre-Bueno, JR, PD Wagner, HA Saltzman, GE Gale, and RE Moon. 1985. Diffusion limitation in normal humans during exercise at sea level and simulated altitude. *Journal of Applied Physiology* 58(3): 989-95.

66. Vaughan, TR, EM DeMarino, and NC Staub. 1976. Indicator dilution lung water and capillary blood volume in prolonged heavy exercise in normal men. *American Review of Respiratory Disease* 113: 757-62.

67. Wagner, PD. 1992. Ventilation-perfusion matching during exercise. *Chest* 101: S192-98.

68. Wagner, PD, GE Gale, RE Moon, JR Tore-Bueno, BW Stolp, and HA Saltzman. 1986. Pulmonary gas exchange in humans exercising at sea level and simulated altitude. *Journal of Applied Physiology* 61: 260-270.

69. Wagner, PD, JR Gillespie, GL Landgren, MR Fedde, BW Jones, RM DeBowes, RL Pieschl, and JJ Erickson. 1989. Mechanisms of exercise-induced hypoxemia in horses. *Journal of Applied Physiology* 66(3): 1227-33.

70. West, JB. 1977. Pulmonary Pathophysiology. 112-19. Baltimore: Waverly Press.

71. West, JB. 1985. *Ventilation/blood flow and gas exchange.* 12-14. London: Blackwell Scientific Publications.

72. Whipp, B, and K Wasserman. 1969. Alveolar-arterial gas tension difference during graded exercise. *Journal of Applied Physiology* 27: 361-65.

73. Younes, M, and J Burks. 1985. Breathing pattern during and after exercise of different intensities. *Journal of Applied Physiology* 59(3): 898-908.

# Appendix A: Literature Review of EIH

## The Limit to Aerobic Performance

Many factors have been attributed to the limit to aerobic performance. If the transport of $O_2$ to metabolically active tissues, such as muscle, is limited, it will have adverse effects on the performance of that muscle. When sustained heavy exercise is performed, large volumes of $O_2$ are required for aerobic performance. Well trained males, can achieve $O_2$ consumption rates in excess of 5 $l \cdot min^{-1}$, and yet their performance is improved when the delivery of $O_2$ is increased artificially. We know that performance is improved when the $O_2$ partial pressure rises, and impaired when $O_2$ partial pressure is lowered. At sea level, normal pressure is 760 torr and $O_2$ comprises 20.93% of that air. This results in a partial pressure of $O_2$ of 159 torr. When air passes into the lungs 47 torr is lost to vapor pressure and contains 149 torr $O_2$. As air moves into the lungs, there is a mixing of inspired and expired air, resulting in a progressive decrease in the partial pressure of $O_2$ due to the presence of $CO_2$ and the transfer of $O_2$ into the capillaries. At the alveoli, the mean $O_2$ pressure is 104 torr. As blood passes through the capillaries, it becomes almost completely equilibrated with the alveolar air, resulting in an arterial $O_2$ pressure of 100 torr. Gas equilibration at the tissue capillaries results in an extracellular and venous $pO_2$ of 40 torr at rest. If there is a reduction in $pO_2$ at any step before delivery to aerobically active tissue, it will lead to a reduction in $pO_2$ delivery, and reduced aerobic potential of the tissues.

## Pulmonary Limitations to the $O_2$ Cascade

$O_2$ transport between alveolus and capillary is not normally considered to be a limitation to aerobic capacity (Asmussen et al., 1960; Saltin et al., 1968). Changes associated with training manifest in cardiovascular and musculoskeletal system adaptations that can be easily measured.

The ground breaking research of Dempsey *et al.* (1984) was the first to demonstrate a reduction in $SaO_2$ to levels below 90% associated with a reduction in arterial $O_2$ ($p_aO_2$). This was measured through a comparison of alveolar and arterial $O_2$ pressures, resulting in the hallmark EIH measure of increased A-a$DO_2$. This discovery displayed the abilities of the pulmonary system being exceeded by very fit individuals. This suggests the pulmonary system has failed to adapt adequately in parallel with the adaptations of the other systems. The presence of EIH in fit athletes has been corroborated by other studies (Hopkins et al., 1989) and is estimated to effect 50% of highly trained male endurance athletes (Powers et al., 1988; Powers et al., 1993; Martin et al., 1992). The presence of EIH in fit athletes at maximum levels of exercise has been shown to be a limit to the attainment of $\dot{V}O_2$ max (Lawler et al., 1988; Dodd et al., 1989; Martin et al., 1993) and maximum performance (Koskolou et al., 1994). This has made research into the causes of EIH an important contributor to the understanding of the limits to elite performance.

**Exercise Induced Hypoxemia**

Inadequate equilibration of arterial to alveolar gas results in a widening of the A-a$DO_2$ at intense levels of exercise (Dempsey et al., 1984; Hammond et al., 1986; Wagner et al., 1986; Asmussen et al., 1960) and is increased further by hypoxia (Torre-Bueno et al., 1985). The increase in A-a$DO_2$, accompanied by hypoxemia in well-trained athletes (Dempsey et al., 1984), suggests a decreased ability to diffuse gas across the pulmonary membrane. An increase in A-a$DO_2$ associated with exercise can be explained by $\dot{V}_A/\dot{Q}_c$ inequality (Wagner et al., 1986), relative alveolar hypoventilation (Dempsey et al., 1984), right to left shunts (Wagner et al., 1986), and diffusion limitation (Wagner et al., 1986).

**Relative Alveolar Hypoventilation**

Some athletes with high aerobic capacities, may have an inadequate ventilatory response to maximal exercise (Dempsey et al., 1984; Wagner 1992) causing a decreased $p_AO_2$. This lowers the driving force of $O_2$ across the pulmonary membrane and consequently reduces the gradient for $O_2$ transfer to the mitochondria. It is possible that, in an attempt to conserve energy, the body will rely on less ventilation than normal and consequently a reduced $pO_2$ supplied to the alveoli. As has been suggested by Dempsey *et al.* (1984, 1986), this may lead to EIH. Although a lack of compensatory hyperventilation has been noted in some athletes (Dempsey et al., 1984; Perrault et al., 1991), a study measuring $p_AO_2$ directly did not find a reduction despite a marked reduction in $p_aO_2$ to about 78 torr (Hopkins et al., 1989). Corroborating this evidence, Powers *et al.* (1992) found a reduced $p_aCO_2$ during maximal exercise in subjects who developed EIH. This has been contested by Caillaud *et al.* (1993) who noted relative reductions in $p_AO_2$ and increased arterial carbon dioxide pressure ($p_aCO_2$) when highly trained subjects were compared to untrained subjects during maximal exercise.

A reduced response to stimuli known to increase ventilation, such as increases in body temperature, acidosis, blood catecholamines, and $P_aCO_2$ as well as decreased $p_aO_2$ may play a part in hypoventilation seen in some athletes. In 1993, Cooper showed a reduced peripheral chemoresponsiveness to hypercapnia in athletes exhibiting EIH, corroborating this theory. The understanding of hypoventilation's contribution to EIH has not been fully elucidated by the current literature, and requires further study. This study has shown that edema may be related to submaximal exercise, and although this study did not demonstrate EIH, it may be that the effects of submaximal hypoventilation contribute to the generation of EIH in some athletes. The

consequence of submaximal hypoventilation would be a reduction in respiratory muscle fitness, as well as increased susceptibility to pulmonary edema.

**Pulmonary Ventilation and Perfusion Inequality**

$\dot{V}_A/\dot{Q}_C$ inequality may also contribute to EIH, with matching of ventilation to perfusion becoming less uniform as exercise intensity increases (Gale et al., 1985; Hammond et al., 1986; Hopkins et al., 1994). $\dot{V}_A/\dot{Q}_C$ inequality occurs due to a combination of two factors. $\dot{V}_A$ is affected by the mixing of inspired gas with lung air that is part of the dead space volume. The gas at the base of the lungs has a greater concentration of dead space air and consequently contains a lower $pO_2$. The apex of the lungs has a lower dead space concentration and therefore has a higher $pO_2$. This leads to a higher $pO_2$ found apically compared to basal pressures. $\dot{Q}_C$ has an opposite pattern, due to gravity, allowing for an increased perfusion gradient towards the base of the lungs. This results in basal pulmonary capillaries being perfused better than those found apically. The low $pO_2$ and high $\dot{Q}_C$ found at the base of the lung, and conversely high $pO_2$ and low $\dot{Q}_C$ apically, creates a scenario whereby the inadequacies of this $\dot{V}_A/\dot{Q}_C$ matching are made apparent with increasing demands for $O_2$ supply.

$\dot{V}_A/\dot{Q}_C$ inequality has been shown to explain 1/3 of the widened A-aDO$_2$ associated with exercise at sea level (Wagner et al., 1986). Further, Hammond *et al.* (1986) showed $\dot{V}_A/\dot{Q}_C$ inequality increasing with exercise intensity, up to a $\dot{V}O_2$ of approximately 3.5 l·min$^{-1}$. When $\dot{V}O_2$ increased beyond 3.5 l·min$^{-1}$, there was no further increase in $\dot{V}_A/\dot{Q}_C$ inequality, but a continued widening of A-aDO$_2$. Hopkins *et al.* (1994) assessed $\dot{V}_A/\dot{Q}_C$ inequality in elite

athletes and found that in well trained subjects the contribution of $\dot{V}_A/\dot{Q}_c$ inequality to A-aDO$_2$ was approximately 60% during heavy exercise.

The reason for the increase in $\dot{V}_A/\dot{Q}_c$ mismatch associated with exercise remains unclear, although Schaffartzik *et al.* (1993) suggested reduced gas mixing in the large airways, a lack of uniformity in pulmonary vasoconstriction, and the onset of pulmonary edema. In 1992, Schaffartzik *et al.* found that $\dot{V}_A/\dot{Q}_c$ mismatch persisted for 20 minutes in half of the subjects following a near maximal exercise bout breathing hypoxic gas. Ventilation and cardiac output returned toward baseline values more rapidly than did $\dot{V}_A/\dot{Q}_c$ relationships. This is consistent with the hypothesis that edema occurs and contributes to $\dot{V}_A/\dot{Q}_c$ mismatch during exercise. $\dot{V}_A/\dot{Q}_c$ appears to contribute to EIH to some extent, but it is likely that some of the inhomogeneity reported occurs due to pulmonary edema (which may also contribute to EIH).


**Veno-Arterial Shunts**

Veno-arterial (v-a) shunts occur due to anatomical and physiological factors. Anatomical shunting occurs when red blood cells are not exposed to alveoli. Physiological or functional shunting occurs when red blood cells are exposed to alveoli that are incapable of transferring O$_2$. V-a shunts have been shown to account for approximately 50 percent of the A-aDO$_2$ at rest (Whipp and Wasserman 1969; Gledhill et al., 1977). It was later shown that breathing hyperoxic gas during maximal exercise caused $p_aO_2$ to return to normal (Dempsey et al., 1984; Powers et al., 1992), suggesting that v-a shunts do not account for any of the A-aDO$_2$ found with EIH. It is not likely that v-a shunts have a significant impact on overall gas exchange (Wagner et al., 1986).

## Diffusion Limitation

A significant contribution to A-aDO$_2$ at sea level appears to be from diffusion limitation (Wagner et al., 1986). The rate of O$_2$ lung diffusion is controlled by five factors including: the concentration gradient from alveoli to capillaries, D$_M$, V$_C$, the rate of combination with hemoglobin ($\theta$), and capillary transit time. The concentration gradient depends on the level of alveolar ventilation, which in turn is dependent on breathing rate and depth, and has been covered in the section on relative alveolar hypoventilation. Capillary transit time is dependent on $\dot{Q}c$ and V$_C$, and will be discussed in the following section. D$_L$ summarizes changes in D$_M$, V$_C$, and $\theta$. This will be covered following capillary transit time.

## Diffusion Limitation – *Pulmonary Capillary Transit Time*

The capillary transit time is measured as the time of exposure of the red blood cell to the alveolus before becoming part of the pulmonary venous circulation. This may occur due to the concommitant expansion of V$_C$ with increasing $\dot{Q}c$, reaching its anatomical limit while $\dot{Q}c$ continues to increase (Dempsey et al., 1984; Hopkins et al., 1994). V$_C$ is estimated to triple with exercise, and is intended to compensate for increasing $\dot{Q}c$ to maintain adequate capillary transit time. It appears that this increase is inadequate. During maximum exercise, $\dot{Q}$ increases to $\sim 33$ l·min$^{-1}$ (Hopkins et al., 1994) and may reach 40 l·min$^{-1}$ (Ekblom et al., 1968). This causes transit time to be reduced to approximately 0.25 seconds in the pulmonary capillary in well trained individuals(West 1985). Hopkins *et al.* (1996) showed that there is evidence suggesting a relationship between D$_L$ limitation and decreased capillary transit times. This rapid rate of mean flow combined with the fact that during systole $\dot{Q}$ will be even higher than mean values, resulting in even shorter transit times for RBC, may account for some of the A-aDO$_2$ widening.

## Diffusing Capacity

Through techniques developed by Roughton $et$ $al.$ (1957), $D_L$ may be partitioned into 2 components: $D_M$ and $V_C$. This may be used to further display the cause for post exercise $D_L$ limitation. Sheel $et$ $al.$ (1998, in press) showed that much of the change in $D_L$ post exercise was attributable to decreases in $V_C$ and that these effects remained 6 hours post exercise. It is likely that there is a plasma ANP regulated blood volume diversion to the recovering musculature (Hanel et al., 1997). The values found for $D_M$ are highly variable, and have not shown a significant decrease in some studies (Miles et al., 1983; Hanel et al., 1994), but have found decreases in others (Sheel et al., 1998). It is likely that Sheel $et$ $al.$ found a difference where others failed due to his large (n = 20) sample size and therefore greater power.

A reduction in $D_L$ following exercise has been reported by numerous authors (Miles et al., 1983; Rasmussen et al., 1988; Manier et al., 1993; Hanel et al., 1994) (Sheel et al., 1998) (McKenzie et al., 1998). $D_L$ decrease may be attributable to three mechanisms. They include an incomplete equilibration of $p_aO_2$ with $p_AO_2$ due to a short red blood cell exposure in the pulmonary capillaries, a reduction in $V_C$ due to shunting of pulmonary blood to working muscles, and pulmonary edema.

## Diffusion Limitation – *Pulmonary Edema*

The accumulation of EW, or pulmonary edema is the second possible mechanism. EW would affect diffusion by increasing the tissue thickness that provides the barrier between alveolar and capillary spaces. The possible mechanism for the accumulation of EW includes increased capillary permeability, increased capillary surface area, increased capillary hydrostatic

pressure, or a lymphatic insufficiency (West 1977). The presence of EW in athletes has been

suggested by a continued $\dot{V}_A/\dot{Q}_C$ mismatch found during 20 minutes of recovery from hypoxic

exercise (Schaffartzik et al., 1992). Previous studies have found acute clinical pulmonary edema

in humans caused by heavy exercise at sea level (McKechnie et al., 1979) and moderate exercise

at high altitude (Marshall et al., 1971). It has also been suggested that brief intense exercise in

athletes with a history suggestive of lung bleeding alters blood-gas barrier function resulting in

higher concentrations of red cells and protein in broncho-alveolar lavage fluid (Hopkins et al.,

1997). When broncho-alveolar lavage was performed on athletes who had performed sustained

submaximal exercise (one hour at 77% of $\dot{V}O_2$ max ), erythrocyte content was not different from

controls (Hopkins et al., 1998). This suggests that extreme levels of exercise are required to alter

the blood-gas barrier integrity (Hopkins et al., 1998). There have also been studies using double

indicator-dilution techniques (Marshall et al., 1971; Goresky et al., 1972) indicating increases

EW during exercise as well as the use of transthoracic electrical impedance during a 30 minute

recovery that suggested elevated intrathoracic fluid volumes (Buono et al., 1982).

Some studies may have failed to detect increases in EW (Marshall et al., 1975; Gallagher

et al., 1988) due to the small fluid volume or the sensitivity of the measurement techniques

(Schaffartzik et al., 1993). Also, in some athletes, there is no increase in $\dot{V}_A/\dot{Q}_C$ inequality

during exercise, and pulmonary edema may not occur in such individuals (Schaffartzik et al.,

1992). It is also possible that previous methods lack the sensitivity to detect the small

differences, as would appear to be the case in the indicator-dilution and X-ray techniques

(Gallagher et al., 1988).

Experiments with $DL_{CO}$ yield indirect evidence of a diffusion limitation relating to the

pulmonary membrane (Manier et al., 1993) (Sheel et al., 1998). The presence of pulmonary

edema has been shown to be tightly correlated with increases in lung density (Hedlund et al., 1983). A recent study used CT to measure changes in EW, and corroborated the presence of EW by the visual inspection of the images for linear and polygonal opacities as well as computing lung density (Caillaud et al., 1995). The subjects were highly trained ($\dot{V}O_2$ max $=4.8 \pm 0.12$ l·min$^{-1}$; mean $\pm$ SE), exercised at a high intensity, and for a long duration ($120 \pm 20$ min; mean $\pm$ SE). The current study also used highly trained ($\dot{V}O_2$ max $= 4.8 \pm 0.12$ l·min$^{-1}$; mean $\pm$ SE), subjects who exercised at a high intensity ($300 \pm 8.8$ watts), and for a long duration (45 minutes). Both the study by Caillaud *et al.* (1995) and the present study found significant changes in EW. Two other studies also using highly trained athletes, but exercising for a short duration at maximum intensity, failed to find changes in EW (Caillaud et al., 1995) (McKenzie et al., 1996). Table A1 compares the data from these four studies, and reveals some interesting similarities. Both short-term, maximum intensity experiments resulted in a lack of significant increases in EW, and both long duration, "sustainable maximum" experiments found significant increases in EW.

*Table A1. A comparison of EW values using various exercise types and EW measures. VO2max ; maximum oxygen uptake before study, duration; time of exercise protocol, intensity of exercise protocol, type of measure, pre and post protocol results, and result; was there a significant change in EW.*

| Study | Unit | Caillaud *et al.* (1995) | Caillaud *et al.* (1995) | McKenzie *et al.* (1998) | Present study (1998) |
|---|---|---|---|---|---|
| $\dot{V}O_2$ max | ($l \cdot min^{-1}$) | Trained | $4.8 \pm 0.12$ | $4.5 \pm 0.2$ | $4.8 \pm 0.14$ |
| Duration | (min) | 13 | 120 | 5 | 45 |
| Intensity | (watts) | to max | Race | $435 \pm 19.9$ | $300 \pm 8.8$ |
| Measure | (Type) | CT | CT | MR | MR |
| Pre | ($g \cdot ml^{-1}$) | N/A | $0.21 \pm 0.009$ | $0.23 \pm 0.02$ | $0.22 \pm 0.008$ |
| Post | ($g \cdot ml^{-1}$) | N/A | $0.25 \pm 0.01$ | $0.22 \pm 0.01$ | $0.24 \pm 0.018$ |
| Result | ($p < 0.05$) | no | yes | No | yes |

*Values are mean ± (SE).*

There was a reduction in $SaO_2$ to a mean of $92 \pm 0.6\%$ (mean ± SE) at $\dot{V}O_2$ max in the current study, and a comparable reduction to $91 \pm 0.3\%$ found by McKenzie *et al.* (1998). Since neither of the short-duration, high intensity studies resulted in a significant increase in EW, it is likely that the changes in EW associated with exercise are the result of the duration of exercise as well as the intensity. Neither $SaO_2$ nor $A\text{-}aDO_2$ was measured during the 45-minute exercise trial, therefore it is not known whether these subjects would have experienced a further increase in $A\text{-}aDO_2$ during the exercise to exhaustion at the end of the trial. It is likely that EW represents only part of the EIH puzzle or that the change in EW with exercise of short duration is not sufficient to be measured with the current level of precision. It is likely that EW is caused by a combination of large increases in $\dot{Q}$, and relative alveolar hypoventilation resulting in low

pressures on the alveolar side and higher pressures on the capillary side. Capillary wedge pressures have been shown to exceed 27 torr and pulmonary arterial pressures 40 torr (Wagner et al., 1986; Reeves et al., 1988). It is possible that this would lead to seepage of fluid into the extravascular space due to stress failure of the pulmonary capillaries. This is consistent with the findings of Hopkins, et al (1997) who found erythrocytes upon the completion of heavy exercise, by means of a bronchoalveolar lavage, although it has also been shown that there is no increase in bronchoalveolar lavage fluid erythrocyte content with submaximal exercise (Hopkins et al., 1998). Injury of the vascular endothelium would also allow fluid to penetrate the interstitium of the lung. 90 minutes post exercise it would be expected that pulmonary lymph flow would have cleared the extravascular space, but it is possible that 45 minutes of exercise resulted in an amount of fluid greater than the lymphatic system could clear in the short term.

Current technical advances in the use of MR to quantify data have resulted in a more sensitive method of detecting changes in EW (Estilaei et al., 1997). The new MR technique has some benefits over the CT method. One obvious benefit is the lack of exposure to ionizing radiation. A second involves the analysis itself, and the ability of the MR analysis to analyze a region of interest within the scan, and compare that area to other regions.

**Pulmonary Edema – Animal Studies**

Special attention needs to be taken when using animal models due to species differences in the regulation of pulmonary hemodynamics and fluid filtration. Measurements of wet-to-dry lung weight ratio (W/D ratio) in 4 dogs after exercise compared the results with the ratio obtained in non-exercised dogs did not find a significant change in EW (Marshall et al., 1975). With dogs there is no increase in the A-aDO$_2$ at high exercise intensities (Schumacker et al.,

1985) and the study examining W/D ratios did not involve heavy exercise making the presence of pulmonary edema unlikely. Horses experience an increase in A-aDO$_2$ with increasing exercise intensities, but do not appear to experience an increase in $\dot{V}_A/\dot{Q}_c$ mismatch associated with that widening (Wagner et al., 1989). Pigs develop a considerable amount of hypoxemia with exercise (Hastings et al., 1982) possibly in part due to increasing $\dot{V}_A/\dot{Q}_c$ mismatch (Schaffartzik et al., 1993), and experience perivascular cuffing, suggestive of pulmonary edema (Schaffartzik et al., 1993) making them a good model for human comparison.

# Appendix B: Magnetic Resonance Imaging

MR is a diagnostic tool capable of rendering a picture of a scanned area based on the characteristics of its mobile hydrogen atoms. It is also non-invasive, and lacks the ionizing radiation involved with other imaging techniques (Hayes et al., 1982) (Morris et al., 1985) (Mayo et al., 1995). The tissue to be scanned is placed within a superconducting electromagnet. The electromagnet emits a magnetic field powerful enough to cause all the mobile hydrogen atoms to align with the external magnetic field. Coils then emit an rf pulse that causes the protons to be spun a certain number of degrees. (often 90° or 180°) The coils are then able to measure two quantities:

1. The longitudinal relaxation ($T_1$) or time for 63% of the nuclei to realign with the magnet,

2. and the transverse relaxation ($T_2$) or time for 63% of the nuclei's precession rates to be out of phase.

The MR unit also consists of gradient coils. The gradient coils emit a sloped field that allows areas within the magnet to be differentiated from one another.

There are many variables within the control of the experimenter when using MR. These parameters will often have positive and negative results of their values; they should therefore be adjusted based on the desired output. For example: a long relaxation time (TR) will result in better resolution, but also increases the scanning time thereby increasing the cost, and the possibility of patient movement. Table B1 summarizes some of the parameters that are adjustable in an MR scan. An increase in the TR, an increase in the number of signal averages ($N_{ex}$), or a narrow bandwidth will all result in an increased signal to noise ratio (SNR). However, an increase in TR or in the $N_{ex}$ increases the scanning time. As increases are made in the voxel size (decreasing matrix size), there is increased SNR and decreased scanning time, but

a relatively large decrease in spatial resolution. Increases in the field of view and the slice

thickness have no effect on scanning time, but will cause an increase in the SNR and a decrease

in the resolution.

*Table B1. Scanning parameter effect on image acquisition*

| Parameter | Value | SNR | Resolution | Scanning Time |
|---|---|---|---|---|
| TR | High | ↑ | ↑ | ↑ |
|  | Low | ↓ | ↓ | ↓ |
| $N_{ex}$ | High | ↑ | ↑ | ↑ |
|  | Low | ↓ | ↓ | ↓ |
| Matrix | High | ↓ | ↑↑ | ↑ |
|  | Low | ↑ | ↓↓ | ↓ |
| FOV | Large | ↑ | ↓ | — |
|  | Small | ↓ | ↑ | — |
| Slice thickness | Thick | ↑ | ↓ | — |
|  | Thin | ↓ | ↑ | — |
| Slice spacing | Narrow | ↓ | ↓ | — |
|  | Wide | ↑ | ↑ | — |
| Bandwidth | Narrow | ↑ | ↑ | — |
|  | Wide | ↓ | ↓ | — |

*From Markisz et al. (1996)*

The type of pulse sequence used in the MR scan is also important to the type of MR

appearance that is desired. The pulse sequence is the combination of rf pulses designed to

produce an image. There are many different techniques including: spin-echo, gradient-echo, fast

spin-echo, and inversion-recovery techniques. Spin-echo is the most common technique and is

produced by exciting the atoms with a 90° rf pulse, then re-phasing their signal with a 180° rf

pulse.

This study was unique in that these measures were performed using a new technique that

analyses the MR data via computer analysis and generates EW values that can be compared,

before and after an intervention, and can select a region of interest to compare within the scan.

The improvement of the MR technique by characterizing $T_2$ distribution as a function of lung

water density has lead to greater accuracy in this measure (Estilaei et al., 1998). It is a clinically useful measure as many lung diseases are accompanied by changes in lung water content (Mayo et al., 1990) (Philips et al., 1989) (Hayes et al., 1982). Validation of this measure using the excised lungs of juvenile pigs, resulted in a strong relationship ($R^2$ = 0.98) between MR and gravimetric analysis (Estilaei et al., 1998).

# Appendix C: Individual Data

*Table C1. Physical and spirometric characteristics of 8 subjects*

| SUBJECT | AGE (years) | HEIGHT (cm) | WEIGHT (kg) | FVC (l) | FEV1·FVC$^{-1}$ (%) |
|---|---|---|---|---|---|
| 1 | 27 | 175.7 | 65.20 | 4.19 | 87 |
| 2 | 28 | 178.9 | 71.80 | 6.54 | 82 |
| 3 | 28 | 184.8 | 85.20 | N/A | N/A |
| 4 | 22 | 177.4 | 79.20 | 6.29 | 81 |
| 5 | 30 | 179 | 71.90 | N/A | N/A |
| 6 | 23 | 191.8 | 80.80 | 7.26 | 63 |
| 7 | 27 | 176.2 | 80.40 | N/A | N/A |
| 8 | 30 | 175 | 74.10 | N/A | N/A |

*Table C2. Maximal oxygen consumption ($\dot{V}O_2max$), peak power, minimum percent arterial oxygen saturation ($SaO_2$), and maximum heart rate (HR) during maximal cycle ergometer test.*

| SUBJECT | $\dot{V}O_2$ max l·min$^{-1}$ | $\dot{V}O_2$ max ml·min$^{-1}$·kg$^{-1}$ | POWER watts | $SaO_2$ % | HR bpm |
|---|---|---|---|---|---|
| 1 | 4.26 | 65.3 | 407 | 92 | 175 |
| 2 | 4.53 | 63.1 | 440 | 95 | 191 |
| 3 | 5.08 | 60.0 | 510 | 92 | 190 |
| 4 | 5.43 | 68.6 | 481 | 91 | 198 |
| 5 | 4.51 | 62.8 | 428 | 92 | 187 |
| 6 | 4.94 | 61.1 | 491 | 88 | 185 |
| 7 | 5.18 | 64.4 | 465 | 94 | 181 |
| 8 | 4.77 | 64.4 | 479 | N/A | 173 |

*Table C3. 45 minute cycling test values for ventilatory threshold maximal oxygen consumption (VT $\dot{V}O_2$), ventilatory threshold power (VT POWER), end of test maximal oxygen consumption (END $\dot{V}O_2$), end of test power (END POWER), and end of test heart rate (END HR).*

| SUBJECT | VT $\dot{V}O_2$ l·min$^{-1}$ | VT POWER watts | END $\dot{V}O_2$ l·min$^{-1}$ | END POWER watts | END HR bpm |
|---|---|---|---|---|---|
| 1 | 3.4 | 260 | 3.7 | 406 | 181 |
| 2 | 3.3 | 308 | 3.8 | 445 | 189 |
| 3 | 4.0 | 310 | 4.7 | 419 | 190 |
| 4 | 4.2 | 348 | 4.5 | 473 | 185 |
| 5 | 3.8 | 300 | 3.7 | 369 | 184 |
| 6 | 3.8 | 289 | 3.8 | 420 | 190 |
| 7 | 3.6 | 300 | 4.0 | 460 | 177 |
| 8 | 3.3 | 285 | 3.7 | 432 | 173 |

*Table C4.  Lung diffusion of carbon monoxide breathing 21% oxygen (DL-21%), lung diffusion of carbon monoxide breathing 90% oxygen (DL-90%), alveolar membrane ($D_M$), and alveolar volume ($V_C$).*

| SUBJECT | DL-21% ($ml \cdot min^{-1} \cdot torr^{-1}$) | | DL-90% ($ml \cdot min^{-1} \cdot torr^{-1}$) | | $D_M$ ($ml \cdot min^{-1} \cdot torr^{-1}$) | | $V_C$ (ml) | |
|---|---|---|---|---|---|---|---|---|
| | pre | post | pre | post | pre | post | pre | Post |
| 1 | 30.55 | 25.92 | 12.12 | 9.94 | 86.1 | 81.7 | 58.4 | 46.9 |
| 2 | 37.70 | 31.92 | 15.10 | 11.77 | 104.1 | 118.2 | 72.2 | 53.4 |
| 3 | 33.33 | 34.04 | 13.63 | 11.82 | 86.6 | 170.5 | 66.4 | 52.1 |
| 4 | 39.81 | 31.32 | 20.50 | 13.85 | 66.4 | 67.6 | 121.7 | 71.5 |
| 5 | 30.72 | 26.46 | 10.89 | 9.17 | 137.3 | 134.8 | 48.3 | 40.2 |
| 6 | 32.69 | 32.68 | 12.35 | 11.29 | 109.2 | 168.2 | 57.3 | 49.8 |
| 7 | 36.54 | 29.13 | 13.76 | 12.71 | 123.8 | 64.8 | 63.5 | 64.8 |
| 8 | 33.78 | 29.78 | 12.14 | 10.59 | 140.8 | 131.4 | 54.2 | 47.0 |

*Table C5.  Pulmonary extravascular water (EW) before (pre) and after (post) a 45-minute bout of cycle ergometry.*

| SUBJECT | EW ($g \cdot ml^{-1}$) | |
|---|---|---|
| | pre test | post test |
| 1 | 0.2427 | 0.2625 |
| 2 | 0.2111 | 0.2090 |
| 3 | 0.2365 | 0.3079 |
| 4 | 0.2553 | 0.3152 |
| 5 | 0.2071 | 0.1987 |
| 6 | 0.1991 | 0.1944 |
| 7 | 0.2359 | 0.2681 |
| 8 | 0.1946 | 0.1980 |

## APPENDIX D: VALIDITY OF THE MR MEASURE – *TIME BETWEEN MEASURES*

Five healthy, male, non-smoking subjects participated in MR measures, to display the effect of time of day on EW. Subjects were asked to avoid exhaustive exercise for 24 hours, caffeine for 12 hours, and food or drink for 2 hours before and between testing. Subjects were scanned twice on one day, in the morning and evening. Results are depicted in table D1.

*Table D1. Pulmonary extravascular water (EW) in the morning and evening.*

| Subject | lung water content (g·ml$^{-1}$) | |
| --- | --- | --- |
| | Morning test | Evening test |
| 1 | 0.209 | 0.218 |
| 2 | 0.298 | 0.292 |
| 3 | 0.232 | 0.241 |
| 4 | 0.216 | 0.239 |
| 5 | 0.230 | 0.288 |
| mean | 0.237 | 0.256 |
| SD | (0.0355) | (0.0326) |

When a paired t-test was performed to check for any statistically significant difference between the measures, the results came back negative ($p = 0.16$, 2-tailed). Although, it is interesting to note that lung water increased in the evening in four of the five subjects.