BLOOD-GAS BARRIER PERMEABILITY IN ATHLETES WITH EXERCISE-INDUCED HYPOXEMIA

by

MICHAEL R. EDWARDS

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Department of HUMAN KINETICS

The University of British Columbia
Vancouver, Canada

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Abstract

The effect of incremental exercise to exhaustion on the change in pulmonary clearance rate (k) of aerosolized Technetium 99m diethylenetriaminepenta acetic acid (Tc-99m DTPA), and the relationship between k and the partial pressure of arterial oxygen (PaO_2) during heavy work, was investigated. Eleven male cyclists (age = 25 ± 2 yrs, height = 181.3 ± 4.0 cm, mass = 80.1 ± 9.5 kg, $\dot{V}O_2max$ = 5.30 ± 0.37 l/min, mean ± SD) completed a pulmonary clearance test shortly following (39 ± 8 min) a $\dot{V}O_2max$ test. Resting pulmonary clearance was completed at least 24 hours before or after the exercise test. Arterial blood was sampled at rest and at one-minute intervals during exercise. Minimum PaO_2 values and maximum A-aDO_2 ranged from 73-92 Torr and 30-55 Torr respectively. No significant difference between resting k and post-exercise k for the total lung (k_T) (0.54 ± 0.19 vs. 0.57 ± 0.17, %/min, $p > 0.05$) was observed. Pearson-product moment correlation indicated no significant linear relationship between change in k_T and minimum PaO_2 ($r = -0.26$, $p > 0.05$). These results indicate that averaged over subjects, pulmonary clearance of Tc-99m DTPA following incremental maximal exercise to exhaustion in highly trained male cyclists is unchanged. Lack of a linear relationship between pulmonary clearance rate and minimum PaO_2 during exercise suggests exercise-induced hypoxemia occurs despite maintenance of alveolar epithelial integrity.

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Introduction

Exercise-induced hypoxemia (EIH), defined as the inability to maintain arterial partial pressure of oxygen (PaO_2) and arterial oxyhemoglobin saturation (SaO_2) during exercise, occurs in some high aerobic power athletes and is associated with a widened alveolar-arterial oxygen gradient ($A\text{-}aDO_2$) (Dempsey et al., 1984; Durand et al., 1999; Hopkins et al., 1994; Torre-Bueno et al., 1985). The precise etiology of EIH and the widened $A\text{-}aDO_2$ remains unclear, but is likely the result of, or an interaction among, relative alveolar hypoventilation, ventilation/perfusion ($\dot{V}_A/\dot{Q}$) inequality and diffusion limitations (Dempsey et al., 1984; Schaffartzik et al., 1992).

$\dot{V}_A/\dot{Q}$ inequality, determined by multiple inert gas elimination technique, increases with exercise (Gale et al., 1985; Schaffartzik et al., 1992). Hopkins et al., (1994) reported that 60 % of the widened $A\text{-}aDO_2$ in highly trained athletes could be explained by $\dot{V}_A/\dot{Q}$ inequality while the remainder was attributed to a diffusion limitation.

Pulmonary interstitial edema may explain both $\dot{V}_A/\dot{Q}$ inequality and diffusion limitations, and may result from increased membrane permeability and/or stress failure of the blood-gas barrier (Durand et al., 1999; West et al., 1991b). High pulmonary capillary perfusion pressure during heavy exercise may result in damage to the pulmonary capillaries and altered integrity of the blood-gas barrier. To permit efficient gas exchange by diffusion, the barrier between the alveoli and the capillaries must be extremely thin. In some sections it may only measure 0.3 μm (Gehr et al., 1978). In contrast, the blood-gas barrier must also be strong enough to withstand elevated pulmonary vascular pressure associated with high cardiac output during intense exercise.

Pulmonary arterial pressures (P_{PA}) were measured by right heart catheterization in 8 former competitive endurance athletes (Reeves et al., 1987). Mean P_{PA} at maximum exercise ($\dot{V}O_2max$ = 51.3 $ml \cdot kg^{-1} \cdot min^{-1}$, 236 watts, at sea level) was 29 ± 3 mmHg, and three subjects had P_{PA} greater than 40 mmHg. The magnitude of these pressures is consistent with other published data (Harms et al., 1998b, Wagner et al., 1986).

West et al., (1991b) introduced the concept of "stress failure" after they demonstrated disruptions to the capillary endothelium, alveolar epithelium, and their respective basement membranes, in a rabbit lung model when pulmonary arterial pressures were raised to and above 40 mmHg (West et al., 1991b). Indirect evidence that may support the theory of stress failure in humans includes several anecdotal reports of human athletes who have coughed up or tasted blood after strenuous exercise (Weiler-Rarell et al., 1995; West et al., 1991a). In addition, Hopkins et al., (1997) reported an increase in red blood cells and protein in bronchoalveolar lavage fluid obtained following seven minutes of maximal exercise. This finding suggested structural failure of the blood-gas barrier. However, PaO_2 or SaO_2 were not measured which leaves the relationship between blood-gas barrier integrity and gas exchange during intense exercise unresolved.

Tc-99m DTPA has been used as a non-specific, but extremely sensitive method for detecting lung pathology (Barrowcliffe & Jones 1989; O'Doherty & Peters 1997). Previous studies have calculated the intra-individual coefficients of variation between 7-18 % (Groth et al., 1989; Nolop et al., 1986; Smith et al., 1992). Tc-99m DTPA is hydrophilic and normally limited to passive diffusion through the intercellular junctions of the alveolar epithelium and the capillary endothelium (Effros & Mason 1983; Jones et al., 1983). Alveolar epithelial junctions are 10 times less permeable than capillary endothelial junctions (Gorin & Stewart 1979) and therefore diffusion of Tc-99m DTPA through the blood-gas barrier is primarily dependent on the permeability of the alveolar epithelial membrane. Damage to the blood-gas barrier during exercise, sufficient to alter permeability of the alveolar epithelium, would likely be detected by the change in the pulmonary clearance rate of technetium-99m diethylenetriaminepenta acetic acid (Tc-99m DTPA). We hypothesized that alveolar epithelial permeability in highly trained cyclists following an incremental exercise test to exhaustion would be increased and related to the impairment in gas exchange.

Methods

Subjects and Preliminary Tests

Subjects reported to the laboratory at least 3 hours postprandial and 24 hours post-exhaustive exercise. Spirometry involved resting measurements of forced vital capacity (FVC), forced expiratory volume in one second ($FEV_{1.0}$), and maximal forced expiratory flow (MFEF) (MedGraphics CPX-D Metabolic Cart, St. Paul, Minnesota). A $\dot{V}O_2$max test was performed on an electronically braked cycle ergometer (Quinton Excalibur, Gronigen, the Netherlands) starting at zero watts and increasing by 30 watts·min^{-1} until volitional fatigue. A minimum $\dot{V}O_2$max of 65 ml·kg·min^{-1} or 5.0 l·min^{-1} (determined as the average of the two highest consecutive 15 second $\dot{V}O_2$ scores) was required for further participation in the study. Ten competitive male cyclists or triathletes who met this criterion were selected. All subjects were nonsmokers and had no history of cardiorespiratory disease. Participants signed an informed consent that described the inherent risks associated with the study. All experimental procedures were approved by the University Clinical Ethics Review Board.

Experimental Protocol

At least 48 hours following preliminary tests subjects returned to the laboratory. On arrival, an arterial catheter was inserted into the radial artery of the non-dominant wrist prior to exercise, and blood samples taken were used to measure PaO_2, carbon dioxide ($PaCO_2$), bicarbonate (HCO_3^-) and pH. Following catheter insertion, subjects completed a $\dot{V}O_2$max test (using the same protocol as described in the preliminary tests). Three milliliter arterial blood samples were collected immediately before the onset of exercise and at one minute intervals (starting at minute four) for the duration of the exercise test. Minute ventilation ($\dot{V}E$), oxygen consumption ($\dot{V}O_2$), and heart rate (HR) were continuously measured during exercise. Pulmonary clearance using 99m Tc-99m DTPA aerosol was determined in all subjects shortly following the exercise test, with an average time interval between the completion of the exercise test and post pulmonary clearance of 38 ± 8 min (mean ± SD). Resting pulmonary clearance was completed at least 24 hours

either before or after, the exercise test. Heart rate was recorded during both lung clearance tests.

Instrumentation

Ventilatory parameters were measured for the duration of the exercise tests using a low resistance, two-way nonrebreathing valve (model 2700B, Hans Rudolph) and a 5 litre mixing chamber for collection of expired gases. From this chamber, continuous samples of air were analyzed for concentrations of oxygen and carbon dioxide at a rate of 300 $ml \cdot min^{-1}$ (S-3A oxygen analyzer and CD-3A carbon dioxide analyzer, Applied Electrochemistry). Analyzers were calibrated with gases of known concentration prior to each test. Inspired ventilation was measured with a flowmeter (Vacumetrics #17150). This device was calibrated by pumping 100 litres of air through the system. Average ventilatory and gas exchange parameters were recorded every 15 seconds using a computerized system (Rayfield, Waitsfield,VT). HR was measured continuously and recorded every 15 seconds using a portable HR monitor (Polar Vantage XL, Kempele, Finland). An oximeter (Ohmeda Biox 3740, Louisville, CO) was attached to the ear to measure oxyhemoglobin saturation (% SaO_2) after a vasodilator nicotine cream (Finalgon, Boehringer Ingelheim, Burlington, ON) was applied to the pinna to improve perfusion. Arterial oxygen hemoglobin saturation levels, determined by ear oximetry and calculated by arterial blood, were significantly correlated ($r = 0.88$, $p < 0.05$). However, there was a significant difference between ear oximetry and calculated arterial SaO_2 (t value = 10.8, $p < 0.05$) obtained during the exercise test. On average, ear oximetry tended to underestimate the arterial blood saturation by 0.9 % (95% CI: 0.75 - 1.08 %).

Following Allen's test for collateral circulation, a 20-gauge arterial catheter was inserted into the radial artery of the non-dominant wrist by percutaneous cannulation using 1 % lidocaine and sterile technique. Minimum volume extension tubing, connected in series with two, three-way stopcocks arranged at right angles, was flushed with saline-heparin solution (1 ml 1:1000 units in 500 ml NS). A rapid response (< 0.01 s) thermister (18T, Physitemp Instruments, Clifton, NJ) was inserted through a Touhy-Borsch heparin lock (Abbott Hospitals, North Chicago, IL) and used to measure peak arterial blood

temperature during collection of the blood sample. Catheter patency was maintained with intermittent heparin infusion (less than 3 ml/hr). At the onset of sampling, twelve milliliters of blood was withdrawn and peak arterial temperature was recorded. The last 2 milliliters were then collected in preheparinized syringes for blood-gas measurements and the remaining 10 milliliters was reinfused into the subject. Blood samples were placed on ice until analyzed for H^+ ion concentration, PO_2, $PaCO_2$, and HCO_3^- (CIBA-Corning 278 Blood Gas System , CIBA-Corning Diagnostics Corporation, Medfield, MA). Blood gases were corrected for temperature. Arterial blood temperature increased 0.8 ± 0.2 degrees Celsius during the VO_2max test. SaO_2 levels were calculated based on PaO_2, changes in body temperature and pH. The alveolar gas equation was used to calculate the alveolar partial pressure of oxygen (PAO_2) and A-aDO_2 (Otis 1964).

The aerosol was created in a nebulizer by introducing 20-30 mCi (740-1110 MBq) of Tc-99m DTPA in 2 ml of saline and driving it with compressed air at a flow rate of 10 l/min (Venti-Scan III Disposable, Biodex Medical, New York). Breathing procedures were rehearsed prior to inhalation of the aerosol to assure optimal delivery. Each subject was fitted with a nose clip and mouth piece, and instructed to breathe through a two-way valve at normal tidal volumes for 3 minutes in the seated position. The exhalate was trapped in a filter. A dose of approximately 1-2 mCi (37-74 MBq) was administered to the subject. The subject was seated with a large field of view gamma scintillation camera (Siemens Orbiter, Iselin, NJ) positioned posteriorly and set to image the entire lungs for 30 minutes. Subjects were instructed to remain motionless during data acquisition.

Data analysis

Output from the gamma camera was processed by computer. Decline in radioactivity over time was recorded in thirty second image frames consisting of a computer image of 128 x 128 pixels. Computer analysis of regions of interest (ROI) were placed around each lung (left and right), then further divided into regions corresponding to apical and basal lung regions. The data obtained for the total, left, right, apical, and basal were corrected for physical decay, then plotted as a function of time. The data for all regions were fit by a mono-exponential function $N = N_o e^{-kt}$, where N_o is the y-intercept or count

rate at $t = 0$, N is the count rate at any time t (min), and k is the rate constant for clearance. Total (k_T), right (k_R), left (k_L), apical (k_A), and basal (k_B) clearance rate constants were determined over the acquisition period and expressed as a percentage of decreased radioactivity per minute ($\%\cdot min^{-1}$). No correction for recirculation was made. Blood samples occasionally clotted. One subject elected not to undergo catheterization and therefore no blood-gas data was obtained for this individual.

Statistical analyses

Differences in resting k_T and post-exercise k_T were analyzed by one-way ANOVA with repeated measures. Two, 2 (resting/post-exercise) x 2 (region) ANOVA with repeated measures on both factors were used to assess differences for k_R and k_L, as well as differences for k_B and k_A. Pearson-product moment correlation was used to determine the strength of the linear relationship between PaO_2 and $A\text{-}aDO_2$, and between the change in k_T (Δk_T = post exercise k_T – resting k_T) and the minimum PaO_2 measured during exercise. A t-test for dependent means was used to determine differences between heart rates measured during the pulmonary clearance tests. A t-test for dependent means was also used to determine differences between PaO_2 measured prior to exercise and at maximum exercise. All significance was set at $\alpha < 0.05$.

Results

Performance

Anthropometic and lung parameters are shown in Table 1. Forced vital capacity (FVC), forced expiratory volume in one-second ($FEV_{1.0}$), and maximal forced expiratory flow (MFEF) were within normal values predicted for men of similar age, height and weight. Performance variables obtained at maximal exercise are shown in Table 2. Maximum respiratory exchange ratio (RER) exceeded 1.15 and peak heart rate exceeded 90% of maximum predicted heart rate (220 – age) in all subjects.

Table 1. *Physical characteristics and spirometry (n = 11).*

Age, yrs	25 ± 2
Height, cm	181.3 ± 4.0
Mass, kg	80.1 ± 9.0
FVC, litres	5.9 ± 0.6
$FEV_{1.0}$, litres	4.8 ± 0.5
$FEV_{1.0}$/FVC	81.4 ± 5.7
MFEF, l/sec	10.5 ± 1.2

Values are means ± SD. FVC, forced vital capacity; $FEV_{1.0}$, expiratory volume in 1 sec; MFEF, maximum forced expiratory flow.

Table 2. *Performance and ventilatory parameters at maximum exercise (n=11).*

HR, bts/min	186 ± 10
$\dot{V}E$, litres	200.7 ± 21.0
Power, watts	482 ± 32
$\dot{V}O_2max$, l/min	5.30 ± 0.37
$\dot{V}O_2max$, ml/kg/min	66.6 ± 6.5
$\dot{V}CO_2$, l/min	6.46 ± 0.38
RER	1.23 ± 0.05

Values are means ± SD. HR, heart rate; $\dot{V}E$, minute ventilation; $\dot{V}O_2max$, maximum O_2 consumption; $\dot{V}CO_2$, maximum CO_2 production; RER, respiratory exchange ratio.

Gas exchange

Figure 1 shows individual PaO_2, A-aDO_2 and $PaCO_2$, respectively, during the progressive exercise test. Mean PaO_2 at maximal exercise decreased significantly from resting levels (114.3 ± 4.4 vs 87.1 ± 8.4 Torr resting and maximal exercise, respectively, $p < 0.05$). In three subjects, the lowest PaO_2 during exercise remained between 90-100 Torr, in five subjects the lowest PaO_2 fell between 80-90 Torr, and in the three remaining subjects, PaO_2 during exercise fell below 80 Torr. The maximum A-aDO_2 ranged from 30 - 55 Torr (42.5 ± 7.0 Torr, mean ± SD). There was a significant negative correlation between PaO_2 and A-aDO_2 ($r = -0.94$, $p < 0.05$). At maximal exercise, five subjects decreased $PaCO_2$ levels below 36 Torr, while in four subjects, $PaCO_2$ values did not fall below 38 Torr. Arterial pH and HCO_3^- levels remained unchanged from light to moderate exercise intensity, but progressively declined during moderate to heavy exercise. Minimal values at maximal exercise for pH and HCO_3^- reached 7.2 ± 0.1 and 14.1 ± 2.5 nmol·litre^{-1} respectively.

Pulmonary clearance

The quality of the goodness of fit (R^2) for the total lung averaged 0.95. The resting and post-exercise k_T for individual subjects is shown in Figure 2. Pulmonary clearance rates, averaged over subjects, for each region, rest and post-exercise, are shown in Figure 3.

One-way ANOVA demonstrated resting and post-exercise k_T were not significantly different averaged over subjects (0.54 ± 0.19 vs 0.57 ± 0.17 %/min, rest and post-exercise, respectively, $F = 0.22$, $p > 0.05$).

The region main effect for k_L and k_R lungs was not significant ($F_{1,10} = 2.25$, $p > 0.05$), indicating that averaged over rest and post-exercise there were no differences in clearance rates between the right (0.58 ± 0.19 %/min) and left lungs (0.53 ± 0.20 %/min). The time main effect for resting and post-exercise was not significant ($F_{1,10} = 0.20$, $p > 0.05$), indicating that averaged over left and right lung regions there were no differences in clearance rates between rest (0.59 ± 0.37 %/min) and post-exercise (0.64 ± 0.42 %/min). The region by time interaction was also not significant ($F_{1,10} = 2.25$, $p > 0.05$),

indicating that differences in k_L and k_R had similar clearance rates at rest and post-exercise.

The region main effect for k_A and k_B was significant ($F_{1,10}$ = 5.76, $p < 0.05$), indicating that averaged over rest and post-exercise there were differences in clearance rates between the apices (0.78 ± 0.36 %/min) and basal regions (0.44 ± 0.34 %/min). The time main effect for rest and post-exercise was not significant ($F_{1,10}$ = 0.44, $p > 0.05$), indicating that averaged over apical and basal lung regions, there were no differences in clearance rates between rest (0.54 ± 0.20 %/min) and post-exercise (0.57 ± 0.20 %/min). The region by time interaction was not significant ($F_{1,10} = 0.33$, $p > 0.05$), indicating that differences in k_A and k_B had similar clearance rates at rest and post exercise.

Heart rates determined during the inhalation of the aerosol were significantly greater post exercise compared to resting (74 ± 12 vs 57 ± 8 bts/min, respectively, t value = 5.4, $p < 0.05$).

Pulmonary clearance and gas exchange

There was no significant relationship ($r = -0.26$, $p > 0.05$) between the minimum PaO_2 obtained during the exercise test and Δk_T (Figure 4). In addition, there was no significant relationship ($r = 0.07$, $p > 0.05$) between the maximal A-aDO_2 and Δk_T.

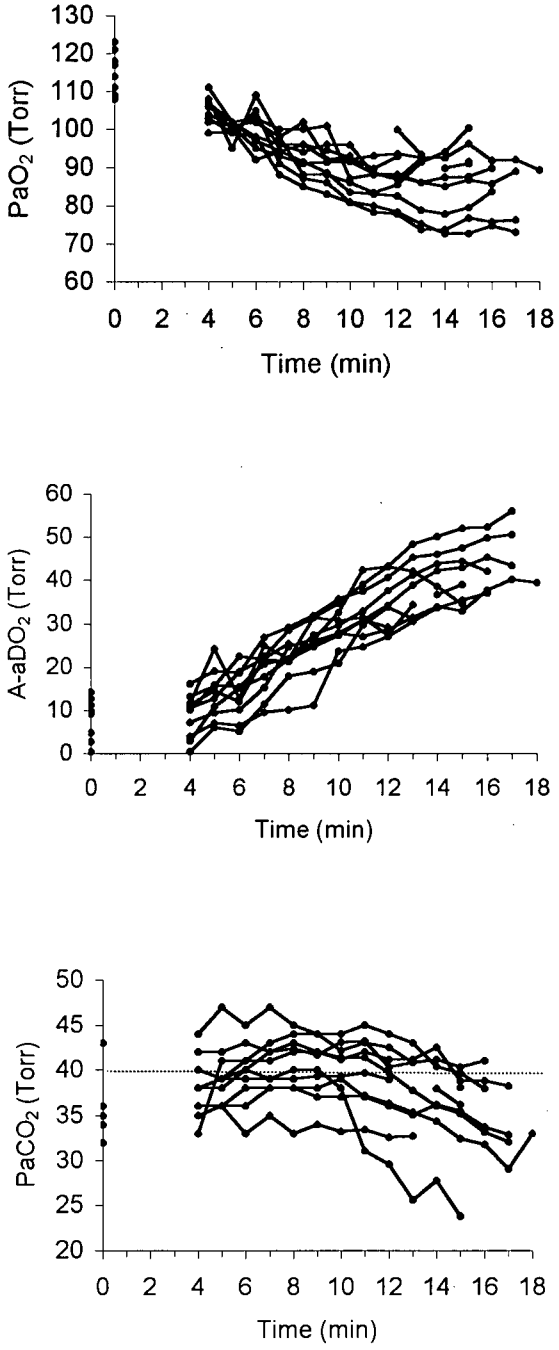

Figure 1. Individual time course data for arterial partial pressure of oxygen, PaO_2 (*top*), alveolar-arterial oxygen gradient, A-aDO_2 (*middle*), and arterial partial pressure of carbon dioxide, $PaCO_2$ (*bottom*) during the progressive exercise test. Vertical line in bottom panel indicates line of identity. Data from 10 of 11 subjects.

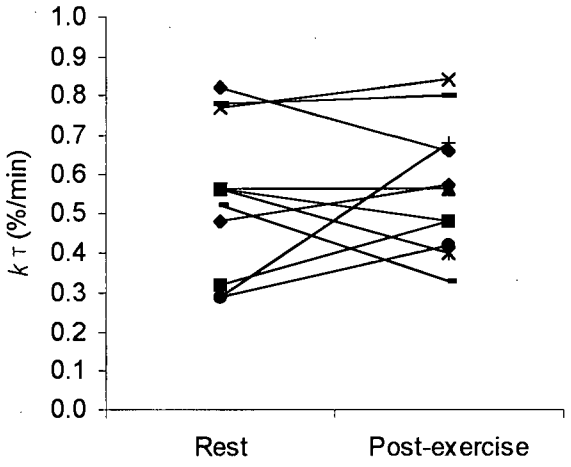

Figure 2. Individual pulmonary clearance rates for the total lung (k_T) at rest and post-exercise.

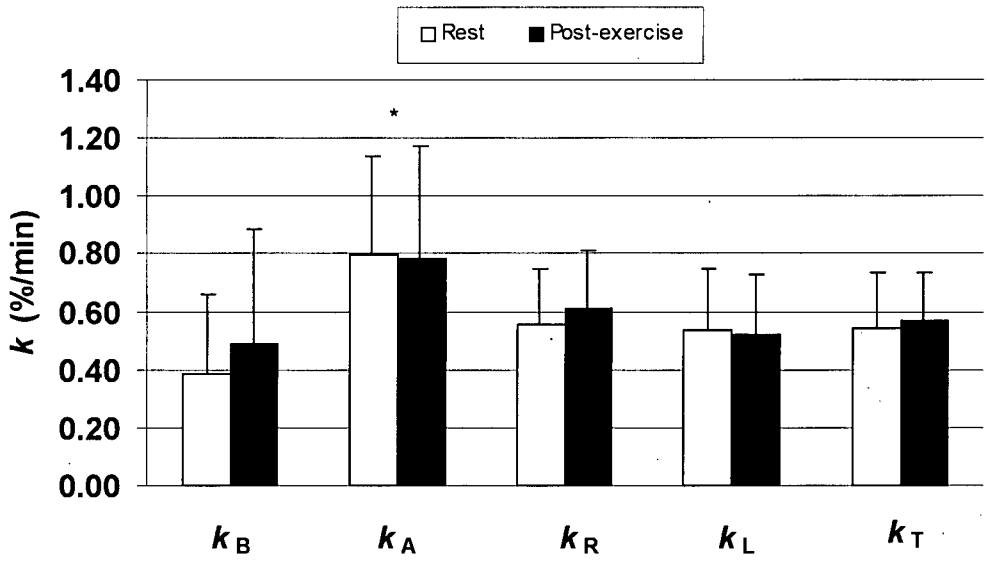

Figure 3. Mean values ($n = 11$) for pulmonary clearance rates (k) at rest and post-exercise and divided into basal (k_B), apical (k_A), right (k_R), and left (k_L) regions as well as total (k_T). Error bars represent SD. * denotes significant difference ($p < 0.05$) between apical and basal regions.

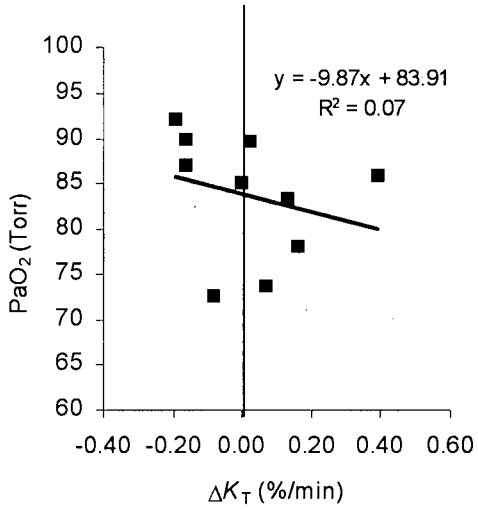

Figure 4. Relationship between the change (post-exercise – resting) in total pulmonary clearance rate (Δk_T) and minimum PaO_2 recorded during exercise test for each subject. Bold line represents linear regression line; equation and R^2 is noted. Vertical line indicates zero change in pulmonary clearance. Data from 10 of 11 subjects.

Discussion

Pulmonary clearance and exercise

$\dot{V}O_2$max values and peak power outputs (Table 2) indicated our subjects were highly trained while the range of EIH and A-aDO_2 during exercise was consistent with other studies involving highly trained male athletes (Dempsey et al., 1984; Powers et al., 1988, Hopkins et al., 1994).

The present study failed to demonstrate a change in alveolar epithelial permeability in humans following incremental exercise to exhaustion. In addition, no relationship between minimum PaO_2 during exercise and change in pulmonary clearance rate was observed, suggesting exercise-induced hypoxemia and an elevated A-aDO_2 occurs in these subjects despite maintenance of alveolar epithelial integrity.

The mechanism(s) of exercise-induced elevation of A-aDO_2 and reduction of PaO_2 remains unclear. However, pulmonary interstitial edema may explain the $\dot{V}_A/\dot{Q}$ inequality observed both during and post-exercise (Schaffartzik et al., 1992). Human pulmonary artery pressures at maximal exercise can reach 40 mmHg (Wagner et al., 1986), which is in the range shown to cause stress failure of the blood-gas barrier in a rabbit lung model (West et al., 1991b). At this magnitude raised transmural pulmonary vascular pressures in rabbits show disruption to the capillary endothelium, the alveolar epithelium, and in some cases red blood cells can be seen passing through the membranes (Tsukimoto et al., 1991; West et al., 1991b).

Hopkins et al., (1997) reported an increase in red blood cells (RBC) and protein in the bronchoalveolar lavage fluid from athletes following seven minutes of heavy exercise compared to resting sedentary controls. These results suggested loss of blood-gas barrier integrity. All six athletes recruited, however, had a history of hemoptysis or tasting blood following intense exercise. None of the athletes in the present study had such a history. Therefore, the conclusions from this study and the present one may not be comparable.

The results from the present study do not exclude pulmonary interstitial edema as a possible mechanism for the EIH and elevated A-aDO_2 observed in our subjects. However, early reports from imaging studies following short term, heavy exercise have been unable to detect evidence of extravascular lung water. Caillaud et al., (1995) reported unpublished data of computerized tomography (CT) scans before and after a 13-minute incremental, exhaustive exercise test, showing no significant changes in lung density. In addition, McKenzie et al., (1996) found no evidence of extravascular lung water with magnetic resonance images (MRI) or CT in athletes after five minutes of intense exercise averaging 435 watts.

There is some evidence for extravascular lung water following prolonged, submaximal exercise. CT scans in highly trained athletes after completing a triathlon (105-135 minutes) found an increased lung density suggestive of a mild subclinical pulmonary edema (Caillaud et al., 1995). Recent MRI data from our laboratory has shown similar results (McKenzie et al., 1999b). In contrast, Manier et al., (1999) failed to observe an increase in post-exercise lung mass with CT following 2 hr of treadmill running at 75% of $\dot{V}O_2max$. Hopkins et al., (1998c) performed bronchoalveolar lavage in athletes following one-hour of submaximal exercise (77 % of $\dot{V}O_2max$) and found no evidence of RBC and protein, indicating no or little damage to the membrane. There was an unexpected finding of RBCs in the lavage fluid of resting control subjects, questioning the sensitivity of this technique for investigating blood-gas barrier integrity. Collectively, it appears prolonged submaximal exercise may alter Starling's forces and result in a net fluid flux with or without altering the structural integrity of the blood-gas barrier. Unfortunately, none of these studies measured PaO_2 or SaO_2, and, therefore, it is unknown if extravascular lung water from submaximal exercise is of sufficient magnitude to impair gas exchange.

Lorino et al., (1989) assessed pulmonary clearance with Tc-99m DTPA in 7 healthy volunteers pre and post 75 min of exercise corresponding to 75% of the subject's $\dot{V}O_2max$. Post exercise, total, apical, and basal clearance rates were all significantly increased. They concluded that the increased alveolar permeability was related to the

mechanical effects of prolonged, high ventilation rates. This is compatible with data from the rabbit model where increasing lung volumes while maintaining constant pulmonary arterial pressure increased the incidence of stress failure in rabbits (Fu et al., 1992). In a pilot study, we investigated whether ventilation rates alone would alter alveolar permeability in five subjects (Appendix IV). Isocapneic ventilation rates were assigned following data previously obtained during a $\dot{V}O_2$max test. Mean $\dot{V}E$ reached a maximum value of 145 ± 29 l/min during isocapneic ventilation. Pre and post (30 ± 5 min) total pulmonary clearance rates remained unchanged (0.54 ± 0.02, 0.57 ± 0.03 %/min, respectively, $p > 0.05$). $\dot{V}O_2$max values in the study by Lorino et al., (1989) only reached 53 ± 3 ml/kg/min and, therefore, we believe stress failure would have been unlikely in these subjects as pulmonary arterial pressures do not reach high values during submaximal exercise, even if exercise is prolonged (Hopkins et al., 1998b). Therefore, it seems unlikely that mechanical effects of sustained ventilation and/or stress failure altered the alveolar epithelial permeability in the study by Lorino et al., (1989).

St. Croix et al., (1998) argued EIH during heavy exercise may reflect a functionally based mechanism, present only during exercise, rather than stress failure because a structural mechanism would be expected to have lasting effects. Their subjects performed a progressive incremental exercise test to $\dot{V}O_2$max followed by a constant load at maximal workload 20 minutes later. A slight improvement in PaO_2 and A-aDO_2 was found during the second exercise bout. Therefore, a functional mechanism, such as decreased mixed venous oxygen saturation, combined with high cardiac output to an already fully dilated and recruited pulmonary vasculature, may decrease pulmonary capillary transit time and result in decreased end-capillary oxygen, widened A-aDO_2 and EIH (Hopkins et al., 1996).

Methodological concerns

All subjects in the present study were non-smokers. Pulmonary clearance rates in our subjects at rest were considered normal for healthy individuals (Smith et al., 1992). Faster apical clearance rates compared to basal rates reflects greater apical ventilation

and greater surface area available for diffusion of Tc-99m DTPA (Marks et al., 1985, Mason et al., 1985; Meignan et al., 1987).

It is possible blood-gas barrier remodeling occurred in the time delay between the exercise test and the post-exercise pulmonary clearance test. This time delay was designed to allow pulmonary blood flow and ventilation to return to near resting values. However, heart rates recorded during the inhalation of the aerosol remained significantly elevated post-exercise compared to resting, and it is uncertain if pulmonary capillary blood volume, and/or the area available for gas exchange, was altered.

Background correction for re-circulation of radioactivity through the pulmonary capillaries is of controversial significance (Coates et al., 1988). Some studies (Lorino et al., 1989; Meignan et al., 1986) have ignored background correction because there may be considerable error in calculation and because background radioactivity does not appear to significantly effect the measured clearance rate (Meignan et al., 1986). In addition, the rate at which the aerosol leaves the blood by normal kidney filtration is 10 fold faster than the rate the aerosol enters the blood through the blood-gas barrier (Hilson et al., 1976). In contrast, using the liver for background correction, Mason et al., (1997) recently demonstrated that when intravenous DTPA was administered prior to inhalation of the aerosol, pulmonary clearance curves were multiexponential. However, using the thigh for background correction yielded pulmonary clearance curves that were monoexponential. Apparently the liver has a closer extravascular-to-intravascular compartment ratio to the lung and, therefore, the authors concluded that liver background correction allows the true shape of the curve to be identified. Clearance curves in the present study were analyzed for 30 minutes and not corrected for re-circulation. Staub et al., (1990) reported that correction for re-circulation was less of a concern if clearance curves were calculated within this time period.

In conclusion, averaged over subjects, pulmonary clearance of Tc-99m DTPA following incremental maximal exercise to exhaustion in highly trained male cyclists was unchanged. Furthermore, there was no relationship between altered pulmonary clearance

and the minimum arterial partial pressure of oxygen during heavy exercise, suggesting exercise-induced hypoxemia occurs despite maintenance of alveolar epithelial integrity.

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Appendix I. Literature review: Exercise-induced hypoxemia

Definition and Incidence

Untrained and moderately trained individuals maintain arterial oxygen tension (PaO_2) and arterial oxygen hemoglobin saturation (SaO_2) homeostasis even during maximal exercise (Asmussen & Nielsen 1960; Powers et al., 1988; Williams et al., 1986). For these individuals the lung is considered more than adequate to meet the demands of the exercising body. In the late fifties and early sixties a few authors suggested that some high aerobic power athletes were unable to maintain PaO_2 and SaO_2 under intense exercise (Holmgren & Linderholm 1958; Rowell et al., 1964). However it was not until 1984 when Dempsey et al., (1984) carefully documented moderate to severely reduced PaO_2 levels in 12 of 16 highly trained runners that exercise-induced hypoxemia (EIH) was recognized as a common occurrence in athletes and was not simply a measurement artifact. Numerous researchers have since confirmed EIH in highly trained athletes (Durand et al., 1999; Hopkins & McKenzie 1989; Powers et al., 1992; Wagner et al., 1986).

Various measurements are used to define EIH. These include: 1) a decrease in PaO_2 to less than 75 mmHg (Dempsey et al., 1984), 2) a SaO_2 of less than or equal to 91 %, (Williams et al., 1986) and 3) a decrease in PaO_2 of 10 mmHg or greater compared to resting levels (Durand et al., 1999; Harms et al., 1998a). Dempsey et al., (1984) demonstrated that in a group of 16 high aerobic power runners ($\dot{V}O_2max$ = 72 $ml \cdot kg^{-1} \cdot min^{-1}$), 50 percent had oxygen hemoglobin saturation levels that fell below 91 percent and PaO_2 that fell below 75 mmHg. The incidence appears to be even greater in male masters athletes (Prefaut et al., 1994; Prefaut et al., 1997) and in women (Harms et al., 1998a). The practical application of this finding is this level of arterial blood desaturation has been shown to impair $\dot{V}O_2max$ (Powers et al., 1989) and possibly reduce endurance performance (Koskolou & McKenzie 1994).

Etiology

The precise etiology of exercise-induced hypoxemia remains unclear despite significant research efforts. Profound physiological changes occur with heavy exercise and the lung is challenged to maintain end-capillary oxygen and thus arterial oxygen saturation homeostasis. Increased blood volume in the already fully dilated and recruited pulmonary vasculature would be expected to increase pulmonary pressures and/or decrease the pulmonary capillary transit time (PCTT). An increase in pulmonary pressures may lead to stress failure of the blood-gas barrier and/or an inflammatory response secondary to stress failure, and/or altered Starlings forces and an increased fluid flux in the interstitial spaces (edema). Furthermore, mismatching and redistribution of blood flow along with relative alveolar hypoventilation may also result in decreased end-capillary oxygen. Investigators have examined these possible mechanisms for EIH and it seems likely that the development of EIH is caused by, or is an interaction among, alveolar hypoventilation, ventilation/perfusion ($\dot{V}_A/\dot{Q}$) mismatch, and diffusion limitations.

Alveolar Hypoventilation

Alveolar hypoventilation has been defined as a ventilation rate lower than necessary to maintain blood gases at resting levels (Powers et al., 1993). Hyperventilation occurs during heavy exercise which functions to increase the partial pressure of alveolar oxygen (PAO_2), thus increasing the "driving pressure" necessary for oxygen diffusion through the blood-gas barrier and into arterial blood. If this hyperventilation does not occur (i.e. relative hypoventilation), as the result of 1) blunted respiratory drives and/or 2) mechanical limitations and/or 3) respiratory fatigue, the PAO_2 will fall. Consequently, PaO_2 will fall and the alveolar-arterial oxygen pressure difference ($A\text{-}aDO_2$) will widen.

Blunted respiratory drives

A blunted ventilatory response to exercise has been proposed to be a major cause of EIH (Dempsey et al., 1984) and may explain 50% of the SaO_2 variability observed during heavy exercise (Harms & Stager 1995). During heavy exercise, metabolic acidosis, norepinephrine and hypoxemia have been shown to increase without a compensatory

ventilatory response (Dempsey et al., 1984). A compensatory ventilatory response is assessed during heavy exercise by $PaCO_2$ levels. As exercise intensity increases, $PaCO_2$ is expected to decline. However the degree of hyperventilation varies and $PaCO_2$ ranges from approximately 28 to normal resting levels of approximately 40 mmHg. A minute ventilation in excess of 200 $l \cdot min^{-1}$ has been estimated to be necessary for an athlete producing 5-6 $l \cdot min^{-1}$ of CO_2 to prevent arterial desaturation (Dempsey 1986).

It has been suggested that a non-compensatory response indicates an inability of receptors to detect changes in chemical stimuli and respond by increasing ventilation (Dempsey 1986). Subjects with the most severe hypoxemia have been shown to have the smallest ventilatory response to exercise (Dempsey et al., 1984). In 36 healthy men, Harms & Stager (1995) found a positive relationship between hypoxic ventilatory response at rest and SaO_2 at maximal exercise ($r = 0.63$, $p < 0.05$) and between hypercapnic ventilatory response at rest and SaO_2 at maximal exercise ($r = 0.62$, $p < 0.05$) suggesting that these subjects had a blunted respiratory drive. More recently, Gavin et al., (1998) studied 13 men during an incremental maximal cycle test and used the ratio of minute ventilation to the rate of carbon dioxide production ($\dot{V}E / \dot{V}CO_2$) as an index for the ventilatory response to exercise. The men were divided into two groups based on maximal exercise $\dot{V}E / \dot{V}O_2$ and PAO_2 estimates. The subjects with low ventilatory response during normoxic exercise had greater arterial desaturation during hypoxic exercise than subjects with a high ventilatory response. However, the group with a low ventilatory response did not have decreased saturation levels during normoxic exercise.

Many of the aforementioned studies addressing blunted respiratory drive, are limited in that indirect indices have been used to characterize a blunted ventilatory response. Large inter-individual variation in the hyperventilatory response to exercise is observed and this may be unrelated to $\dot{V}E / \dot{V}O_2$, or simply $\dot{V}E$. In addition, the hypoxic ventilatory response has been traditionally measured at rest and may not reflect the response at exercise. Because of these criticisms, more research is required on the role of blunted chemoreceptors and EIH.

A dog model has recently been used to study the role of carotid body hypocapnia and ventilation (Smith et al., 1995). In this preparation, one carotid body was separated from the systemic arterial circulation and artificially perfused while the other carotid body was denervated. The results demonstrated that carotid body hypocapnia decreased the ventilatory response proportionally to the magnitude of the hypocapnia. In a follow up study, these authors found that hypocapnia can decrease the carotid body chemoreceptive response to hypoxia and reduce the ventilatory response (Smith et al., 1997). Although these studies were performed in the resting dog, similar levels of hypocapnia occur in humans performing heavy exercise and indicate that both stimulatory and inhibitory impulses occur during exercise. In addition, the role of the carotid bodies in producing hyperventilation during heavy exercise has been questioned. Bilateral denervation of the carotid body in ponies did not blunt the hyperventilatory response to heavy exercise (Pan et al., 1986). In fact, ventilation increased following denervation.

Mechanical limitations

There is some evidence to suggest that a mechanical flow limitation may result in alveolar hypoventilation and possibly contribute to hypoxemia during exercise. An expiratory flow limitation means that if expiratory flow were to increase, it must occur at an increased volume. Volume may be constrained by the elasticity of chest wall. In addition, intrathoracic pressures continue to increase during incremental exercise but are ineffective in increasing flow. In this situation, respiratory muscles are performing inefficient work.

The resting maximal volitional flow-volume envelope defines the capacity of the respiratory system for volume and flow. In four of sixteen highly trained runners, Dempsey et al., (1984) demonstrated flow-volume envelopes during heavy exercise, matched or exceeded resting flow-volume envelopes. The authors suggested that this may represent a mechanical limitation to ventilation. Similar results were found by Johnson et al., (1992).

Athletes with and without an expiratory flow limitation were compared in their ability to increase ventilation during maximal exercise while breathing a hypoxic gas mixture

(Chapman et al., 1998). The results indicated that expiratory-flow limited athletes were unable to increase ventilation during maximal exercise while breathing the hypoxic gas. However, there was no relationship between ventilation and SaO_2 and the authors concluded that mechanical limitations to ventilation were not responsible for arterial desaturation. It has been previously suggested that an expiratory flow limitation plays a secondary role in the development of hypoxemia (Johnson et al., 1992).

In another study, ten male cyclists with mean $\dot{V}O_2max$ of 72 $ml \cdot kg^{-1} \cdot min^{-1}$ and maximum $\dot{V}E$ of 147 $l \cdot min$ did not have an expiratory flow limitation during maximum exercise (Mota et al., 1999). These authors assessed flow limitation by applying a negative expiratory pressure to the mouth and the flow-volume curve of the ensuing expiration was compared with the preceding breath. This technique is thought to be more valid than previous research imposing resting flow-volume loops upon exercise flow-volume loops.

Unloading the respiratory muscles either by breathing a low density gas or by a pressure assist device provides evidence of a mechanical limitation. Breathing a low density gas (helium-oxygen mixture) increases ventilation (Dempsey et al., 1984) and the flow-volume loop by reducing the mechanical impedance as high flow rates maintain a more laminar flow. Indeed, breathing a heliox gas mixture has been shown to increase performance time (Aaron et al., 1985). A pressure assist device adds positive pressure on inspiration rather than negative pressure produced by the respiratory muscles and has been found to reduce the work of breathing.

The major consequence of a mechanical limitation to breathing appears to be an increased energy expenditure of the respiratory muscles and/or respiratory muscle fatigue which is possibly related to EIH.

Respiratory muscle fatigue

The diaphragm is composed primarily of slow twitch fibers which, combined with its high capillary density and high concentration of aerobic enzymes, makes this muscle somewhat resistant to fatigue. However, theoretical respiratory muscle fatigue, namely diaphragmatic fatigue, during exercise could prevent the respiratory muscles from

generating the required pressure for inspiration and expiration and alveolar hypoventilation may ensue.

Using various methodologies, respiratory muscle fatigue has been demonstrated following heavy short-term exercise and prolonged submaximal exercise including marathons and ultra-marathons (Bender & Martin 1985; Ker & Schultz 1996; Loke et al., 1982; McConnell et al., 1997). Inducing extreme inspiratory fatigue by repeated voluntary efforts at rest altered the ventilatory response to high-intensity exercise (Sliwinski et al., 1996).

A technique called bilateral phrenic nerve stimulation (BPNS), used post-exercise to test diaphragmatic muscle force production, has recently gained popularity as a non-subjective and motivational independent test (Johnson et al., 1996). BPNS of 10 and 20 Hz was performed in twelve healthy subjects following exercise intensities of 80 % $\dot{V}O_2max$ to exhaustion. The results indicated decreased trans-diaphragmatic pressure in one-half of the subjects following an intensity of 80-85 % of $\dot{V}O_2max$ and decreased pressure in almost all subjects following 90-95 % of $\dot{V}O_2max$. This reduction (approx. 20-40 % of resting levels) is, apparently, indicative of diaphragmatic fatigue (Babcock et al., 1998; Johnson et al., 1993). In addition, pressures remained reduced for one hour following exercise indicating that the diaphragm did not immediately recover from the exercise (Johnson et al., 1993).

Other than alveolar hypoventilation, the consequences of respiratory muscle fatigue are unclear. Present research suggests that respiratory muscles during exercise may contribute 10-15 % of $\dot{V}O_2max$ (Aaron et al., 1992) and a substantial 14-16 % of the total cardiac output (Harms et al., 1998b). Competition from the locomotor muscles may prevent optimal blood flow to the respiratory muscles. Harms et al., (1997) demonstrated that increased respiratory work may significantly compromise leg blood flow during maximal exercise. Reduction in blood flow to working muscles has been shown to decrease muscle force output (Ward et al., 1992).

Respiratory muscle training

If respiratory muscle fatigue contributes to alveolar hypoventilation it seems reasonable to suggest that respiratory muscle training could hypothetically improve performance. However, the effect of respiratory muscle training on performance has been controversial (Boutellier et al., 1992; Boutellier & Piwko 1992; Fairbarn et al., 1991; Morgan et al., 1987).

Fairbarn et al., (1991) enrolled five subjects in a four week respiratory muscle endurance training program via isocapnic hyperventilation. At the end of the training period, maximum voluntary ventilation increased significantly from 155 to 174 $l \cdot min^{-1}$ ($p < 0.01$) while the control subjects remained unchanged (155 to 150 $l \cdot min^{-1}$, $p > 0.05$); a finding consistent with other studies (Leith & Bradley 1976; Morgan et al., 1987). However, there was no change in $\dot{V}O_2max$, maximal cycling endurance time at 90 % of peak power output, or $\dot{V}E$ at $\dot{V}O_2max$ for either the experimental or control group ($p > 0.05$) suggesting that increasing respiratory muscle endurance is possible but may not enhance performance. In agreement with these studies, Morgan et al., (1987) documented no change in $\dot{V}O_2max$ or endurance cycling time at 95 % of $\dot{V}O_2max$ following 3 weeks of respiratory muscle training. Based on these findings, it was concluded that ventilation does not limit exercise in the highly trained individual (Fairbarn et al., 1991).

In contrast, other authors found that four weeks of respiratory training increased cycle endurance time by 38 % in trained individuals, at a predetermined anaerobic threshold (77 % $\dot{V}O_2max$) (Boutellier et al., 1992). In a previous study, Boutellier & Piwko (1992) reported 4 weeks of respiratory training increased endurance time in sedentary subjects by 50 %. The mechanism for this increase in performance remains purely speculative (Boutellier 1998). However, possible mechanisms include a reduced work of breathing resulting in less competition for blood flow by the working muscles (Harms et al., 1997), and/or decreased dyspnea.

Unfortunately, the methodology investigating respiratory muscle training is of some concern. Many studies have not used a control group and/or have used an unacceptable

control group. Subjects in the control groups were simply not performing any activity and therefore it is difficult to distinguish the effects of learning, dyspnea, and motivation from muscle training. In addition, the studies showing markedly improved endurance from respiratory muscle training (Boutellier et al., 1992; Boutellier & Piwko 1992) have not since been replicated. The relationship among respiratory fatigue, respiratory muscle training and EIH remains unclear.

In conclusion, the evidence seems to suggest that relative hypoventilation may be partly responsible for the development and/or exacerbate the EIH seen in athletes. The athlete may be left with a non-conscious decision: maintain arterial oxygen saturation by increasing ventilation but weigh the additional metabolic costs of doing so and/or the possible redistribution of blood flow from the exercising muscles. Other factors may be integrated into this decision including mechanical limitations to flow, respiratory muscle fatigue, and/or the sensation of dyspnea.

Ventilation-Perfusion Mismatch

A ventilation perfusion mismatch ($\dot{V}_A/\dot{Q}$) is when there is a non-uniformity in some regions of the lung between the air flow entering and leaving the alveoli and the blood flowing through the capillaries. This non-uniformity may be the result of inadequate alveoli ventilation, poor capillary blood flow, or a combination of both. The consequence is often poor or inefficient gas exchange of oxygen and carbon dioxide.

Multiple inert gas elimination technique (MIGET) is an indirect method of assessing $\dot{V}_A/\dot{Q}$ inequalities and intrapulmonary shunt. A multicompartmental model was developed (Wagner et al., 1974) to predict the A-aDO_2 gradient based on data from infusion and elimination of inert gas. The predicted A-aDO_2 is then compared to the observed A-aDO_2. The predicted value is thought to represent a $\dot{V}_A/\dot{Q}$ inequality and shunt whereas the difference between the predicted and observed A-aDO_2 is thought to represent a diffusion limitation and post-pulmonary shunt. Post-pulmonary shunt has been disregarded as breathing 100 % O_2 eliminates the A-aDO_2 gradient.

Using MIGET, $\dot{V}_A/\dot{Q}$ inequality has been shown to increase with exercise (Gale et al., 1985; Hopkins et al., 1994; Schaffartzik et al., 1992). In moderately trained subjects ($\dot{V}O_2$ = 3.0 l·min^{-1}), the observed A-aDO_2 was greater than the predicted A-aDO_2 suggesting a diffusion limitation in these individuals (Hammond et al., 1986). Furthermore, Wagner et al., (1986) tested 8 healthy individuals with a mean $\dot{V}O_2$max of 3.7 l·min^{-1} and showed that two-thirds of the A-aDO_2 can be explained by a diffusion limitation and remainder is explained by $\dot{V}_A/\dot{Q}$ mismatch. However, recent work suggests that $\dot{V}_A/\dot{Q}$ mismatching is a major contributor to the widening of the A-aDO_2 gradient in highly trained athletes (Hopkins et al., 1994). In this MIGET study, ten high aerobic power athletes ($\dot{V}O_2$max > 5.0 l·min^{-1}) performed exercise at various intensities (150 watts, 300 watts and maximal exercise (372 ± 22 watts)). Although the observed A-aDO_2 increased significantly more than the predicted A-aDO_2, the authors reported that 60 % of widening A-aDO_2 was explained by $\dot{V}_A/\dot{Q}$ mismatch while the remainder could be attributed to a diffusion limitation.

Although MIGET studies provide insight into gas exchange, they are unable to discern the exact mechanism for the cause of widening A-aDO_2 and have resulted in much speculation.

Diffusion Limitations

Increased oxygen extraction by the working muscles widens the pulmonary arterial-venous oxygen gradient and therefore imposes a further demand on diffusion to maintain oxygen homeostasis. Maximal aerobic power and cardiac output can increase from resting levels more than ten fold. The pulmonary vascular system is a unique organ in that it must accept the entire cardiac output. During heavy exercise the pulmonary vasculature is thought to be fully recruited and distended. Any further increase in cardiac output and left ventricular filling pressures is expected to decrease red blood cell (RBC) pulmonary capillary transit time (PCTT) or increase pulmonary pressure.

PCTT is one aspect of EIH that has been inadequately investigated. The reason may lie in the difficulty of measuring such a phenomenon. It is only possible to measure the mean RBC transit time from right to left heart and not individual RBC transit times. Therefore, PCTT can only be estimated.

Cardiac output increases with exercise intensity. The pulmonary vasculature accepts the entire cardiac output through distension and recruitment of pulmonary capillaries. It is hypothesized that at a cardiac output of 25 $l \cdot min^{-1}$ or a $\dot{V}O_2$ of 3.5 $l \cdot min^{-1}$, the pulmonary vascular reaches its maximum morphological limit (Dempsey et al., 1982). Theoretically, increasing the cardiac output further would decrease the transit time of a RBC through the capillary bed and consequently result in an oxygen disequilibrium and a widening of the A-aDO_2 gradient.

At cardiac outputs of 25 $l \cdot min^{-1}$, PCTT is thought to be 0.35-0.40 seconds, adequate for O_2 equilibrium (Dempsey et al., 1982). Since cardiac output in some athletes may be as high as 40 $l \cdot min^{-1}$ (Ekblom & Hermansen 1968) this may further reduce the PCTT.

To address the issue of PCTT, ten high aerobic power subjects, previously showing evidence suggestive of diffusion disequilibrium by MIGET (Hopkins et al., 1994), underwent RBC labeling to determine pulmonary transit times while exercising at an intensity above 90 % of $\dot{V}O_2max$ (Hopkins et al., 1996). Cardiac output, measured by radiocardiography, during exercise reached 33 $l \cdot min^{-1}$ and whole lung transit time decreased from 9.32 to 2.91 seconds; estimating PCTT at 0.39-0.41 seconds. However, the frequency distribution of transit times estimated that 40 % of RBC had PCTT of less than 0.30 seconds and 15 % of RBC had PCTT of less than 0.14 seconds which would be inadequate for efficient gas exchange.

In contrast, Warren et al., (1991) exercised subjects on a bicycle ergometer at multiple workloads reaching 88 % of $\dot{V}O_2max$ ($\dot{V}O_2$ = 4.31 $l \cdot min^{-1}$) and measured capillary volume (Vc) by diffusion capacity of carbon monoxide (DLCO) and cardiac output by echocardiogram. PCTT was determined by dividing Vc by $\dot{Q}$. The authors concluded that although the A-aDO_2 increased and PO_2 decreased, they were not caused by a

plateau in capillary blood volume and consequently a decline in PCTT. However, this study has been criticized (Hopkins et al., 1996) as intensity may not have been of sufficient magnitude to cause a plateau in Vc.

PCTT is often overlooked as a mechanism for EIH as both athletes with and without EIH have similarly high $\dot{V}O_2$max levels and presumably similar cardiac outputs and therefore thought to have similarly sized vasculature beds. However, pulmonary blood capacity has not been determined in athletes and it is conceivable that those with EIH have a smaller pulmonary vasculature and hence, chronically faster PCTT. This idea has been alluded to by (Hopkins et al., 1998b). Hopkins et al., (1998b) found a high correlation ($r = -0.97$, $p < 0.05$, $n = 6$) between lung size and $\dot{V}_A/\dot{Q}$ inequality in athletes. It was speculated that individuals with smaller lung sizes might also have smaller airways and blood vessels. Similar findings show that subjects susceptible to high altitude pulmonary edema (HAPE) have a smaller forced vital capacity and more $\dot{V}_A/\dot{Q}$ inequalities (Eldridge et al., 1996).

In conclusion it seems reasonable to suggest that PCTT plays a significant role in the widening of the A-aDO_2 gradient during heavy exercise. More research is necessary to clarify this issue.

Elevated Pulmonary Pressures and its Consequence

Changes in pulmonary hemodynamics may be responsible for the formation of EIH. Compared to the systemic circulation, the lung is generally thought to be a low-pressure system. However, because of the invasiveness of the Swan-Ganz catheter, there have been limited studies in which pulmonary arterial pressures have been measured in healthy humans under resting conditions. The estimated mean arterial pressure and standard deviation for human adults is approximately 14 ± 3 mmHg. Based on these values, less than 1 % of normal adults would be expected to have pulmonary arterial pressures in excess of 25 mmHg; a value indicative of pulmonary hypertension (Marshall & Marshall 1991).

There have been only a few studies investigating pulmonary hemodynamics while exercising at high intensities. In Operation Everest II, mean pulmonary arterial pressures were measured by right heart catheterization in 8 formerly competitive endurance athletes (Reeves et al., 1987). Subjects had a mean $\dot{V}O_2max$ of 51.3 $ml \cdot kg^{-1} \cdot min^{-1}$ in a barometric chamber simulating sea level. Additional measurements were performed at 6100 m and 7620 m-altitudes equivalent to climbing Mount Everest. Mean pulmonary arterial pressure at maximum exercise (236 watts) was 29 ± 3 mmHg. Three subjects had pulmonary pressures greater than 40 mmHg. As the barometric pressure decreased simulating the higher altitude, pressures further increased. The magnitude of these pressures is consistent with other researchers using catheter studies to measure pulmonary artery (Harms et al., 1998b) and wedge pressures (Wagner et al., 1986). Wagner et al., (1986) exercised 8 subjects to a $\dot{V}O_2max$ of 3.7 $l \cdot min^{-1}$ in an upright position and pulmonary arterial and wedge pressures at 90% of $\dot{V}O_2max$ were found to be 37.2 ± 6.1 mmHg and 21.0 ± 3.7 mmHg respectively. Maximum workloads reached only 271 watts.

To date, pulmonary capillary pressures have not been directly measured in exercising humans or experimental animals. However, based on micropuncture studies in anesthetized cats, it is estimated that capillary pressures are likely half way between pulmonary artery pressures and pulmonary artery wedge pressures (Bhattacharya et al., 1982). Following this reasoning, West & Mathieu-Costello (1992a) estimated capillary pressures in these subjects to be 29 mmHg, using the reported pulmonary artery and wedge pressures by Wagner et al., (1986) (37.2 and 21 mmHg respectively). Furthermore, pulmonary pressures are observed in the middle region of the lung. Due to a hydrostatic gradient, vascular pressures at the base of the lung are calculated to be at least 7 mmHg greater. Thus capillary pressures in the base of the lung in the aforementioned study may exceed 36 mmHg. Indeed pulmonary capillary pressure at high blood flow in dogs is closer to arterial than venous pressures and therefore the pressure of 36 mmHg may be an underestimate (Younes et al., 1987).

The consequences of these raised pressures during exercise are uncertain. It has been proposed that these high estimated pulmonary arterial pressures could result in 1) structural stress failure of the blood-gas barrier, 2) an inflammatory reaction and increased membrane permeability secondary to stress failure and/or 3) increased fluid filtration (altered Starlings forces) combined with an inadequate removal by the lymphatic system producing pulmonary edema.

Stress Failure of the Blood-gas Barrier

The maintenance of the integrity of the blood-gas barrier is critical for efficient gas exchange. High pulmonary pressures seen during heavy exercise may result in damage to the pulmonary capillaries and thus alter the integrity of the blood-gas barrier. To permit rapid CO_2 and O_2 exchange by diffusion, the barrier between the alveoli and the capillaries must be extremely thin. In fact, some sections of the blood gas barrier in humans have been shown to be only 0.3 μm (Gehr et al., 1978). In contrast, this barrier must be strong enough to withstand the pressures associated with accepting the high cardiac outputs during intense exercise. Due to this "physiological dilemma" (West & Mathieu-Costello 1993), it seems reasonable to expect stress failure of the blood-gas barrier during heavy exercise. Consequently, regions of the lung would be potentially poorly ventilated, poorly perfused or both. In addition, another function of blood-gas barrier is to act as a barrier to prevent fluid from entering the interstitium. Damage to the membrane would permit fluid to enter the interstitial space and thus result in pulmonary interstitial edema.

As discussed previously, human pulmonary pressures seen at maximal exercise are often above 40 mmHg which is in the range shown to cause capillary stress failure in a rabbit lung model[1]. There has been some indirect evidence to support the theory of stress failure in humans including several anecdotal reports of human athletes who have coughed up blood or tasted blood after strenuous exercise (Weiler-Rarell et al., 1995). In addition, Hopkins et al., (1997) reported an increase in red blood cells and protein in

[1] As a caution, it cannot be assumed that stress failure occurs in humans and rabbits at the same pressures, as other studies show that stress failure in the dog and horse lung occur at much higher pressures than in the rabbit (Birks et al., 1994; Mathieu-Costello et al., 1995).

bronchoalveolar lavage fluid following 7 minutes of maximal exercise. This finding suggests failure of the blood-gas barrier. However, all of the subjects in this study had a history of blood in their lungs following intense exercise and it is conceivable that this finding could occur without an alteration to the blood-gas barrier. In a follow-up study, Hopkins et al., (1998c) had a similar group of athletes perform a submaximal exercise (77 % of $\dot{V}O_2max$) for one hour. Upon bronchoalveolar lavage, there was no evidence of RBC and protein indicating no or little damage to the membrane. There was an unusual and unexpected finding of the presence of RBC in the lavage fluid of control subjects, questioning the validity of this technique as the control subjects did not perform any exercise. Nevertheless, the authors concluded that it is the high pressures associated with maximal exercise that are necessary to alter the integrity of the blood-gas barrier.

Studies using animal models have provided valuable insight into the structure and function of the blood-gas barrier. Much of the evidence for stress failure is from experimental observations in anesthetized rabbits (West et al., 1991b). The rabbits had cannulas inserted into the pulmonary artery and left atrium to accurately measure transmural pressure. The lungs were perfused first with autologous blood and then fixed for electron microscopy.

Using this preparation, pressures of 39 mmHg caused disruptions to the capillary endothelium, basement membranes, and alveolar epithelium (West et al., 1991b). As demonstrated by West & Mathieu-Costello (1992b) frequent damage to the endothelium and/or epithelium occurred while the basement membrane remained intact. The interstitium increased in thickness and platelets and RBC appeared to be attached to the basement membrane when the endothelium and/or epithelium was damaged. Epithelial disruptions often occurred near the intercellular junctions but not at the junctions. At even higher pressures, red blood cells were occasionally seen passing through all layers of the membrane (West & Mathieu-Costello 1992b). No disruptions of the membrane occurred at pressures of 9 mmHg and few occurred at pressures of 24 mmHg (West et al., 1991b).

In a follow-up study, Kurdak et al. (1995) supports the two studies by Hopkins et al. (Hopkins et al., 1997; Hopkins et al., 1998c) in reference to the intensity of exercise and damage to the blood-gas barrier. Traditionally, studies investigating stress failure in the rabbit have artificially exposed pulmonary capillaries to high pressures for only short durations (approx. 5 minutes). However, Kurdak et al., (1995) perfused rabbit lungs with 26 mmHg for 10 and 100 minutes and found no statistical differences in the degree of membrane disruption. These results suggested that applying a moderate pressure for a longer duration does not increase the incidence of stress failure. Therefore stress failure seems to result from single insults of high pressure rather than multiple insults at moderate pressure. However, as the duration increased, the thickness of the blood-gas barrier also increased (Kurdak et al., 1995) which is consistent with the theory of interstitial edema. Addition studies are necessary in humans to resolve this issue.

High lung volumes may also result in structural damage to epithelial cells of the blood-gas barrier. When anesthetized rabbits were exposed to high lung volumes while pulmonary capillary pressure remained constant, there was a great increase in the frequency of capillary stress failure (Fu et al., 1992). In addition, at higher lung volumes and constant capillary pressures of 32.5 $cm{\cdot}H_2O$, the thickness of the blood-gas barrier was greater; indicative of interstitial edema. However in humans performing maximal exercise, tidal volumes only reach approximately 50% of vital capacity (Dempsey et al., 1996), suggesting that high lung volumes are not associated with heavy exercise.

As previously discussed, it is unclear if these artificially induced pressures, which result in altered blood-gas barrier permeability, are in the "physiological range" that occur during exercise. Guery et al., (1998) increased pulmonary artery pressure or alveolar pressure to clinically relevant levels in the rat lung model and found no change in the permeability of the alveolar-capillary membrane. Permeability was assessed by measuring fluorolabeled dextran in broncholavage fluid and measuring extravascular lung water as well as electron microscopy to measure structural integrity. However, the authors did find that simultaneously increasing both airway and capillary pressures increased the degree of blood-gas barrier disruption.

Other than humans, Thoroughbred racehorses, Greyhound dogs and racing camels have shown evidence of stress failure following heavy exercise (Akbar et al., 1994; King et al., 1990; Whitwell & Greet 1984). Thoroughbred racehorses have, for example, been selectively bred to have extremely high aerobic capacities. This high $\dot{V}O_2max$ is dependent on a high cardiac output. Pulmonary arterial pressures in these athletic animals have been measured to exceed 100 mmHg (West et al., 1993). Horses have higher pulmonary arterial pressures than humans even when corrected for body weight (Dempsey et al., 1996). Almost all thoroughbred horses performing heavy exercise hemorrhage into their lungs (Whitwell & Greet 1984) and show blood-gas barrier disruption upon electron microscopy (West et al., 1993). The Shetland Pony has much lower pulmonary pressures but also shows disruptions in the blood-gas barrier with intense exercise (Erickson et al., 1997).

There is some evidence to suggest that endothelial damage is transient and possibly reversible. Using the rabbit lung model, Elliott et al., (1992) demonstrated that reducing pressures during perfusion decreased the magnitude and the number of disruptions in the epithelial and endothelial membranes. The disruptions which disappeared were initially small and associated with intact basement membranes. Other studies have shown disruptions in the membranes at raised pressures and in more than 50 % of these disruptions, the basement membranes remained intact (West et al., 1991b). This has lead to the suggestion that once pressures return to normal levels, the cells may travel along their basement membrane and close the gap (West et al., 1991b). In addition, there is some evidence to suggest that vascular remodeling occurs in response to chronically elevated pulmonary pressures. For example, patients with mitrial stenosis have thickened basement membranes (West & Mathieu-Costello 1998). A useful study would be to assess and compare the hemodynamic response to heavy exercise in individuals with EIH and those without.

In conclusion, stress failure appears to be a viable mechanism for the development of EIH. The high pulmonary pressures with heavy exercise combined with the thin membranes makes the blood-gas barrier particularly vulnerable to stress injury. The mechanism of stress failure has been identified in other human pathological conditions

including high-altitude pulmonary edema, severe left ventricular failure, mitral stenosis and neurogenic pulmonary edema.

Stress Failure and Altered Permeability

It has been recently proposed that an inflammatory response may occur secondarily to stress failure (Durand et al., 1999) and further impair gas exchange. EIH may be caused by an increased histamine release as a result of an inflammatory response near the pulmonary capillaries. Such a response may alter the permeability of the membrane. Using a radiolabled aerosol to determine lung clearance Braude et al., (1984) found that inhaled histamine increased lung epithelial permeability. Anselme et al., (1994) demonstrated a significant relationship between the change in plasma histamine and the change in PO_2 with progressive exercise ($r = 0.8$, $p < 0.01$) in seven young and seven Masters athletes. However, the authors conceded that it could not be determined if the histamine response caused the decrease in PO_2 or was simply a response to injury. In an additional study, inhibition of the histamine response, through administration of nedocromil sodium, improved PO_2 at maximum exercise from 71.1 to 83.4 mmHg as well as significantly improved the A-aDO_2 gradient in seven Master athletes (Prefaut et al., 1997). However, the improvement in PO_2 did not alter $\dot{V}O_2max$. The authors proposed a link between EIH and histamine in Masters athletes.

Increased Filtration Pressures

As previously discussed, pulmonary vascular pressures increase with exercise intensity. The increase in pressure may alter the integrity of the blood-gas membrane either structurally or chemically, or simply increase Starlings forces. Increases in Starlings forces may increase the amount of fluid filtration and result in a mild subclinical edema.

Edema

Interstitial edema is fluid that accumulates between the epithelium of the alveolus and endothelium of the capillary. Since diffusion of blood gases is inversely proportional to thickness and the resultant edema increases membrane thickness, gas transport across the

membrane is impaired. An increased thickness need not be the only mechanism for altered gas exchange. Alternatively, alveolar edema may reduce lung compliance and compress capillaries which may exacerbate the $\dot{V}_A/\dot{Q}$ inequalities (Hopkins et al., 1998b). Evidence from the pig suggests that the when interstitial edema is evident, it surrounds the medium-sized airways and blood vessels (Schaffartzik et al., 1993). This constriction may alter the diameter of the airway, artery or both and lead to a $\dot{V}_A/\dot{Q}$ inequality.

Methods for Determination

There has been limited anecdotal or case study evidence of athletes who have experienced edema as the result of heavy exercise. In one case study, McKechnie et al., (1979) reported radiographic evidence suggesting that two athletes suffered from acute pulmonary edema during a 90-km running race. In another case study, Weiler-Rarell et al., (1995) reported severe dyspnea and hemoptysis in 8 swimmers during the first 45 minutes of a swimming event, forcing 5 of these athletes to retire early from the event. Upon hospitalization, two of these athletes had radiographic evidence of clinical edema.

Although it is proposed that a mild pulmonary edema may occur in some elite athletes, it has yet to be conclusively shown. To date, there is no acceptable measurement of subclinical edema. Several indirect methods have been used and include analysis of post exercise lung mechanics, post exercise diffusion capacity (DLCO) tests, $\dot{V}_A/\dot{Q}$ mismatch during and post exercise, radiographic tests, and scanning tests.

Lung mechanics following exercise

It has been speculated that rapid shallow breathing following heavy exercise may be indicative of interstitial pulmonary edema (Youngs & Burks 1985). Caillaud et al., (1993) found a change in tidal volume during exercise and recovery for the same ventilation level and concluded that interstitial edema may be responsible. This reduction in tidal volume also occurred in an untrained group but to a lesser extent. In addition, Schaffartzik et al., (1992) also found reductions in vital capacity following exercise

without concomitant changes in expiratory flow rates. These findings suggested early closing volume possibly due to subclinical pulmonary edema.

Post exercise diffusion capacity

Post exercise diffusion capacity (DLCO) tests have identified a reduction in DLCO following high intensity exercise. It is speculated that this decline in diffusion capacity is due to fluid accumulation in the interstitial space. However, similar declines in DLCO post exercise have also recently been shown to occur in untrained subjects where edema is not expected (Sheel et al., 1998). Significant declines in DLCO have also occurred following exercise at only 30 % of $\dot{V}O_2$max (Warren et al., 1999), an intensity where the formation of edema is highly unlikely. DLCO as an index for edema, has recently received criticism from other groups (Hanel et al., 1997; McKenzie et al., 1999a). In fact, it has been suggested that reduction in post exercise diffusion may be more reflective of peripheral blood pooling rather than fluid accumulation. Using regional electrical impedance, Hanel et al., (1997) showed that approximately one-half of the post-exercise reduction in DLCO can be explained by a decrease in pulmonary blood volume.

Ventilation/perfusion mismatch

As previously stated, ventilation/perfusion studies using MIGET have suggested that a diffusion limitation may explain 33 to 60 % of the A-aDO_2 gradient in humans (Hopkins et al., 1994; Torre-Bueno et al., 1985). There are three indirect lines of evidence that $\dot{V}_A/\dot{Q}$ may reflect edema in humans. $\dot{V}_A/\dot{Q}$ inequalities persist 30 minutes into recovery following intense exercise despite ventilatory parameters and cardiac output returning to baseline values (Schaffartzik et al., 1992). Secondly, breathing 100% oxygen improves $\dot{V}_A/\dot{Q}$ mismatch (Gale et al., 1985) as it is thought to decrease pulmonary arterial pressure and driving pressure for edema (Hopkins et al., 1998a). Indeed the addition of oxygen during inspiration also increases the $\dot{V}O_2$max in proportion to the EIH experienced during breathing room air (Harms et al., 1998a; Nielsen et al., 1998; Powers et al., 1989). Finally, $\dot{V}_A/\dot{Q}$ increases in a hypoxic environment (Wagner et al., 1987).

Radiographic tests

Gallagher et al., (1988) was unable to detect any evidence of pulmonary edema with human radiological studies. However, the sample population was not highly trained, ($\dot{V}O_2max$ = 3.07- 3.77 l/min) and was unlikely to develop pulmonary edema. Furthermore, radiographic X-rays can only detect clinical changes in edema greater than 20 percent. Fluid accumulation in trained individuals during heavy exercise may amount to only six percent.

Scanning studies

Computerized tomography (CT) scans in highly trained athletes after completing a triathlon found an increased lung density and an increased number of opacities (index of interstitial fluid accumulation) (Caillaud et al., 1995). These results suggested the existence of mild subclinical edema despite a post-triathlon scan time interval of 105-135 minutes. These authors also reported unpublished data of CT before and after a 13 minutes incremental, exhaustive exercise test and found non-significant results.

Recent work from our laboratory with magnetic resonance imaging (MRI) has found mixed results. Images following high intensity exercise, near $\dot{V}O_2max$, were unable to detect the presence of edema (McKenzie et al., 1996). However, in agreement with Caillaud et al., (1995), significant increases in edema were found after 45 minutes of cycling at 75 % of $\dot{V}O_2max$ (McKenzie et al., 1999b). In the later study using MRI, the subjects were highly trained however, there was no measurement of PO_2, which limits the applicability to EIH. As with DLCO, a limitation to the CT and MRI scanning studies is the difficulty of accounting for the increased blood volume in the pulmonary vasculature seen post exercise.

Animal models

After 6-7 minutes of heavy exercise, histological evidence of perivascular edema is shown in the pig (Schaffartzik et al., 1993). The authors found a significantly greater percentage of pulmonary arteries with perivascular edema in exercising pigs than in

nonexercising controls (33.8 vs. 20 %, respectively). The authors concluded that perivascular edema can occur following heavy, short term exercise.

Functional vs Structural Mechanism

Although there is some debate as to whether edema occurs in humans performing intense exercise, there is also debate as to whether the proposed edema is of sufficient magnitude to impair gas exchange. This issue has been investigated by having subjects perform two consecutive, high intensity exercise bouts while measuring SaO_2 (McKenzie et al., 1999a). In this study, SaO_2 remained unchanged while DLCO increased after a second bout of intense exercise suggesting that if edema was present it was not of sufficient magnitude to impair gas exchange and cause EIH.

In a similar study, 28 women performed an incremental $\dot{V}O_2max$ test followed by a constant-load treadmill test 20 minutes later. $\dot{V}O_2max$ and PO_2 were not altered by prior exercise (St. Croix et al., 1998). These authors speculated that the mechanism of EIH may be more functional than structural. These authors further argue that if edema occurs, it is very transient in nature. Any structural abnormality should theoretically require more than 20 minutes for repair. In addition, this study documented the incidence of EIH to be much higher in women than men. Women have smaller lung volumes, decreased airway diameter and decreased diffusion capacity compared to age and height matched men (McClaran et al., 1998).

Future Research Directions

Exercise-induced hypoxemia is a common occurrence in highly trained athletes yet the mechanisms responsible have not been clearly established. More research is needed to investigate the pulmonary hemodynamic response to heavy exercise, to investigate alternative methods for determining stress failure of the blood-gas barrier and finally, to confirm the incidence of EIH in women.

Appendix II. Literature review: Pulmonary clearance of Tc-99m DTPA, the blood-gas barrier and exercise.

Background

Pulmonary clearance of technetium-99m diethylene triamine penta-acetic acid (Tc-99m DTPA), a radioactive labeled aerosol, can be used in a clinical setting for investigating blood-gas barrier permeability. The technique is non-invasive, relatively inexpensive and available in most nuclear medicine departments (Smith et al., 1992). Pulmonary clearance of Tc-99m DTPA has been used for many years in nuclear medicine as a non-specific, but extremely sensitive method for detecting lung pathology (O'Doherty & Peters 1997).

Pulmonary clearance involves the deposition of an aerosol, small droplets of an isotonic solution containing a radio-labeled particle, on the surface of the pulmonary epithelium. Following inhalation, the rate at which Tc-99m DTPA diffuses across the alveolar-capillary membrane over time can be measured with a gamma camera. This rate is used as an index of alveolar-capillary permeability (Coates & O'Brodovich 1986). Once Tc-99m DTPA is in the capillary blood, it is distributed uniformly within the total body intra-vascular fluid, filtered by the kidneys and ultimately excreted in the urine.

Aerosol generation and delivery

The aerosol is generated by introducing approximately 20-30 mCi (740-1110 MBq) of Tc-99m DTPA in 2 ml of saline and driving it with compressed oxygen at a flow rate of 10 $l \cdot min^{-1}$. The aerosol produced is approximately 1 μ mass median aerodynamic diameter. It is recommended that the radiochemical purity of the aerosol be tested; free 99m-Pertechnetate (TcO_4) should not exceed 5 % of the composition (Coates et al., 1988).

Breathing procedures are rehearsed and subjects rest for 20 minutes prior to inhalation of the aerosol to avoid alterations in clearance rates from initial deep breathing and assure optimal delivery (Smith et al., 1992). Each subject is fit with a nose clip and mouth piece and instructed to breathe at normal tidal volumes for several minutes in a seated or supine position. The subject is then seated or supine with a large field of view gamma camera, positioned posteriorly and set to image the entire lungs.

Output from the gamma camera is sent to a computer for data processing. The decline in radioactivity over time is recorded by acquiring image frames. Pixel counts are taken and corrected for physical decay then plotted as function of time. The data are often fit to a mono-exponential function $N = N_o e^{-kt}$, where N_o is the y-intercept or count rate at $t = 0$, N is the count rate at any time t, k is the slope of the line or rate constant for clearance. If the curves are indeed mono-exponential, the rate constant (k), is determined over thirty minutes and expressed as a percentage of decreased radioactivity per minute (% min^{-1}). This time frame is recommended as inflammatory mediators (macrophages and neutrophils) release oxygen free radicals and are capable of splitting Tc from DTPA at 30 minutes (Braude et al., 1984). Clearance half-times (T½) are also frequently reported in the literature and can be easily derived from k values.

Pulmonary clearance, obtained by monoexponential analysis, in healthy, normal subjects is approximately 1 % min^{-1} or T½ of 70 minutes (O'Doherty & Peters 1997; Smith et al., 1992). Ninety-five percent of the population should have clearance rates slower than 1.5 % min^{-1} or a T½ of greater than 46 minutes (Smith et al., 1992). Of interest, clearance rates of 4 % min^{-1} or T½ of 20 minutes is common in smokers (O'Doherty & Peters 1997).

In severely damaged lungs, the clearance curve can often be bi-exponential. Bi-exponential curves can occur when there are regions in the lung with different clearance rates. An accumulation of fluid in the interstitium of some alveoli would likely result in a slower exponential curve. Alternatively, a slower exponential curve could indicate normal clearance and a faster curve could represent clearance from a damaged blood-gas membrane. Clinically, any curve that is bi-exponential is deemed abnormal (O'Doherty & Peters 1997).

Comparing results

It is often difficult to compare studies obtained in different laboratories, as the methodology is not often standardized. In addition, Several factors appear to influence pulmonary clearance rates including aerosol particle size, lung volumes and breathing frequency during the inhalation of the aerosol (Rizk et al., 1984), positive end-expiratory

pressure (Barrowcliffe et al., 1989; Marks et al., 1985), scanning position (Meignan et al., 1987), scanning time (Mason et al., 1997), and the method of analysis.

Correction factor

Background correction for re-circulation of radioactivity through the pulmonary capillaries is of controversial significance (Coates et al., 1988). Some studies (Lorino et al., 1989; Meignan et al., 1986) have ignored background correction as there may be considerable error in calculation and such background radioactivity does not appear to significantly effect the measured clearance rate (Meignan et al., 1986). In addition the rate at which the aerosol leaves the blood by normal kidney filtration is 10 fold faster than the aerosol enters the blood through the blood-gas barrier (Hilson et al., 1976). In contrast, Mason et al., (1997) recently demonstrated that intravenous DTPA administered prior to inhalation of the aerosol and using the liver for background correction, pulmonary clearance curves were multiexponential. However, using the thigh for background correction yielded pulmonary clearance curves that were monoexponential. Apparently, the liver has a closer extravascular-to-intravascular compartment ratio to the lung and therefore the authors concluded that liver background correction allows the true shape of the curve to be identified. Staub et al., (1990) reported that correction for re-circulation was less of a concern if clearance curves were calculated within the first 20-30 minutes.

Reliability

Present research reports this measure as being reproducible and capable of following a patient over the period of treatment (O'Doherty & Peters 1997). Previous studies determining pulmonary clearance of Tc-99m DTPA have calculated the intra-individual coefficients of variation between 7-18 % (Groth et al., 1989; Nolop et al., 1986; Smith et al., 1992; Thunberg et al., 1989). Coates et al., (1987) measured pulmonary clearance three times over a one month period in 40 healthy men and the correlation coefficient was 0.57 for the right lung and 0.37 for the left lung. The reason(s) for this poor reproducibility is not entirely clear. Some error in measurement could be the result of different methodological procedures used in different laboratories.

Radiation Doses

As discussed, the Tc-99m DTPA passes through the pulmonary blood-gas barrier into the blood and is then filtered by the kidneys and excreted in the urine. Whole-body effective dose equivalents are independent of clearance rates (Barber 1985). The organ with the highest radiation dose appears to be the bladder wall. Oral fluid administration and regular voiding can readily reduce the effective dose (Barber 1985).

Clinical applications

Pulmonary clearance has been reported to be an extremely sensitive and valuable technique for detecting and quantifying a wide range of lung injuries. Faster clearance rates have been observed with smoking, oxygen toxicity, sarciodosis, surfactant removal, adult respiratory distress syndrome, inhalation burns, pulmonary fibrosis, exercise, inhaled histamine, and HIV-positive patients with early pneumocystis pneumonia (O'Doherty & Peters 1997, Sundram 1995, Staub et al., 1990).

Delimitations

It should be emphasized that pulmonary clearance of Tc-99m DTPA is a non-specific measure of blood-gas barrier permeability and therefore the precise etiology of altered permeability cannot be ascertained.

Theoretical basis of pulmonary clearance

Properties

Tc-99m DTPA has a physical half-life of 6.04 hours. It has a molecular weight of 492 daltons and the stability constant for Tc-99m DTPA is 10^{17}.

Blood-gas Barrier

The blood-gas barrier consists of epithelial and endothelial cells with their fused basement membranes, separated by the interstitial space. Respiratory gases (O_2 and CO_2) as well as carbon monoxide are lipophilic and can readily pass through the membrane by

simple diffusion. These molecules are often blood flow dependent. However, Tc-99m DTPA is a hydrophilic aerosol. Hydrophilic molecules are normally limited to passive diffusion through the intercellular junctions of the alveolar epithelium and the capillary endothelium. These molecules are perfusion independent although complete occlusion of pulmonary blood flow alters clearance rates (Rizk et al., 1984). The intercellular gap junctions in the alveolar epithelium (zona occludentes) are much tighter (0.8 to 1.0 ηm) than the capillary endothelium intercellular junctions (4.0 to 8.0 ηm) (Taylor & Gaar 1970). Therefore, the alveolar epithelium is thought to be approximately 10 times less permeable than capillary endothelium (Gorin & Stewart 1979). Thus it is the alveolar epithelium which is thought to be the limiting membrane preventing movement of fluids and solvents as well as Tc-99m DTPA into the interstitial space.

Pulmonary clearance and exercise

Theory

The maintenance of the integrity of the blood-gas barrier is critical for efficient gas exchange. High pulmonary pressures seen during heavy exercise may result in damage to the pulmonary capillaries thus altered integrity of the blood-gas barrier. To permit gas exchange by diffusion, the barrier between the alveoli and the capillaries must be extremely thin for rapid exchange of CO_2 and O_2. In fact, some sections of the blood gas barrier in humans have been shown to be only 0.3 μm (Gehr et al., 1978). In contrast, this barrier must be strong enough to withstand the pressures associated with accepting the high cardiac outputs during intense exercise. Due to this "physiological dilemma" (West & Mathieu-Costello 1993), it seems reasonable to expect failure to the blood-gas barrier. Consequently, regions of the lung would be potentially poorly ventilated, poorly perfused or a combination of both. In addition, a primary function of blood-gas barrier is to act as a barrier and prevent fluid from entering the interstitium. Severe damage to the membrane would permit fluid to enter the interstitial space.

Human pulmonary pressures seen at maximal exercise are often above 40 mmHg which is in the range shown to cause capillary stress failure in a rabbit lung model. There has

been some indirect evidence to support the theory of stress failure in humans including several anecdotal reports of human athletes who have coughed up blood or tasted blood after strenuous exercise (Weiler-Rarell et al., 1995). In addition, Hopkins et al., (1997) reported an increase in red blood cells and protein in bronchoalveolar lavage fluid following 7 minutes of maximal exercise suggesting failure of the blood-gas barrier during heavy exercise.

Literature

To the author's knowledge, only two studies have investigated blood-gas barrier permeability with Tc-99m DTPA and exercise. In one study, seven nonsmoking, healthy subjects inhaled the aerosol and sat on a bicycle ergometer with their backs against a gamma camera for 27 minutes (Meignan et al., 1986). At minute 20, subjects cycled at 50 watts for seven minutes while clearance was measured. There were significant increases in apical clearances (3.40 min^{-1} vs. 1.82 min^{-1}, respectively, $p < 0.01$) while basal clearances were non significant ($p > 0.05$). The authors concluded that the increase in apical clearance was primarily caused by an increase in apical blood flow. Other researchers have demonstrated that Tc-99m DTPA is perfusion independent and therefore the increase in apical clearance was likely the result of increased surface area available for gas exchange rather than increased perfusion (Lorino et al., 1989).

In a second study, seven nonsmoking males ($\dot{V}O_2max = 53 \pm 3$ $ml \cdot min^{-1} \cdot kg^{-1}$) performed 75 minutes of constant load exercise on a treadmill at 75 % of $\dot{V}O_2max$. Approximately 25 minutes following the exercise session when heart rate, tidal volume and breathing frequency had returned to resting levels, pulmonary clearance was determined with Tc-99m DTPA. Total, apical, and basal clearance significantly increased post exercise ($p <$ 0.01, 0.05, and 0.05 respectively) compared with pre exercise values. The authors concluded that sustained exercise results in an alteration in the intercellular tight junctions, secondary to the mechanical effects of sustained high ventilation rates. Alternatively, increased permeability may have been the result of stress failure of the blood-gas barrier. However, based on $\dot{V}O_2max$ values, subjects in this study were not highly trained and therefore unlikely to encounter stress failure as pulmonary arterial

pressures do not reach exceedingly high values during submaximal exercise; even if the exercise is prolonged (Hopkins et al., 1998b). Furthermore, Hopkins et al., (1998c) found that prolonged exercise at submaximal intensities did not alter the integrity of the blood-gas barrier. However, it is conceivable that subjects in both of these studies (Hopkins et al., 1998c; Lorino et al., 1989) developed edema from altered Starlings forces without any structural damage to the blood-gas barrier.

There is some debate as to the effectiveness of determining pulmonary edema with pulmonary clearance. Pulmonary edema without a concomitant increase in permeability does not appear to effect clearance of Tc-99m DTPA. Mason et al., (1985) tested six patients with cardiogenic pulmonary edema and only one subject had clearance rates greater than healthy controls. In contrast, 11 of the 14 patients with noncardiogenic pulmonary edema, had significantly greater clearance rates (Mason et al., 1985). It is presently unknown if the subclinical pulmonary edema thought to occur in athletes performing exercise is related to other clinical disease processes and whether the proposed edema could alter pulmonary clearance rates.

Alternative causes of accelerated clearance

It is difficult to differentiate the individual roles of increased blood flow and increased ventilation during exercise on changes in pulmonary clearance. As discussed, theory and some data suggest pulmonary hemodynamics may be responsible for altered clearance rates. Alternatively, breathing high lung volumes or high ventilation rates may induce an alteration to the membrane.

Explanations for an increase in clearance rates, while breathing higher lung volumes, have recently been reviewed by Smith et al., (1992). Some of the more plausible factors include: 1) stretching the intercellular junctions 2) mechanical injury resulting in alteration of the integrity of the blood-gas barrier 3) increasing alveolar ventilation thus unfolding of lung surface and decreasing the distance required for diffusion and/or increasing surface area for diffusion, and 4) depletion of surfactant resulting in an increased clearance independent of any change in permeability or surface area.

Egan (1980) showed that increases in lung volume resulted in increased alveolar permeability as measured by the rate of efflux of several different radiolabeled solutes of known molecular size. The increased permeability was most likely from alterations to the size of the intercellular junctions. The increased size of the gap junctions from high lung volumes were not immediately reversible when lung volumes were decreased and suggested that the gap junctions (zona occludentes) may not be elastic. Given the similar size of the intercellular junctions and Tc-99m DTPA molecule, it seems logical that this mechanism may explain altered clearance rates.

The expansion in lung volume may not necessarily be associated with stretching of the alveolar walls (Gil et al., 1979). Alternatively, high lung volumes may result in structural damage (often in the form of breaks) to epithelial cells of the blood-gas barrier. When anesthetized rabbits were exposed to high lung volumes while pulmonary capillary pressure remained constant, there was a great increase in the frequency of capillary stress failure (Fu et al., 1992). In addition, at higher lung volumes and constant capillary pressures of 32.5 cmH_2O, the thickness of the blood-gas barrier was greater; indicative of interstitial edema.

Furthermore, it is also conceivable that increases in pulmonary clearance from high lung volumes may be due to an increase in alveolar surface area from alveolar distention. This hypothesis is supported by Meignan et al., (1987) who found that subjects measured in a seated position had an increased clearance rate in the upper lung regions compared to the lower regions of the lung. Due to gravitational effects on transpleural pressures, upper lung regions have greater alveolar ventilation.

Surfactant depletion by lung lavage has been shown to increase pulmonary clearance (Evander et al., 1987). Surfactant is lipophilic whereas Tc-99m DTPA is hydrophilic. The interaction of these molecules may prevent a wide distribution of Tc-99m DTPA in the alveoli. Mechanical effects of increased ventilation may alter surfactant production (Lorino et al., 1989). However, it seems unlikely that exercise could alter surfactant levels and hence influence clearance rates.

All of these theories can be supported by investigations involving pulmonary clearance tests with positive end-expiratory pressure (PEEP). PEEP of 10-15 cmH_2O increased pulmonary clearance rates (Barrowcliffe et al., 1989, Marks et al., 1985, Rizk et al., 1984). However, the applicability of these theories to an exercise model is uncertain. It is uncertain if exercise may induce periodic increases of lung over-inflation, thus altering the integrity of the epithelium, and result in a faster diffusion of Tc-99m DTPA.

During maximal exercise tidal volumes only reach approximately 50% of vital capacity (Dempsey et al., 1996). These physiological tidal volumes during heavy exercise are much less than the mechanically ventilated volumes used in clinical research studies. After approximately 70% of $\dot{V}O_2max$ the increased minute ventilation with progressive exercise is the result of increased respiratory rate (Steinacker et al., 1993). Breathing frequency does not appear to influence pulmonary clearance. Rizk et al., (1984) studied this hypothesis in two groups of mechanically ventilated dogs. One group was assigned a respiratory rate of 25 breaths·min^{-1} with a tidal volume of 10 ml·kg^{-1} and the other group was assigned a respiratory rate of 15 breaths·min^{-1} with a tidal volume of 15 ml·kg^{-1}. There was no significant effect of increasing respiratory rate while performing a pulmonary clearance test. In contrast, dogs were mechanically ventilated with high-frequency oscillatory ventilation (HFOV) for four hours (Man et al., 1987). (HFOV is provides gas exchange using high-frequency small volume oscillations). Pulmonary clearance was measured 24 minutes later and it was discovered that HFOV significantly increased pulmonary clearance. The authors concluded that HFOV may damage the intercellular junctions. However the relevance of the later study to exercise in humans is questionable. The effect of hyperventilation (without exercise) prior to pulmonary clearance requires further investigation.

Conclusion

In conclusion, pulmonary clearance with Tc-99m DTPA has proven to be valuable in detecting certain lung pathologies however, there are several limitations with this technique. To date, it is uncertain if the integrity of the blood-gas barrier is altered in highly trained athletes performing heavy exercise and if pulmonary clearance with Tc-

99m DTPA will be able to detect this possible altered permeability. Further research is necessary to address these issues.

Appendix III. Raw Data

Table 3. *Age, height and mass for individual subjects.*

Subject	Age, yrs	Height, cm	Mass, kg
BT	25	179.4	77.2
CM	28	176.5	72.3
GM	23	180.5	72.6
CH	24	187.7	87.4
JP	21	185.9	75.7
SK	28	183.2	103.0
MV	23	174.5	74.5
PL	28	179.0	74.1
DT	25	180.1	84.7
RR	27	184.8	80.3
NS	24	182.2	79.6

Table 4. *Pulmonary function parameters for individual subjects.*

Subject	FVC, liters	FEV_1, liters	FEV_1/FVC, %	FEFmax, l/sec
BT	6.4	5.3	82.9	8.9
CM	5.2	4.8	93.2	11.8
GM	5.3	4.8	91.8	11.1
CH	7.1	5.9	82.6	9.9
JP	6.5	5.3	81.6	12.1
SK	6.1	5.0	80.6	11.8
MV	6.2	4.9	79.2	11.7
PL	5.3	4.4	83.8	10.5
DT	5.7	4.4	76.3	9.1
RR	5.8	4.5	76.9	8.9
NS	5.3	4.1	76.8	9.9

FVC, forced vital capacity; $FEV_{1.0}$, expiratory volume in 1 sec; MFEF, maximum forced expiratory flow.

Table 5. *Ventilatory parameters at maximum exercise for individual subjects.*

Subject	$\dot{V}E$, liters	$\dot{V}CO_2$, liters	$\dot{V}E/\dot{V}CO_2$,	$\dot{V}E/\dot{V}O_2$,	RER
BT	188.0	6.61	28.4	36.1	1.27
CM	196.0	5.96	32.9	41.5	1.26
GM	223.0	5.87	38.0	43.9	1.16
CH	225.0	6.98	32.2	41.3	1.28
JP	238.0	6.37	37.4	41.3	1.15
SK	175.0	6.70	26.1	31.2	1.19
MV	207.0	6.76	30.6	36.9	1.21
PL	195.0	6.40	30.5	40.5	1.33
DT	198.0	6.27	31.6	38.9	1.23
RR	192.7	6.98	27.6	33.3	1.21
NS	170.0	6.20	27.4	33.2	1.21

$\dot{V}E$, minute ventilation; $\dot{V}CO_2$, maximum CO_2 production; ratio; $\dot{V}E/\dot{V}O_2$ and $\dot{V}E/\dot{V}CO_2$, ventilatory equivalents for O_2 and CO_2 respectively; RER, respiratory exchange.

Table 6. *Performance parameters at maximal exercise for individual subjects.*

Subject	HR, bts/min	$\dot{V}O_2max$, liters/min	$\dot{V}O_2max$, ml/kg/min	Power, watts
BT	188	5.21	67.5	479
CM	174	4.73	65.4	431
GM	193	5.08	70.0	452
CH	199	5.45	62.3	512
JP	192	5.77	76.2	519
SK	170	5.61	54.5	501
MV	177	5.61	75.3	512
PL	198	4.82	65.0	457
DT	173	5.09	60.1	450
RR	186	5.78	72.0	520
NS	192	5.12	64.3	466

HR, heart rate; $\dot{V}O_2max$, maximum O_2 consumption.

Table 7. *Individual time course data for core temperature measured during the progressive exercise test. Following sample taken before the start of exercise, the first blood sample was withdrawn at minute 4.*

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	35.0	36.4	36.1	36.4	36.6	36.7	36.5	35.6	36.6	36.7	None
4	35.0	36.4	36.4	36.3	36.6	36.7	36.0	36.0	36.6	36.7	
5	36.0	36.5	36.4	36.5	36.0	36.7	35.8	36.2	36.6	36.6	
6	35.8	36.6	36.8	36.8	36.6	36.8	36.0	36.3	36.8	36.8	
7	35.5	36.7	36.8	36.8	36.5	36.9	36.0	36.3	36.6	37.0	
8	35.8	36.7	36.9	37.0	36.9	36.9	35.8	36.5	36.9	37.2	
9	36.5	36.8	36.9	37.0	36.8	36.9	36.5	36.6	37.2	37.4	
10	36.8	37.0	37.1	37.2	37.0	37.1	36.9	37.1	37.2	37.6	
11	36.8	37.0	37.3	37.2	37.1	37.0	36.7	37.0	37.4	37.7	
12	36.8	37.4	37.4	37.4	37.2	37.1	37.0	37.3	37.6	37.8	
13	37.0	37.5	37.5	37.5	37.2	37.1	37.0	37.5	37.5	38.1	
14	37.2	37.6	37.8	37.7	37.2	37.3	37.1	37.7	37.6	38.3	
15	37.4	37.8	37.6	37.8	37.3	37.5	37.3	37.7	37.8	38.4	
16	37.6			37.8	37.6	37.5	37.5			38.2	
17				37.8	37.8	37.8	37.6				
18					37.7						

X denotes no temperature recorded.

Table 8. *Individual time course data for corrected arterial partial pressure of oxygen measured during a progressive exercise test. Following the sample taken before the start of exercise, the first blood sample was withdrawn at minute 4.*

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	111	117	109	111	111	118	121	108	114	123	none
4	111	104	108	102	107	99	103	107	106	107	
5	102	101	95	101	100	99	99	100	102	102	
6	103	102	109	95	92	98	105	103	98	97	
7	100	98	98	93	94	88	91	96	96	94	
8	100	88	102	91	96	85	87	94	96	91	
9	101	88	92	91	92	83	86	96	95	89	
10	87	86	93	91	92	81	81	96	92	83	
11	88	83	113	88	93	78	80	88	90	83	
12	88	85	100	88	93	78	78	87	93	82	
13	86	91	93	86	92	74	75	92	X	79	
14	87	94	X	85	92	74	73	X	90	78	
15	88	100	X	87	96	77	73	91	91	79	
16	90			86	92	76	75				
17				89	92	76	73				
18					89						

PaO_2, arterial partial pressure of oxygen. X denotes no sample analyzed; often due to sample clotting.

Table 9. *Individual time course data for corrected arterial partial pressure of carbon dioxide measured during the progressive exercise test. See Table 8 for detailed description of layout.*

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	36	32	36	35	36	36	32	43	36	34	None
4	33	38	36	42	35	40	35	44	38	38	
5	41	39	36	42	36	39	36	47	38	39	
6	41	39	33	43	38	41	36	45	40	40	
7	43	39	35	42	38	41	38	47	39	42	
8	44	40	33	43	38	42	38	45	39	42	
9	44	40	34	42	37	42	38	44	39	42	
10	44	38	33	41	37	41	39	42	39	43	
11	45	31	33	41	37	42	37	43	40	43	
12	44	29	33	40	36	41	36	43	39	40	
13	43	26	33	38	35	41	35	41	X	41	
14	40	28	X	36	34	43	36	X	38	41	
15	40	24	X	35	32	39	35	38	36	40	
16	38			33	32	39	34			41	
17				32	29	38	33				
18					33						

PCO_2, arterial partial pressure of carbon dioxide. X denotes no sample analyzed; often due to sample clotting.

Table 10. *Individual time course data for the alveolar oxygen determined during the progressive exercise test. See Table 8 for detailed description of layout.*

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	116	117	122	122	125	127	118	111	124	120	None
4	115	111	120	113	120	115	114	107	116	110	
5	109	110	119	114	116	118	114	106	118	113	
6	109	112	122	114	114	117	117	108	113	112	
7	110	113	123	115	116	115	113	107	117	112	
8	110	113	124	116	117	114	116	112	118	113	
9	112	114	123	116	118	115	117	115	119	116	
10	111	119	124	119	120	116	116	117	120	113	
11	113	126	124	119	120	116	119	118	120	116	
12	115	129	127	122	122	118	122	121	122	120	
13	117	133	128	125	124	119	124	123	X	120	
14	121	133	X	127	126	120	123	X	126	122	
15	123	135	X	130	129	124	125	126	130	124	
16	127			131	130	126	127			126	
17				132	132	127	129				
18					129						

PAO_2, alveolar partial pressure of oxygen X denotes no sample analyzed; often due to sample clotting.

Table 11. *Individual time course data for the alveolar-arterial oxygen gradient determined during the progressive exercise test. See Table 8 for detailed description of layout.*

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	5	0	13	11	14	9	-3	3	10	-3	None
4	4	7	12	11	13	16	11	0	10	3	
5	7	9	24	13	16	19	15	6	16	11	
6	6	10	13	19	22	19	12	5	15	15	
7	10	15	25	22	22	27	22	11	21	18	
8	10	25	22	25	21	29	29	18	22	22	
9	11	26	31	25	26	32	31	19	25	27	
10	24	33	31	28	28	36	35	21	27	30	
11	25	42	11	31	27	37	39	30	31	33	
12	27	43	27	34	29	41	43	34	29	37	
13	31	42	34	39	31	45	48	31	X	41	
14	34	39	X	42	34	46	50	X	37	44	
15	35	34	X	43	33	48	52	35	39	44	
16	37			45	38	50	52			42	
17				43	40	51	56				
18					39						

$A\text{-}aO_2$, alveolar partial pressure of oxygen. X denotes no sample analyzed; often due to sample clotting.

Table 12. *Individual time course data for arterial oxygen saturation calculated from PaO_2 during the progressive exercise test. See Table 8 for detailed description of layout.*

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	98.2	98.4	98.0	98.2	98.1	98.4	98.5	97.9	98.2	98.5	None
4	98.2	97.8	98.0	97.7	98.0	97.5	97.8	97.9	97.9	97.9	
5	97.6	97.6	97.3	97.7	97.7	97.6	97.6	96.7	97.7	97.7	
6	97.7	97.6	98.1	97.3	97.2	97.4	97.8	97.6	97.4	97.4	
7	97.5	97.4	97.6	97.2	97.3	96.7	96.9	97.0	97.3	97.1	
8	97.4	97.0	97.8	96.9	97.5	96.3	96.6	97.0	97.3	96.8	
9	97.5	96.0	97.2	97.0	97.2	96.1	96.5	97.1	96.9	96.3	
10	96.3	96.0	97.0	96.9	97.2	95.7	95.8	97.0	96.8	95.5	
11	96.4	96.0	98.1	96.3	97.1	95.3	95.8	96.2	96.3	95.4	
12	96.2	96.0	97.4	96.2	97.1	95.0	95.6	95.6	96.3	94.9	
13	95.9	96.0	96.8	95.9	97.0	94.1	95.1	95.8	X	93.8	
14	95.9	96.0	X	95.5	97.0	93.7	94.2	X	95.3	92.3	
15	95.5	96.0	X	95.1	97.1	93.6	93.9	94.4	94.7	91.6	
16	95.2			94.4	96.3	92.6	93.7			92.0	
17				93.3	95.9	90.6	92.1				
18					95.2						

% SaO_2, arterial hemoglobin saturation. X denotes no sample analyzed; often due to sample clotting.

Table 13. Individual time course data for arterial oxygen saturation measured by an oximeter during the progressive exercise test.

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	99.1	None	None	96.1	97.2	98.2	98.1	None	97.4	97.3	98.1
4	X			95.9	96.7	97.2	97.0		97.0	97.0	97.0
5	X			95.9	95.9	97.2	96.9		97.1	96.8	96.9
6	X			95.5	96.1	97.0	96.5		96.6	96.6	96.5
7	X			95.4	96.0	96.6	95.9		96.7	96.3	95.9
8	X			95.3	96.0	96.4	95.8		96.4	96.2	95.8
9	X			94.8	95.9	95.9	95.4		96.4	95.5	95.4
10	X			94.8	95.9	95.4	94.8		96.2	94.7	94.8
11	96.3			94.2	95.8	95.5	94.8		95.9	94.9	94.8
12	95.8			94.0	95.3	94.9	94.7		95.5	94.0	94.7
13	96.5			94.0	95.5	94.5	94.2		95.2	93.2	94.2
14	95.6			93.6	93.9	94.2	93.8		94.9	91.7	93.0
15	95.2			93.0	95.5	93.9	93.7		94.5	90.7	92.0
16	94.8			92.4	95.1	93.3	92.8			88.3	91.0
17				91.2	94.4	91.5	91.9				90.5
18					93.9						

% SaO_2, arterial hemoglobin saturation. X denotes no measurement.

Table 14. *Individual time course data for pH measured during the progressive exercise test. See Table 8 for detailed description of layout.*

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	7.43	7.43	7.40	7.44	7.40	7.43	7.43	7.41	7.40	7.42	None
4	7.41	7.40	7.41	7.41	7.43	7.40	7.41	7.40	7.40	7.41	
5	7.40	7.39	7.41	7.42	7.43	7.41	7.41	7.38	7.39	7.41	
6	7.41	7.39	7.42	7.41	7.42	7.40	7.40	7.38	7.39	7.40	
7	7.40	7.39	7.42	7.42	7.43	7.40	7.40	7.37	7.39	7.40	
8	7.39	7.37	7.42	7.40	7.43	7.40	7.39	7.38	7.38	7.40	
9	7.39	7.37	7.42	7.41	7.43	7.40	7.40	7.37	7.38	7.39	
10	7.37	7.37	7.42	7.40	7.43	7.39	7.39	7.36	7.37	7.37	
11	7.37	7.35	7.42	7.37	7.42	7.38	7.39	7.34	7.36	7.37	
12	7.35	7.33	7.40	7.38	7.41	7.37	7.40	7.32	7.33	7.35	
13	7.34	7.31	7.38	7.37	7.40	7.35	7.39	7.29	X	7.33	
14	7.33	7.28	X	7.35	7.39	7.34	7.37	X	7.27	7.28	
15	7.30	7.23	X	7.31	7.38	7.31	7.36	7.19	7.21	7.22	
16	7.26			7.26	7.34	7.26	7.33			7.18	
17				7.15	7.30	7.19	7.28				
18					7.27						

X denotes no sample analyzed; often due to sample clotting.

Table 15. *Individual time course data for bicarbonate concentration measured during the progressive exercise test. See Table 8 for detailed description of layout.*

Time	BT	CM	GM	CH	JP	SK	MV	PL	DT	NS	RR
0	24	21	22	24	23	24	22	27	23	22	None
4	21	24	22	27	23	25	22	27	24	24	
5	25	24	23	27	23	25	23	28	23	24	
6	26	23	21	27	24	25	23	27	24	25	
7	27	23	23	27	25	25	23	27	24	26	
8	26	23	22	27	25	26	23	26	24	25	
9	26	23	22	26	24	26	23	26	23	25	
10	26	22	22	25	24	25	23	23	23	25	
11	26	17	22	24	24	25	23	23	22	24	
12	24	15	20	23	23	24	22	22	20	21	
13	23	13	19	21	22	23	21	19	X	20	
14	21	13	X	19	20	22	21	X	17	19	
15	19	10	X	17	19	19	20	14	14	16	
16	16			14	17	17	17			14	
17				11	14	14	15				
18					15						

HCO^{3-}, arterial bicarbonate concentration. X denotes no sample analyzed; often due to sample clotting.

Table 16. *Individual heart data during rest and post-exercise pulmonary clearance test. Time period between completion of* $\dot{V}O_2max$ *test and start of aerosol delivery.*

Subject	Rest HR, bts/min	Post-exercise HR, bts/min	Time period, min
BT	56	68	30
CM	52	62	30
GM	53	65	40
CH	56	90	35
JP	59	75	50
SK	48	69	35
MV	58	62	40
PL	60	85	40
DT	X	X	50
RR	69	75	50
NS	75	90	30

HR, heart rate; X denotes no measurement.

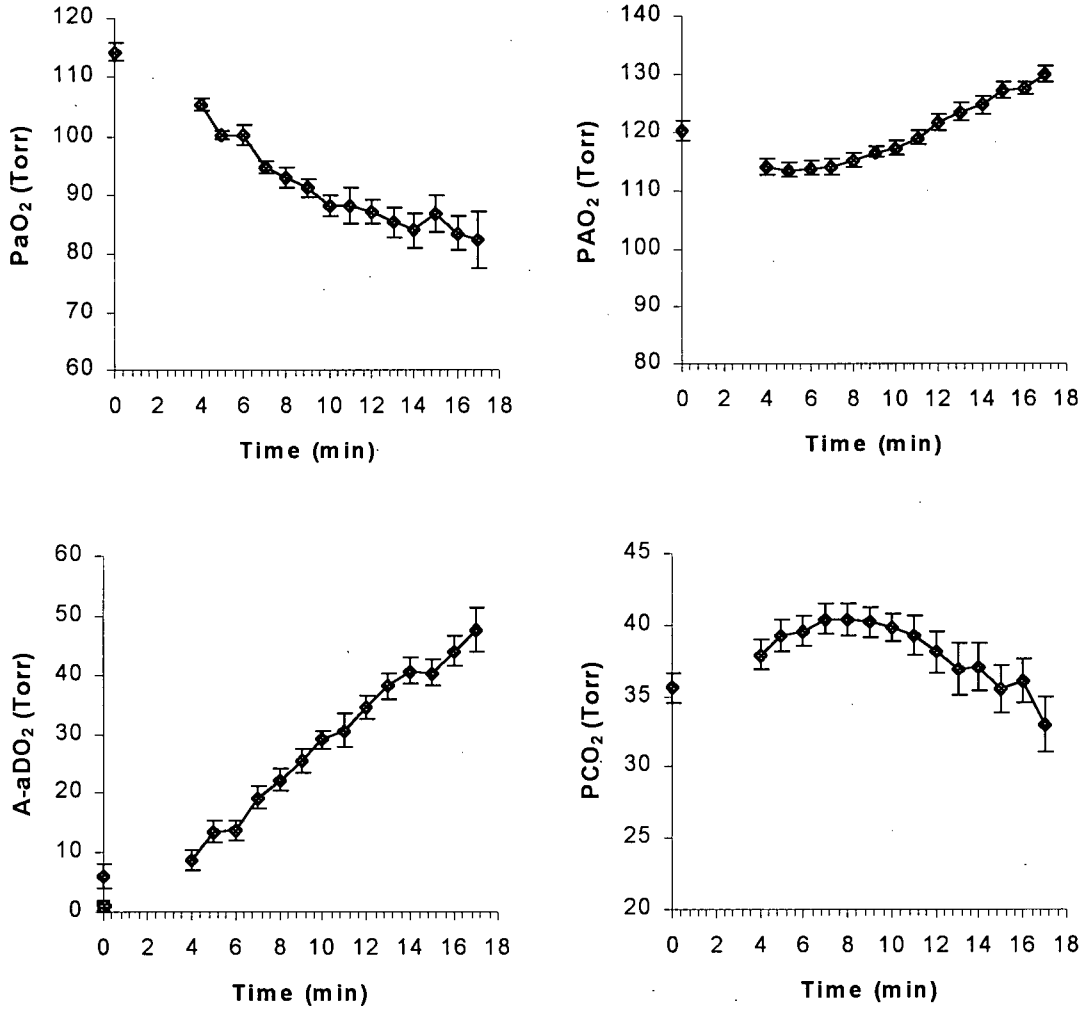

Figure 5. Mean ($n = 10$) time course data for arterial partial pressure of oxygen (PaO_2), alveolar partial pressure oxygen (PAO_2), alveolar-arterial oxygen gradient ($A\text{-}aDO_2$), and arterial partial pressure of carbon dioxide (PCO_2) during the progressive exercise test. Error bars represent SE.

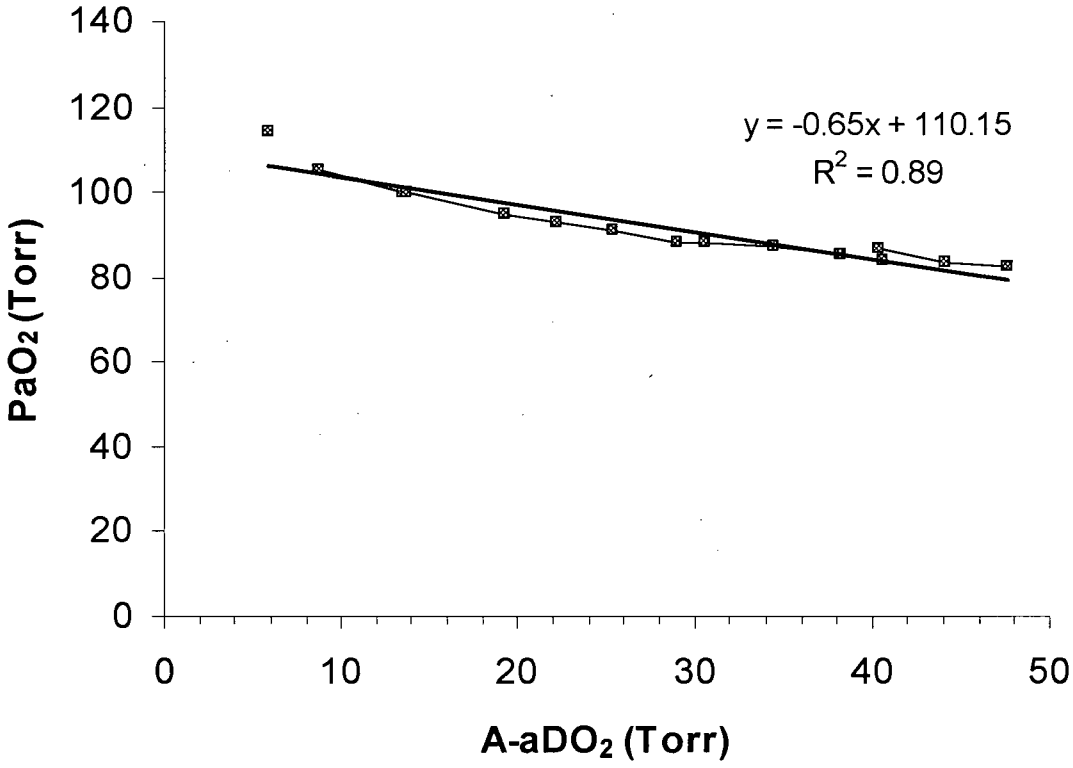

Figure 6. Relationship between PaO_2 and A-aDO_2. Linear regression line and equation is noted.

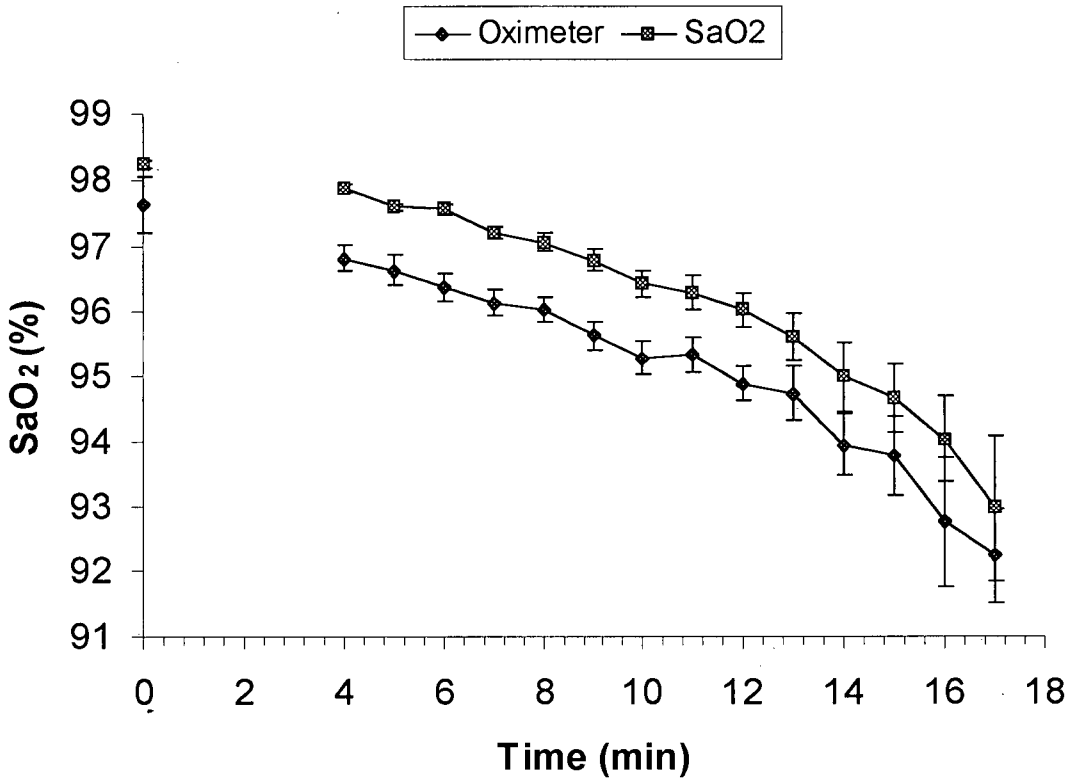

Figure 7. Percent arterial hemoglobin saturation (SaO_2) determined by oximeter and calculated by arterial blood gas data. Error bars represent standard error. Data from 7 of 11 subjects.

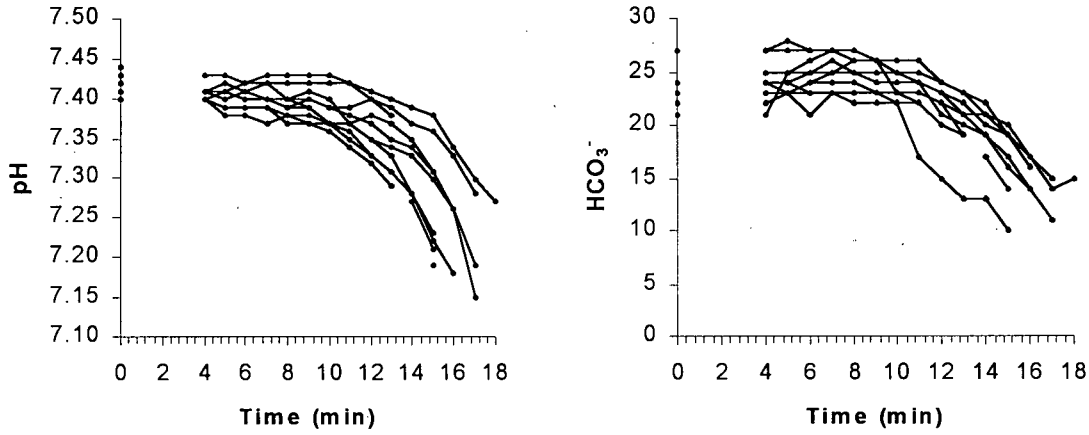

Figure 8. Individual time course data for arterial pH (*left*) and bicarbonate (HCO_3^-) (*right*) during the progressive exercise test.

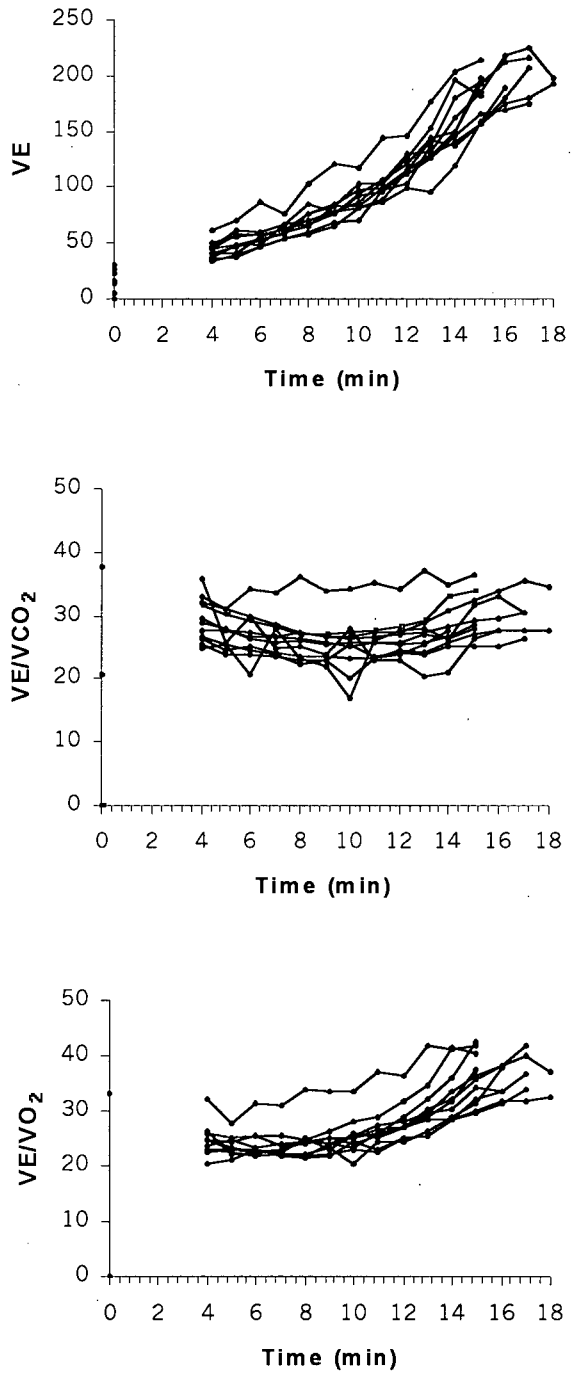

Figure 9. Individual time course data for minute ventilation ($\dot{V}E$) (*Top*), and ventilatory equivalents for oxygen ($\dot{V}E / \dot{V}O_2$) (*middle*) and carbon dioxide $\dot{V}E / \dot{V}CO_2$ (*bottom*).

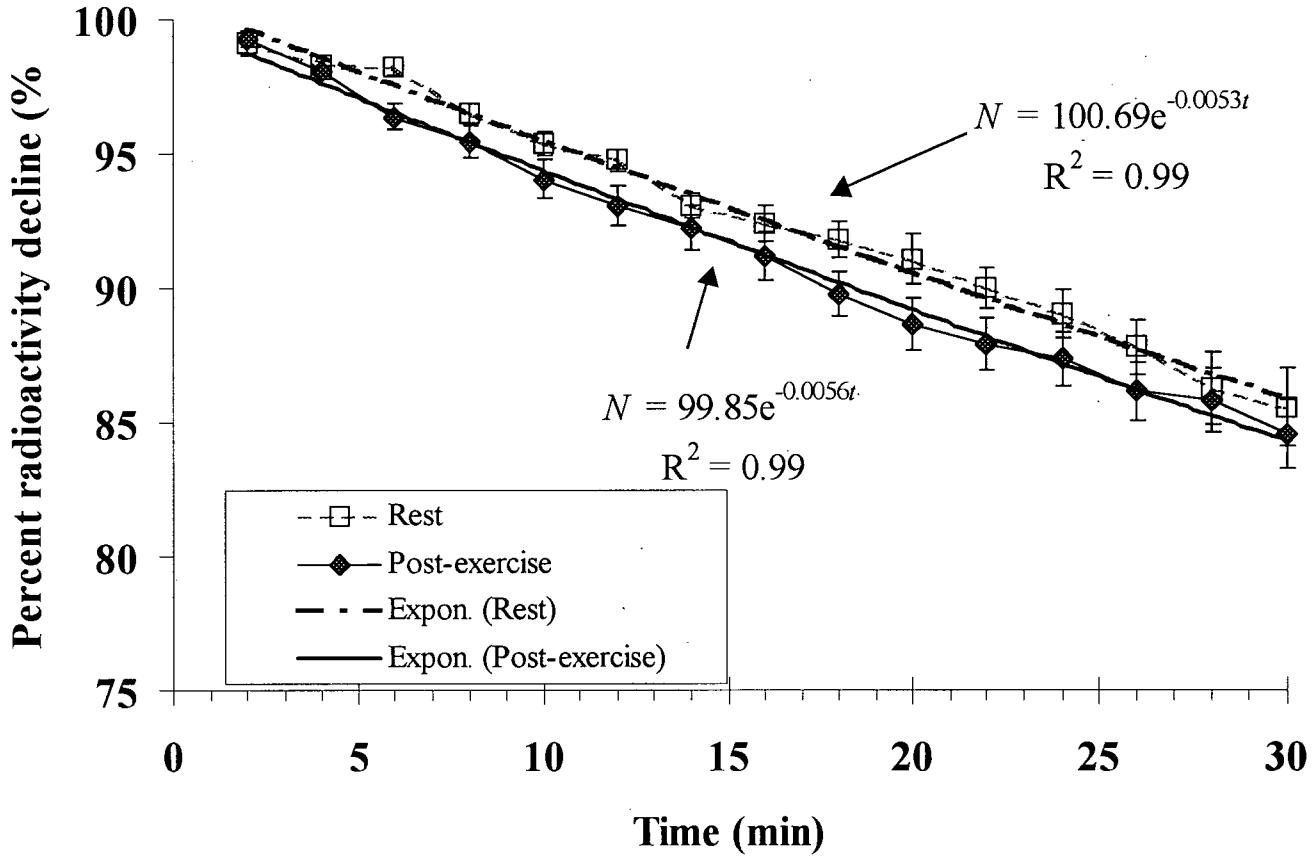

Figure 10. Mean ($n = 11$) for total (k_T) pulmonary clearance values acquired for 30 minutes, at rest and post $\dot{V}O_2max$. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period. Exponential regression lines and equations for rest and post-exercise are indicated.

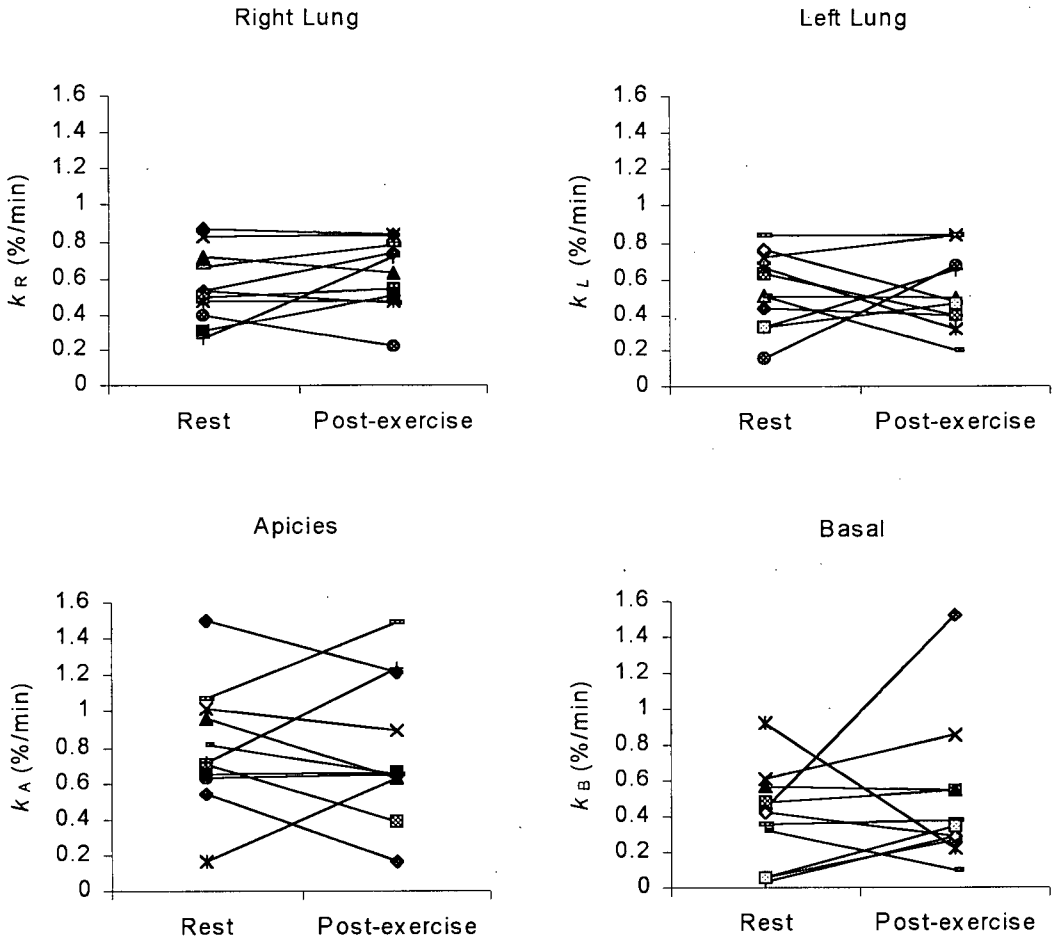

Figure 11. Individual pulmonary clearance rates ($n = 11$) for the right (k_R), left (k_L), apical (k_A), and basal (k_B) regions at rest and post-exercise.

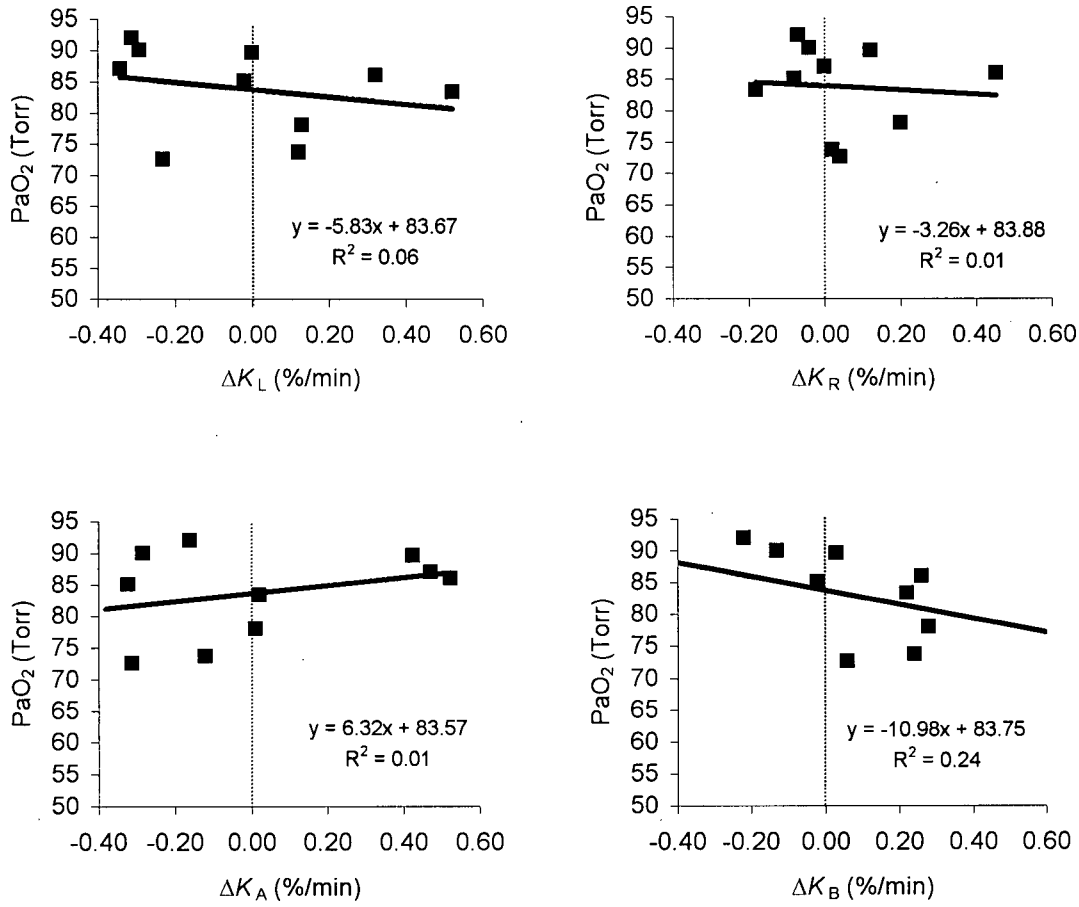

Figure 12. Relationship between the change (post-exercise - resting) in pulmonary clearance rates for left (k_L), right (k_R), apical (k_A) and basal (k_B) regions of the lung and the minimum PaO_2 recorded during exercise test for each subject. Bold line represents regression line and equation and R^2 is noted. Broken line represents zero change in pulmonary clearance rate. Data from 10 of 11 subjects.

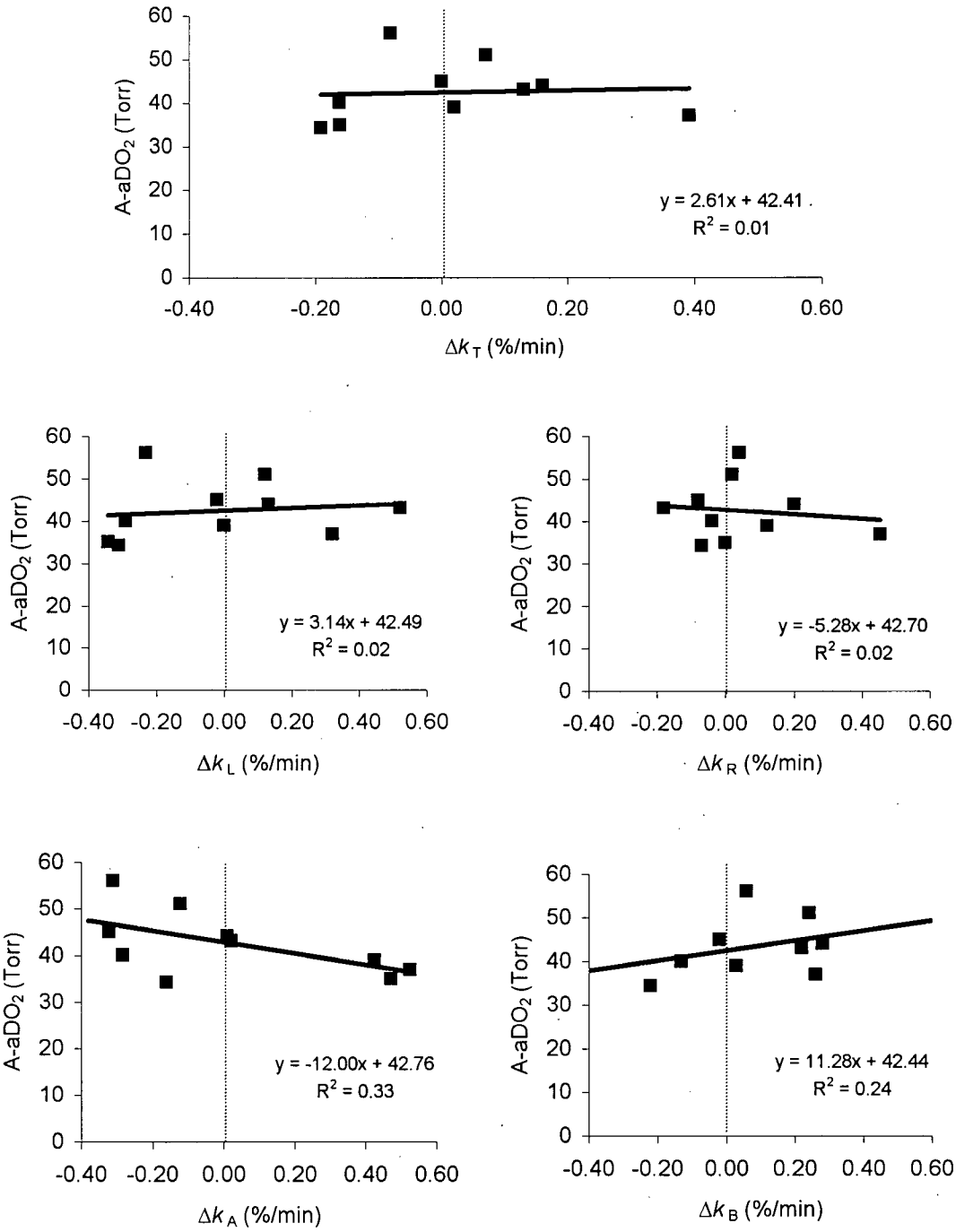

Figure 13. Relationship between the change (post-exercise - resting) in pulmonary clearance rates for total (k_T), left (k_L), right (k_R), apical (k_A) and basal (k_B) regions of the lung and the maximum A-aDO_2 recorded during exercise test for each subject. Bold line represents regression line and equation and R^2 is noted. Broken line represents zero change in pulmonary clearance rate. Data from 10 of 11 subjects.

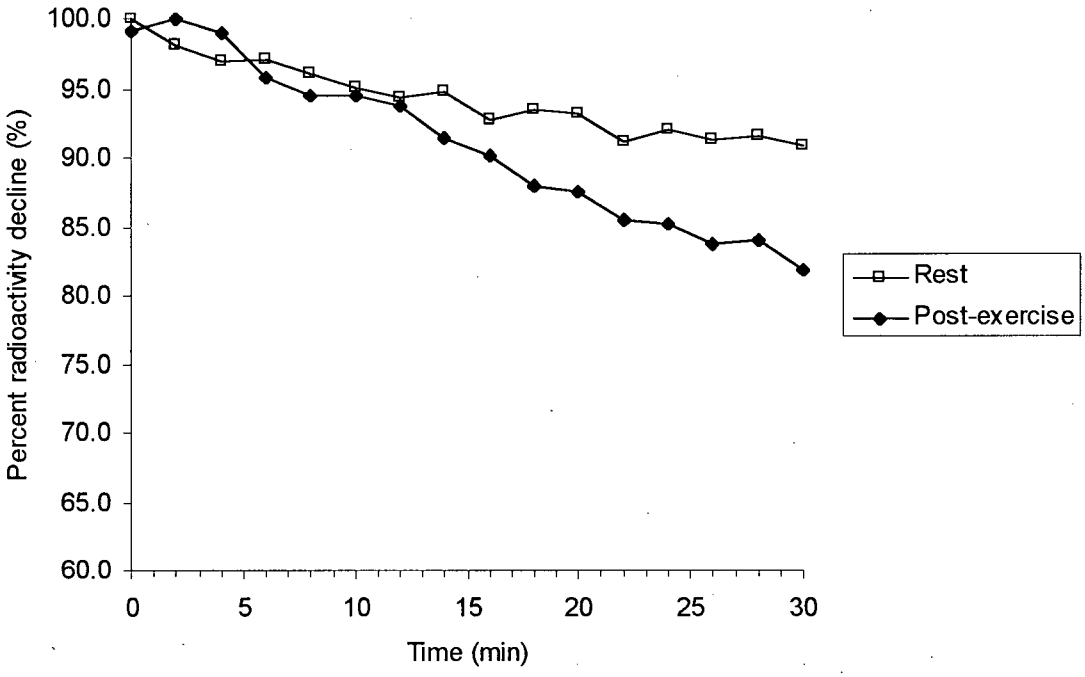

Figure 14. Subject BT pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period.

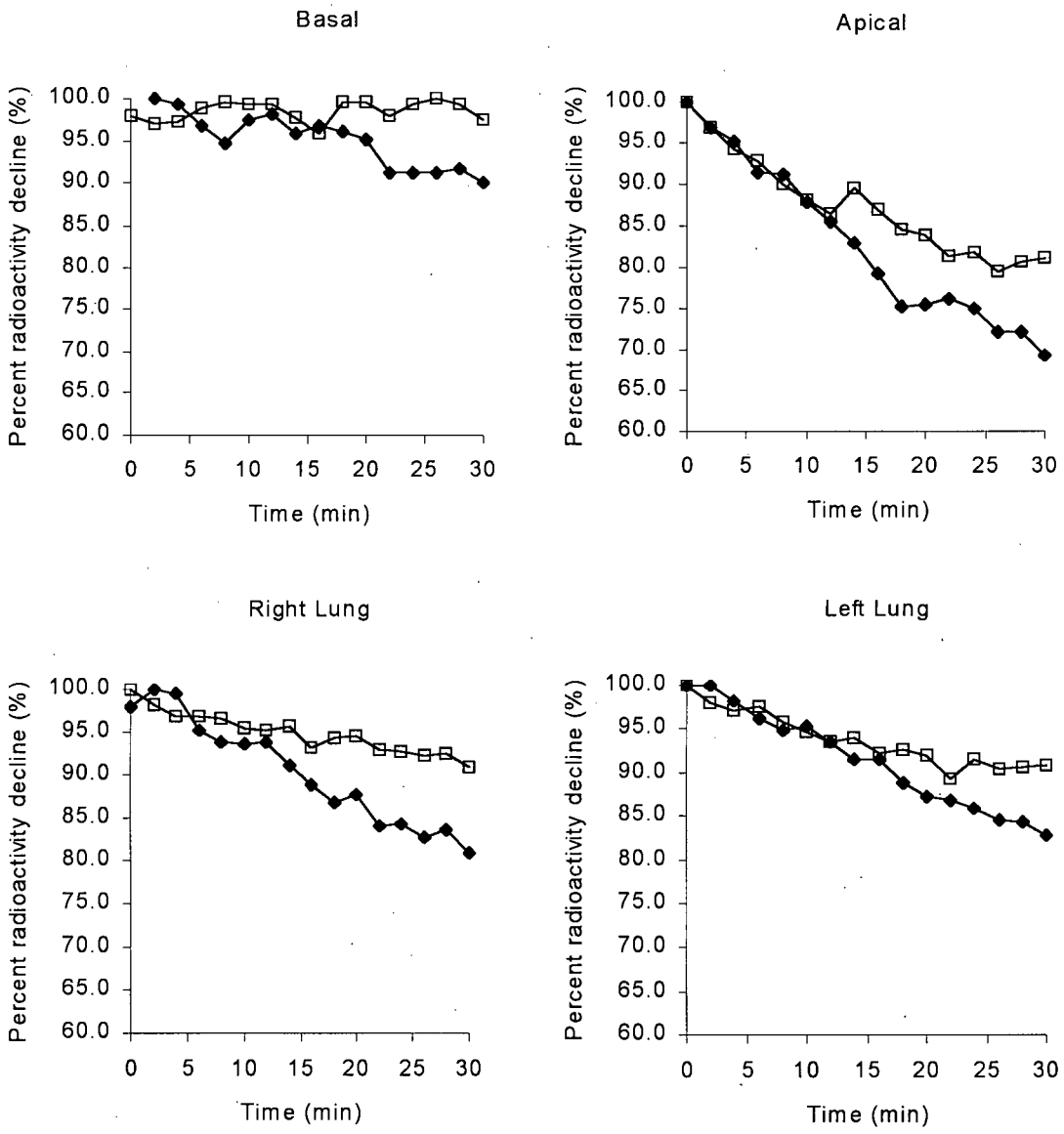

Figure 15. Subject BT pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

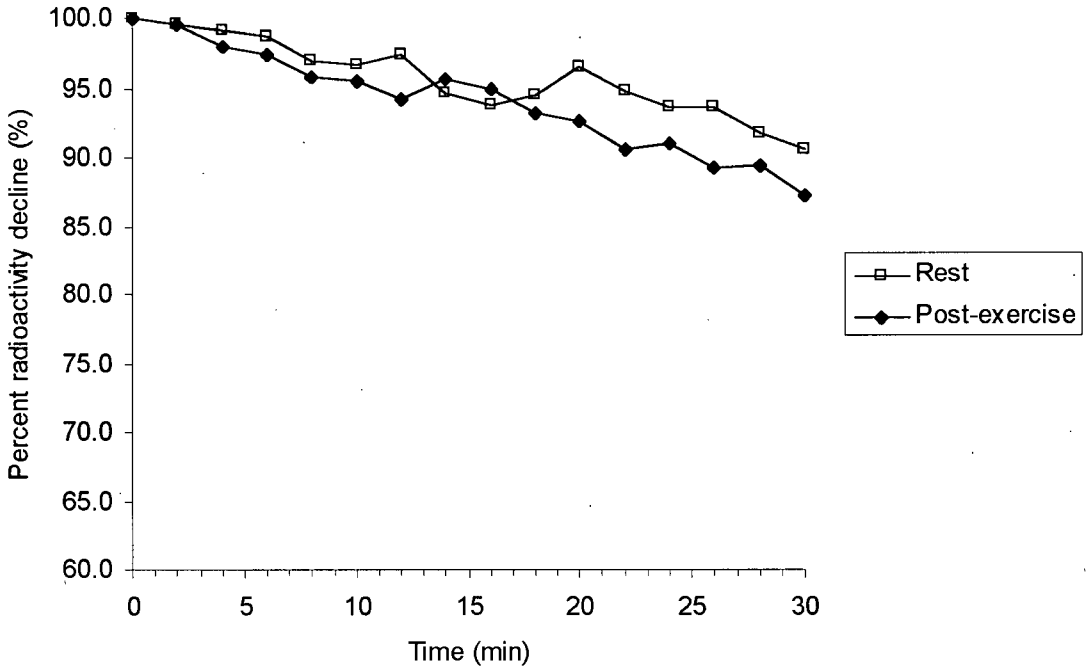

Figure 16. Subject CM pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

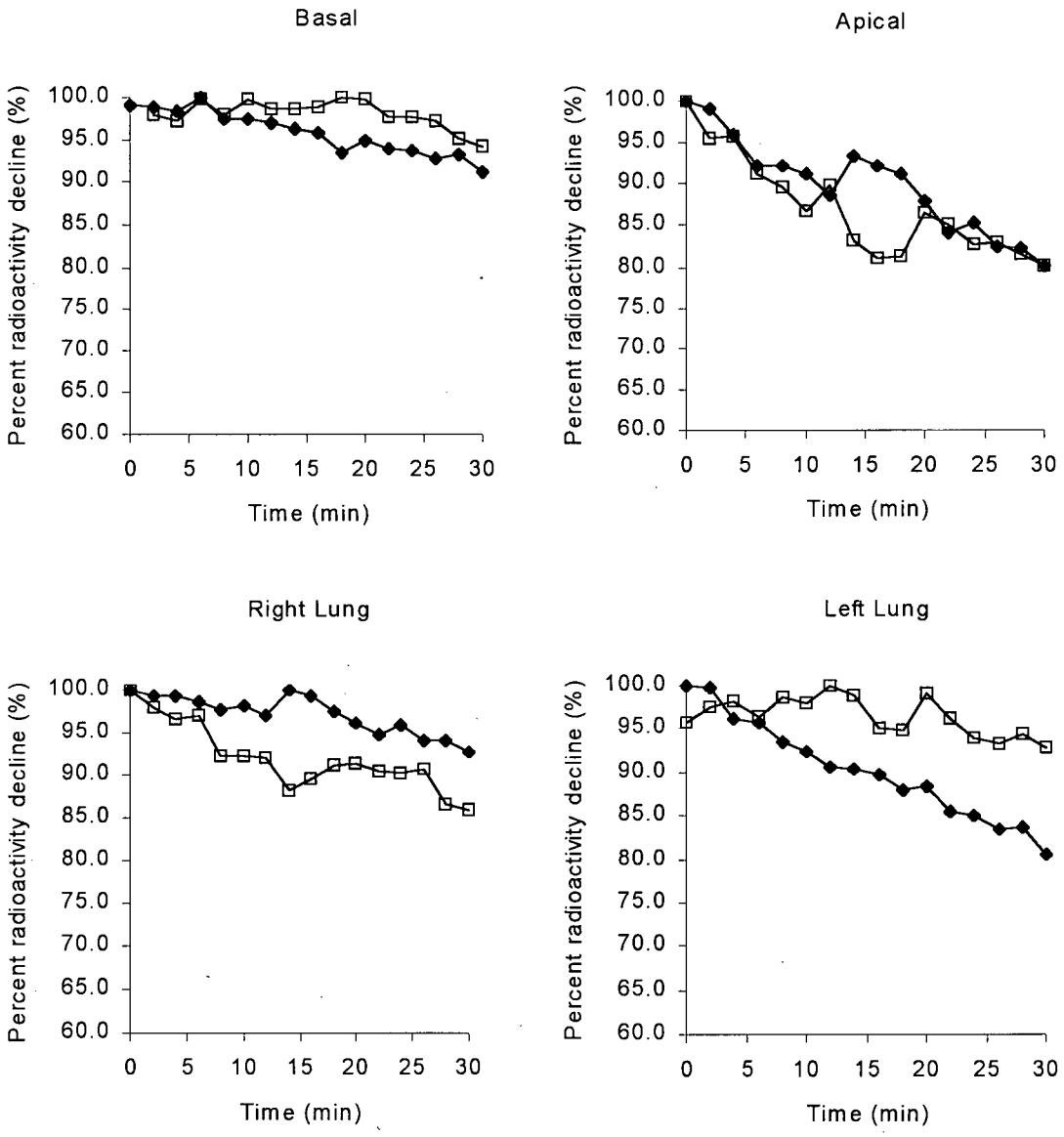

Figure 17. Subject CM pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

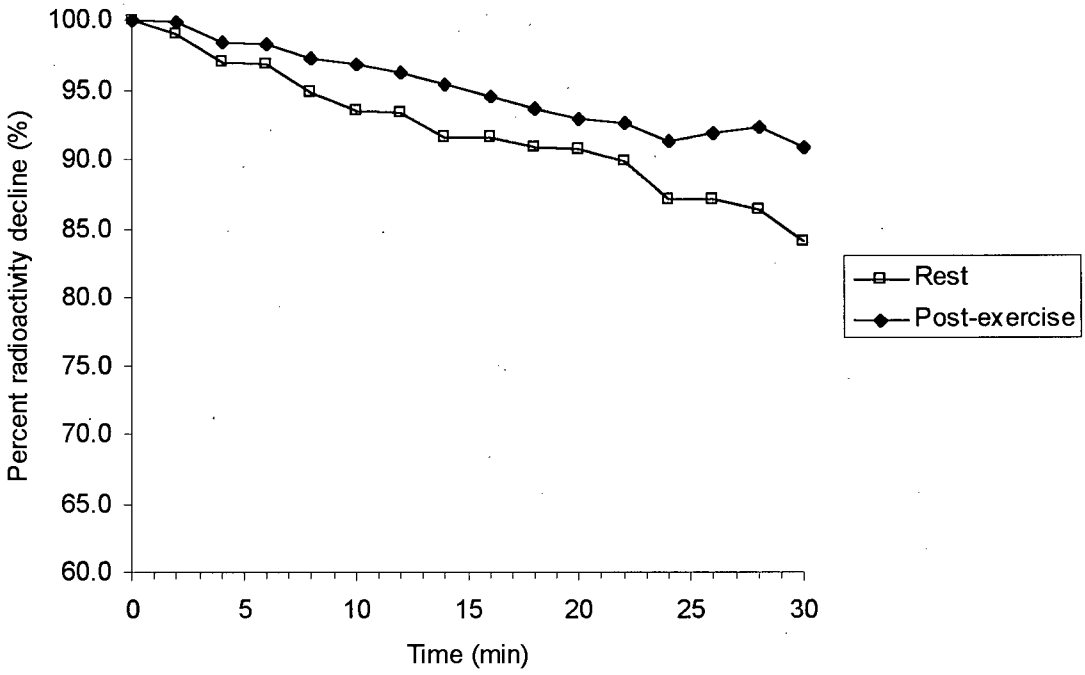

Figure 18. Subject GM pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

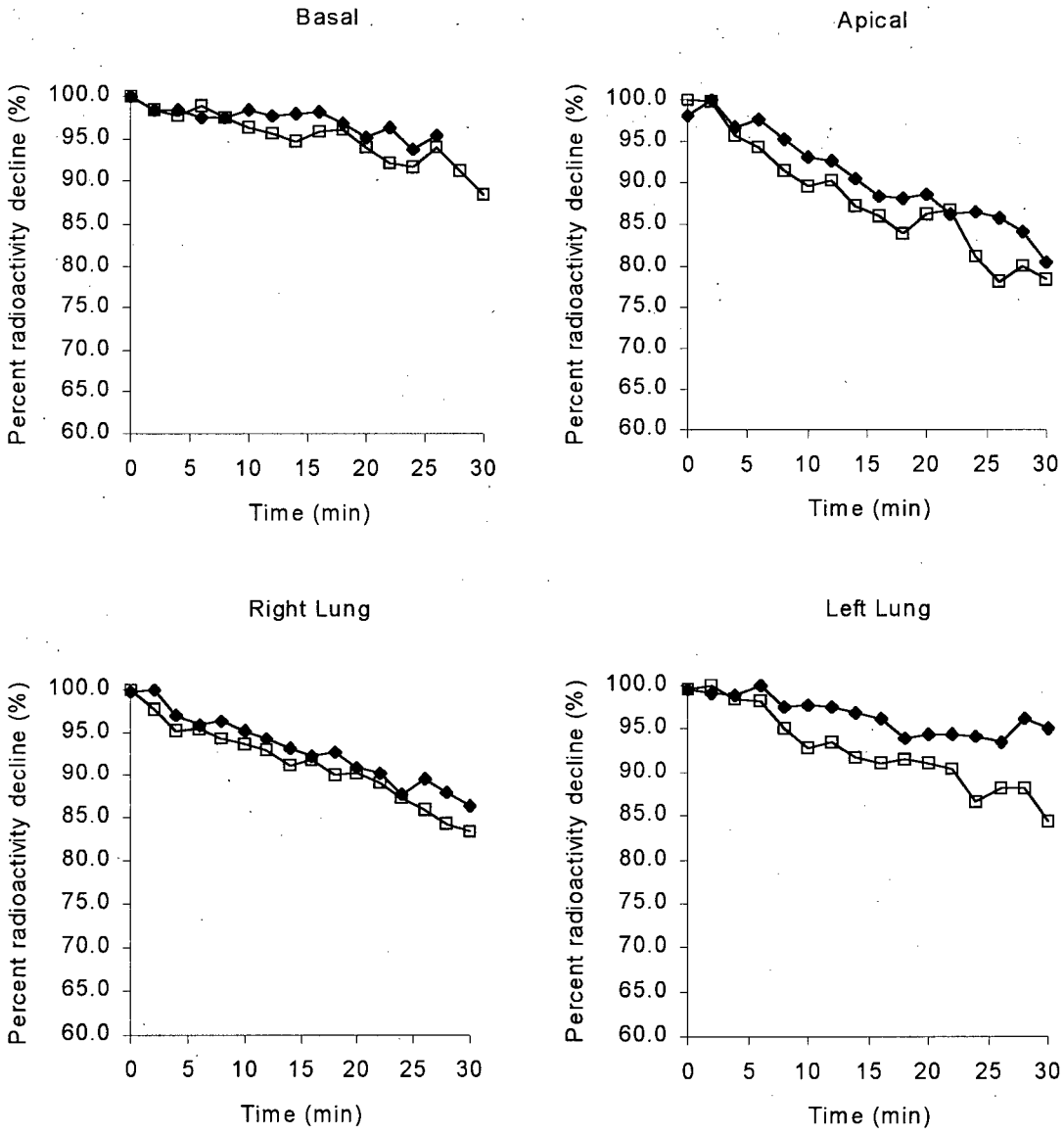

Figure 19. Subject GM pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

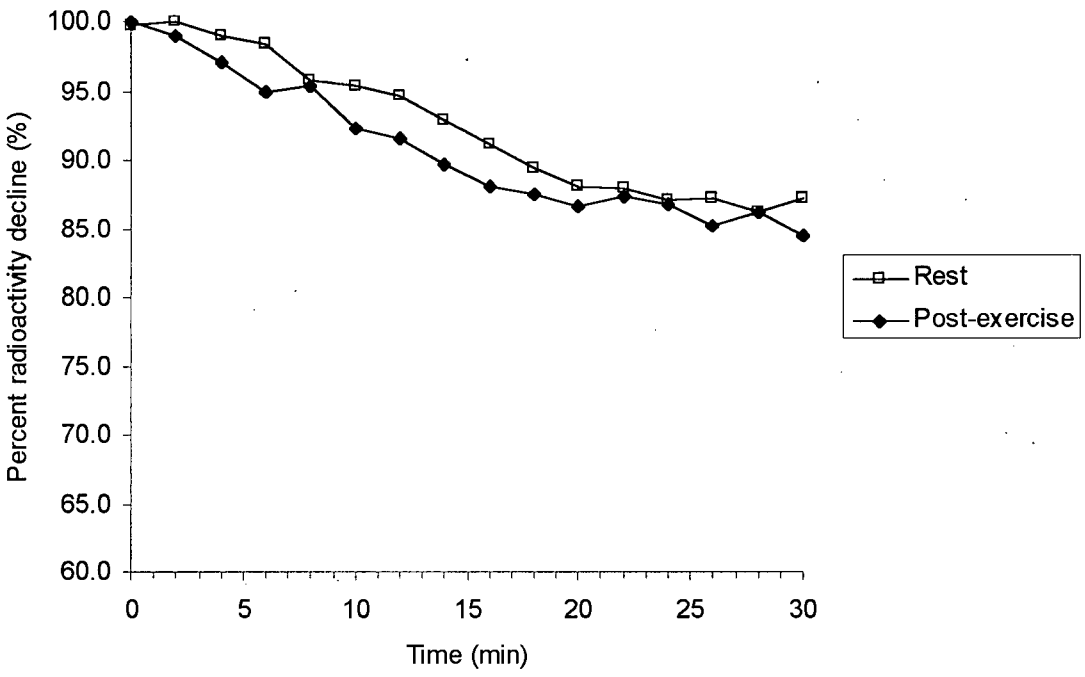

Figure 20. Subject CH pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

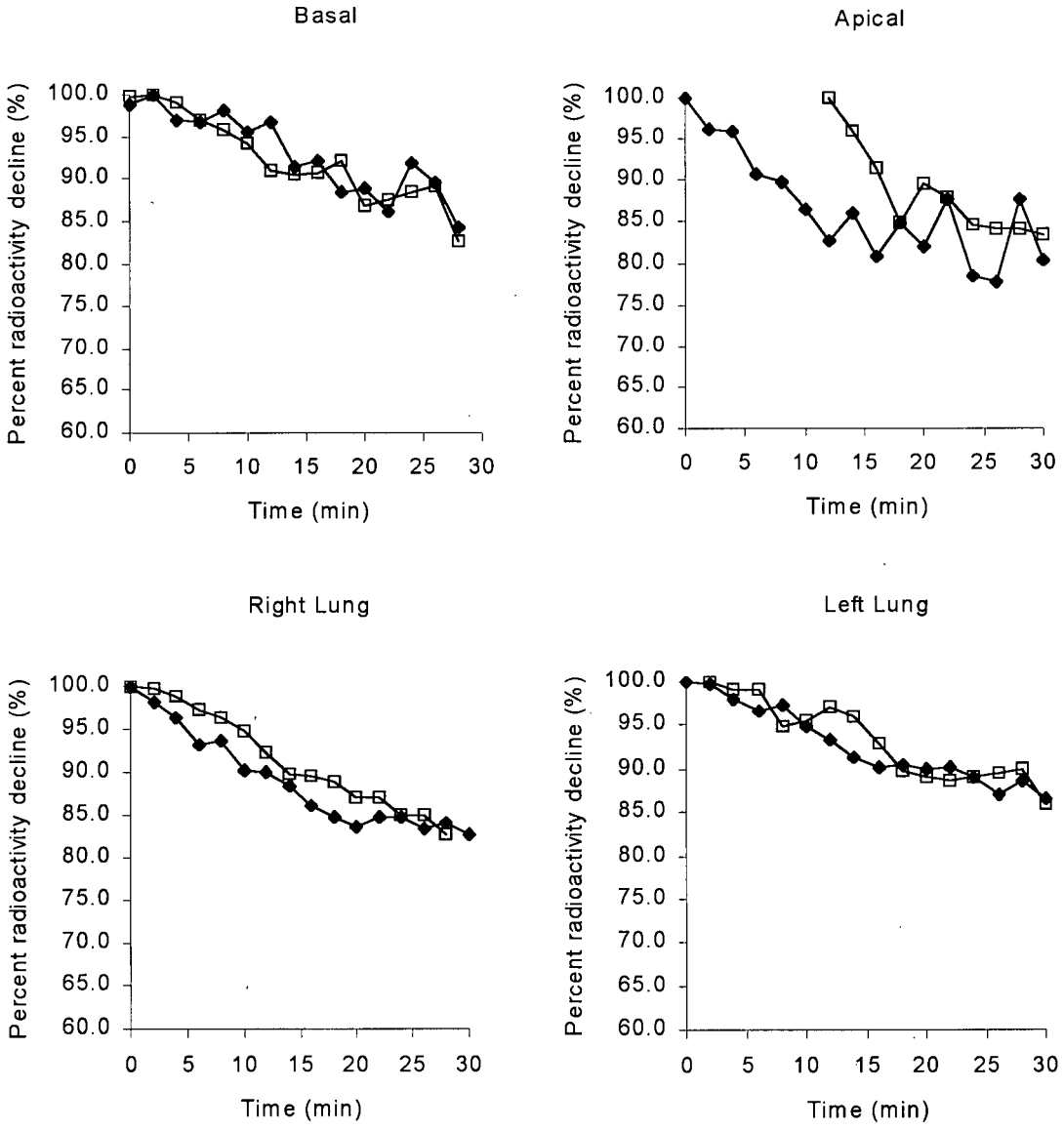

Figure 21. Subject CH pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

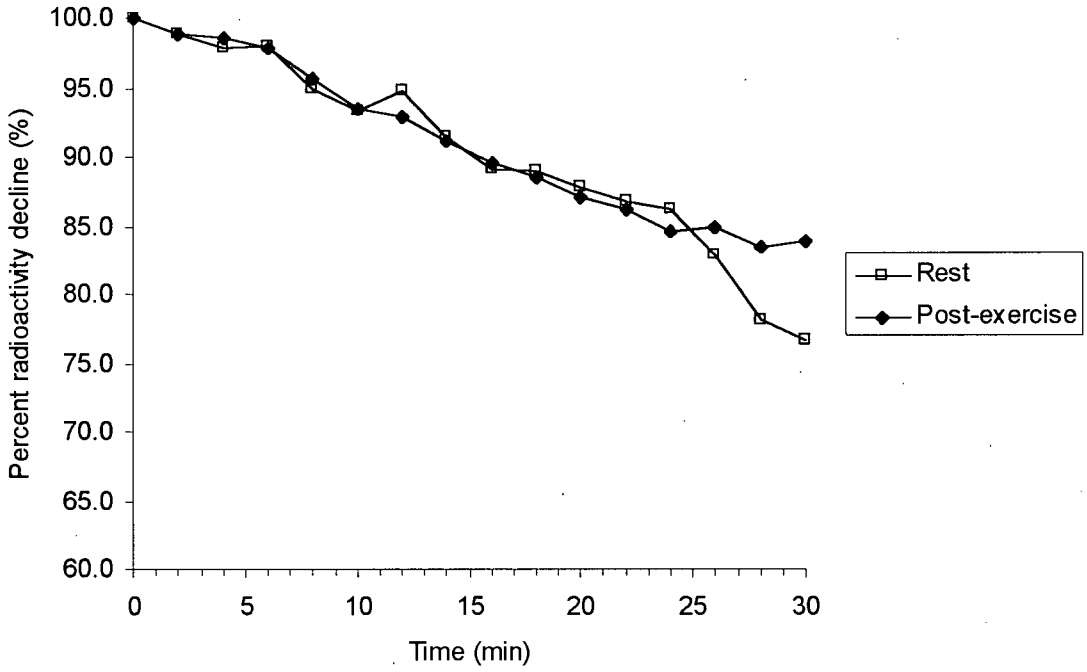

Figure 22. Subject JP pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

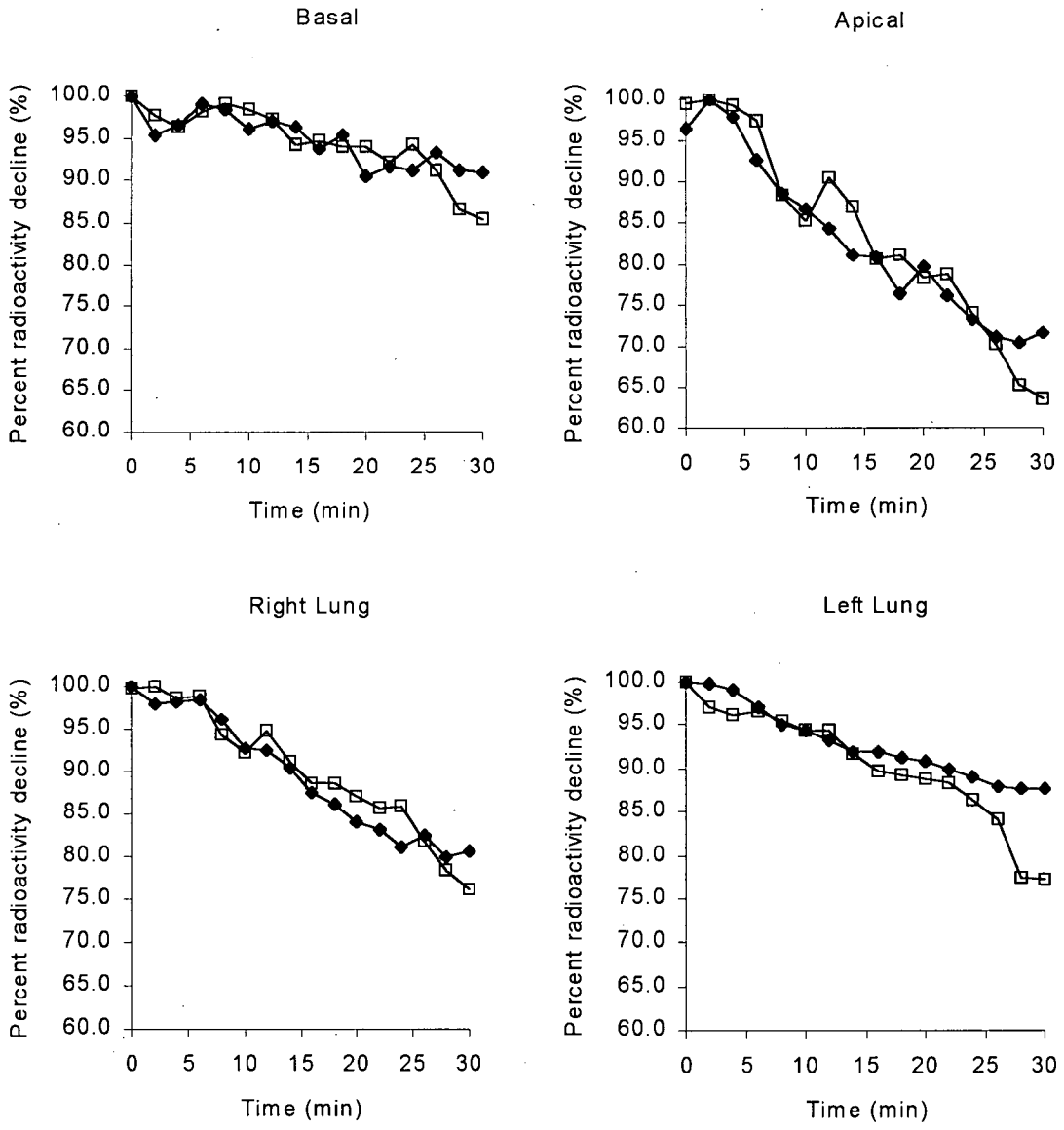

Figure 23. Subject JP pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

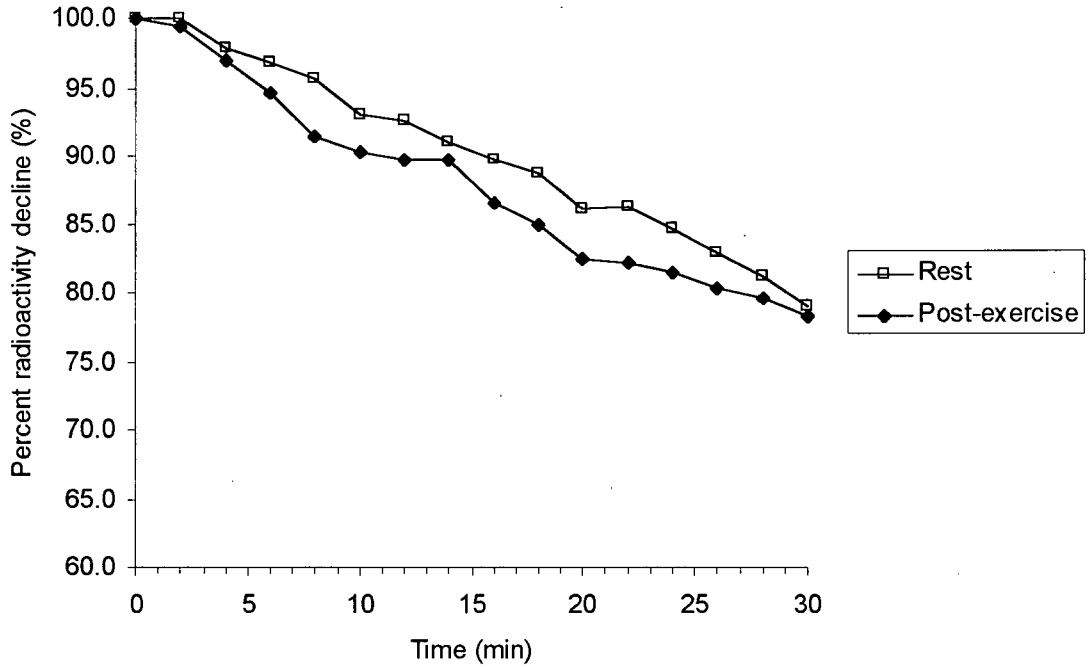

Figure 24. Subject SK pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

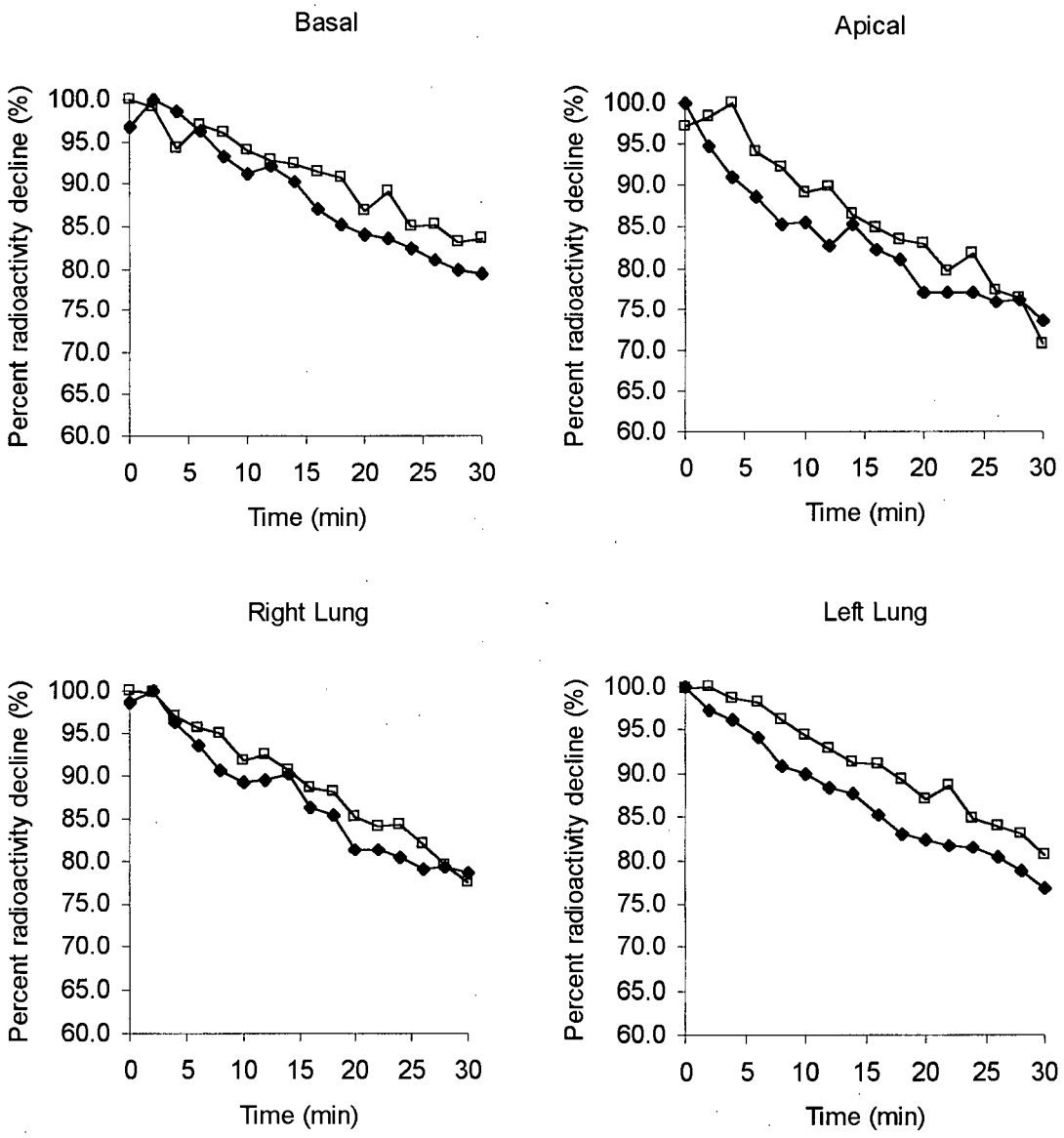

Figure 25. Subject SK pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

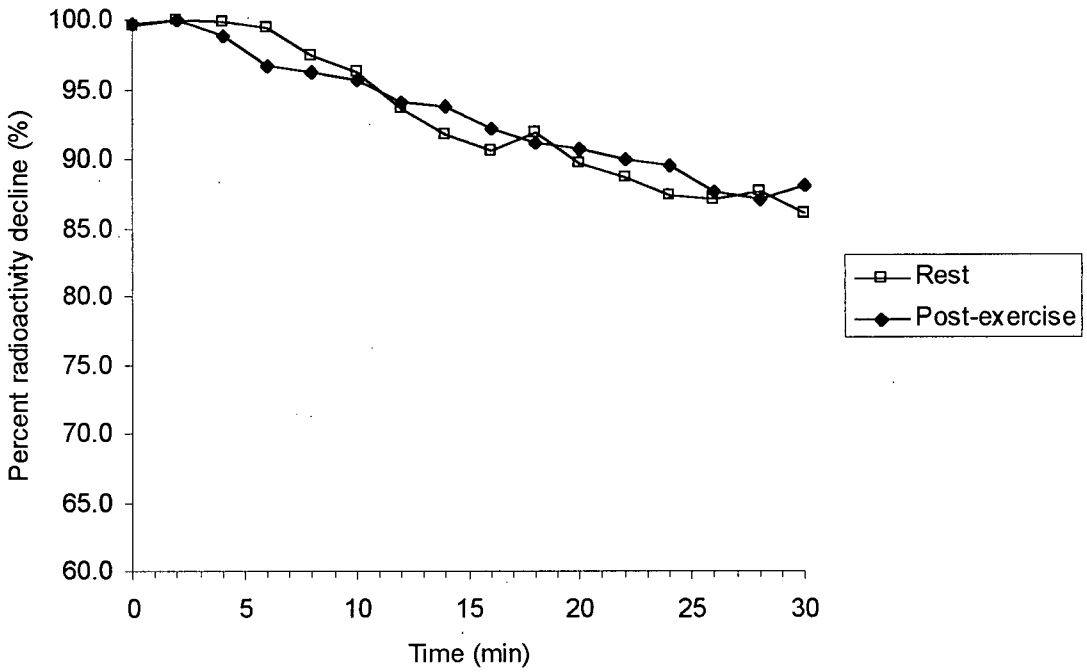

Figure 26. Subject MV pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

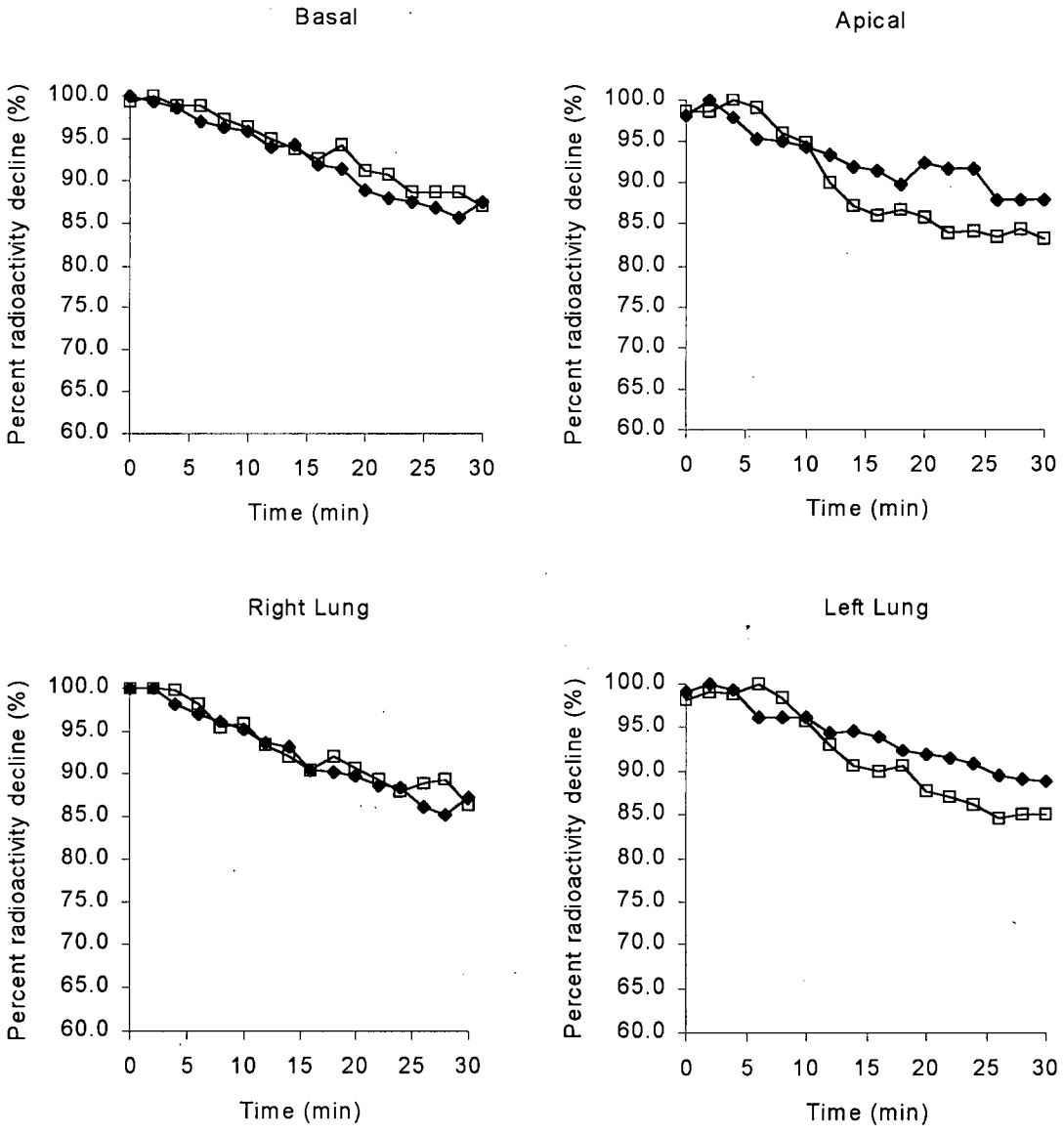

Figure 27. Subject MV pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

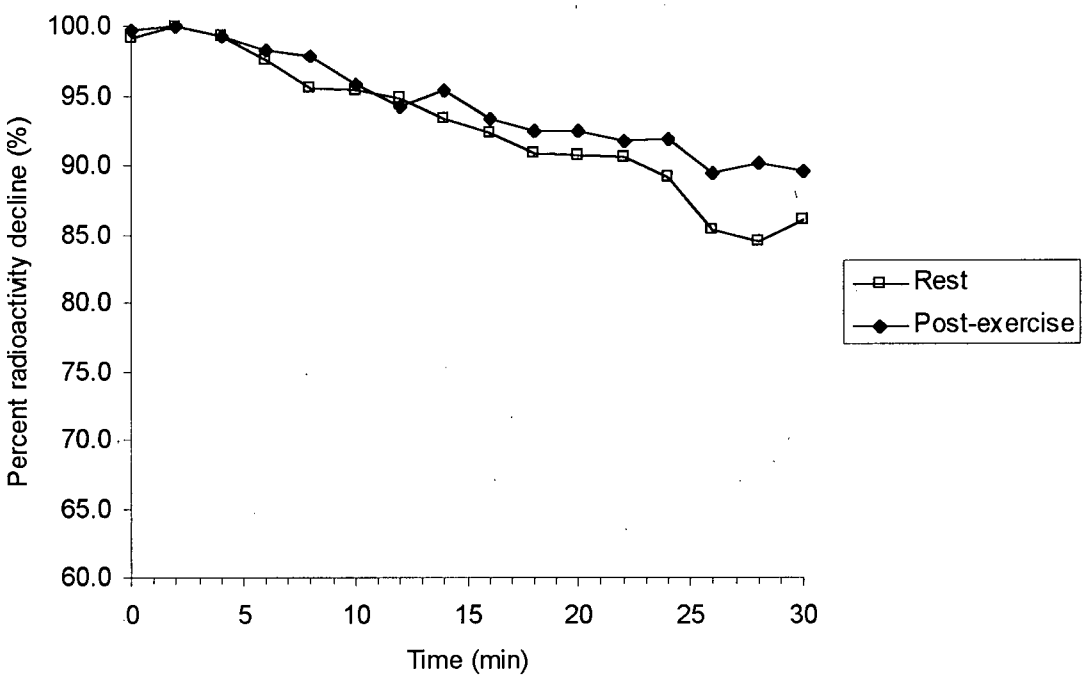

Figure 28. Subject PL pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

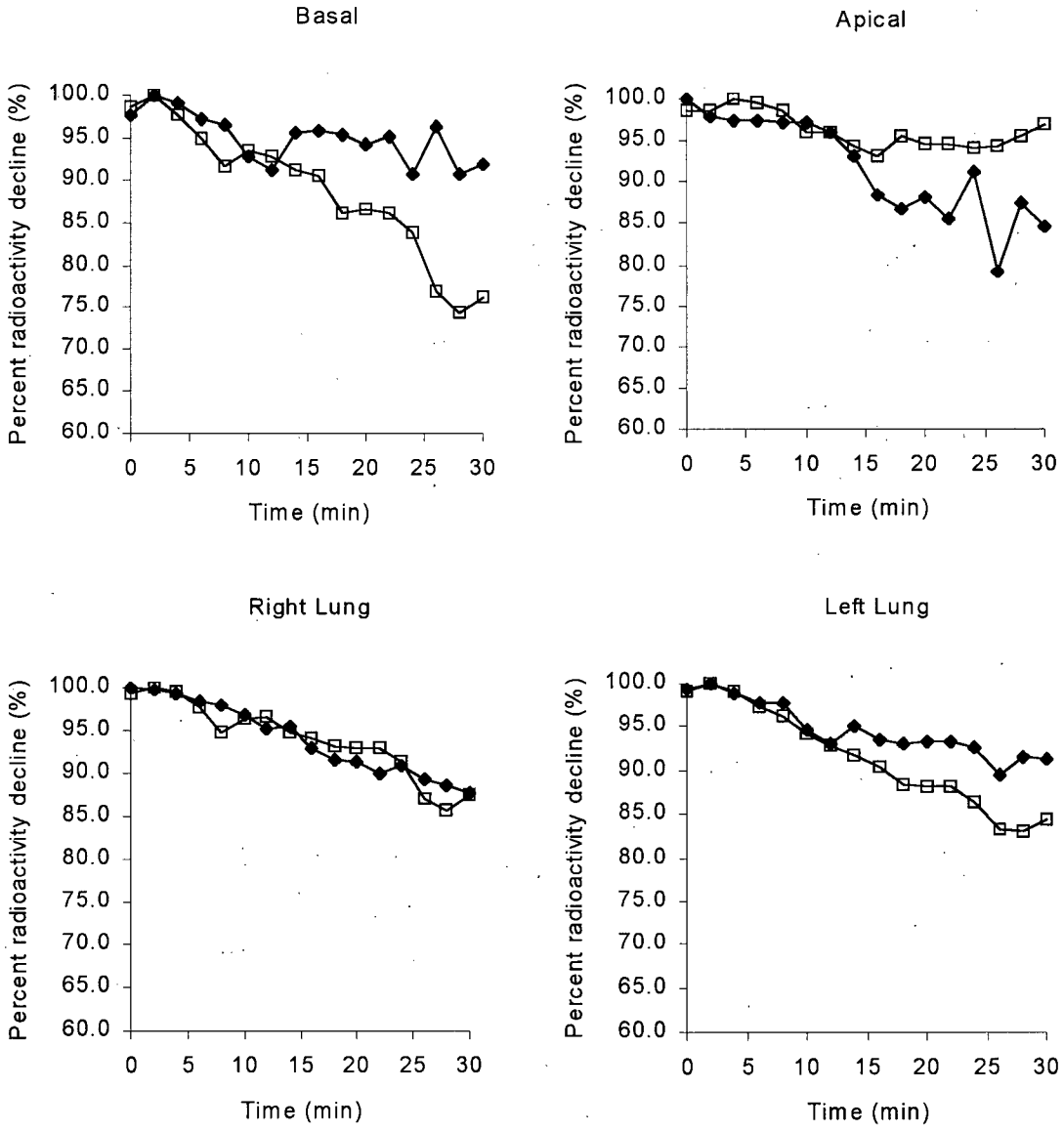

Figure 29. Subject PL pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

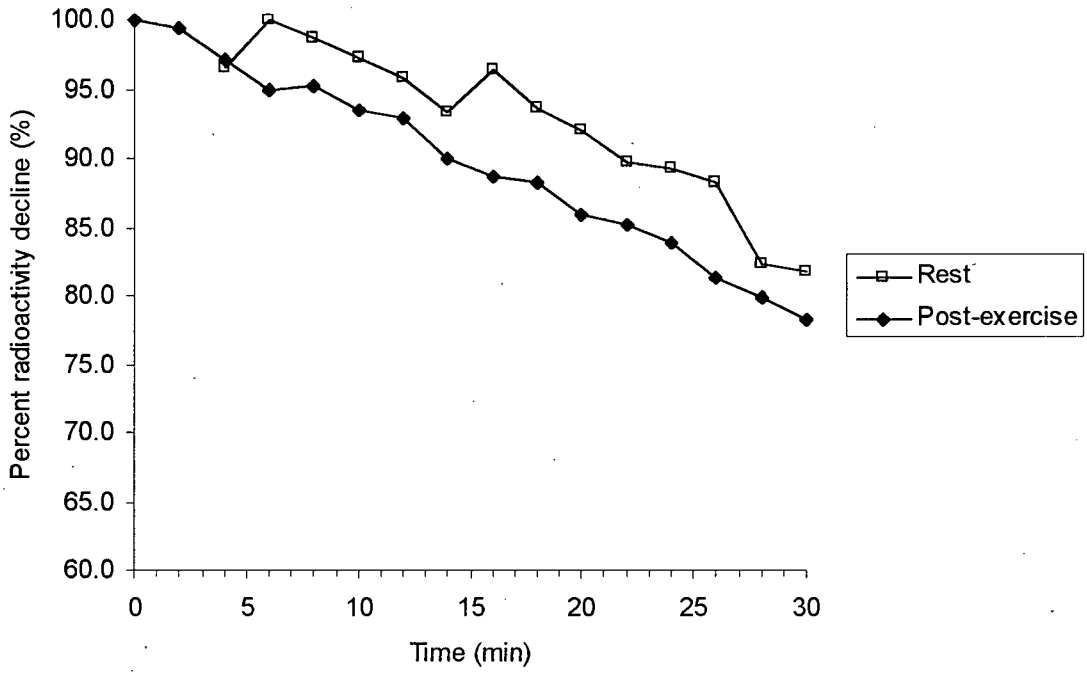

Figure 30. Subject PT pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

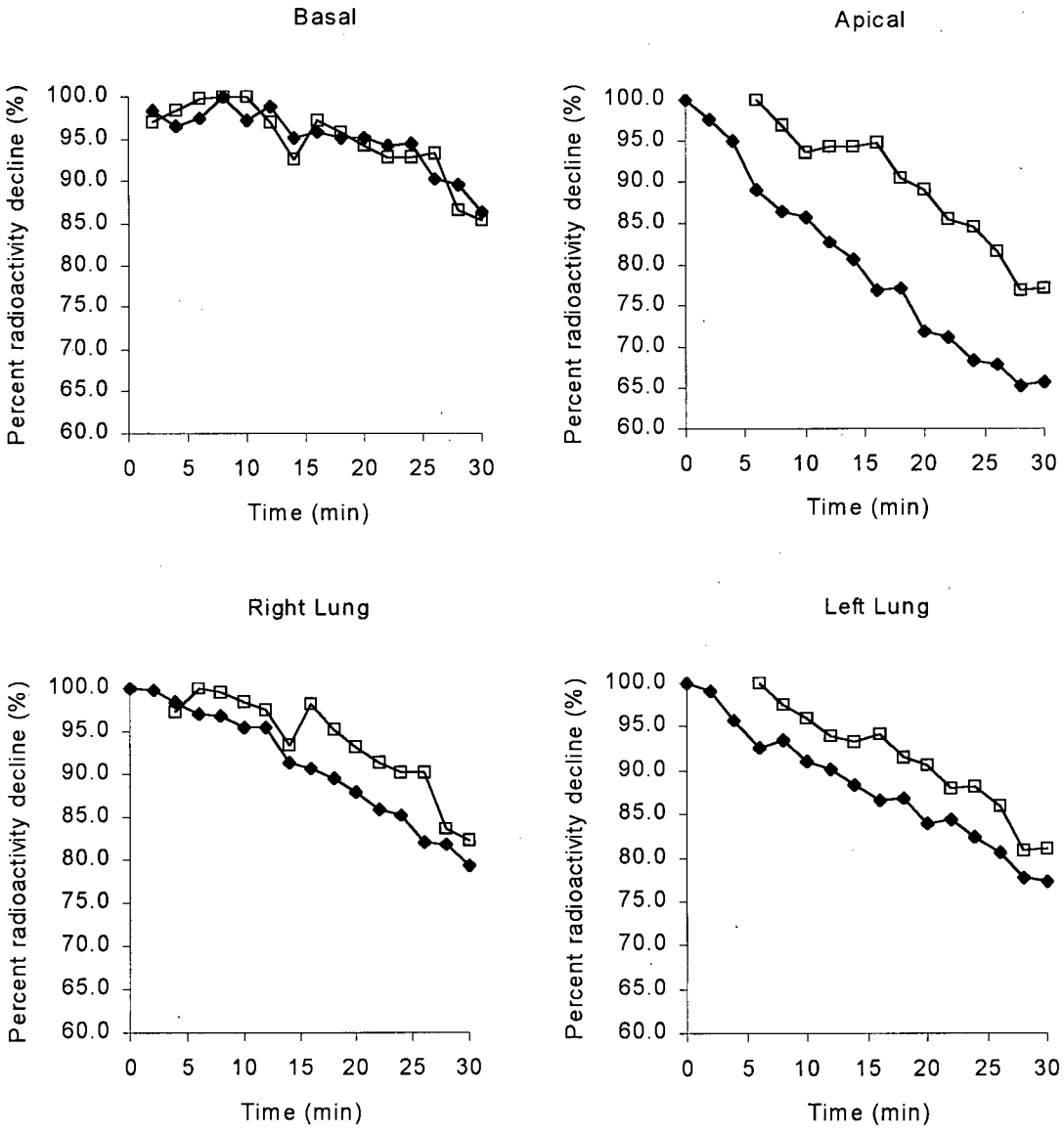

Figure 31. Subject PT pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

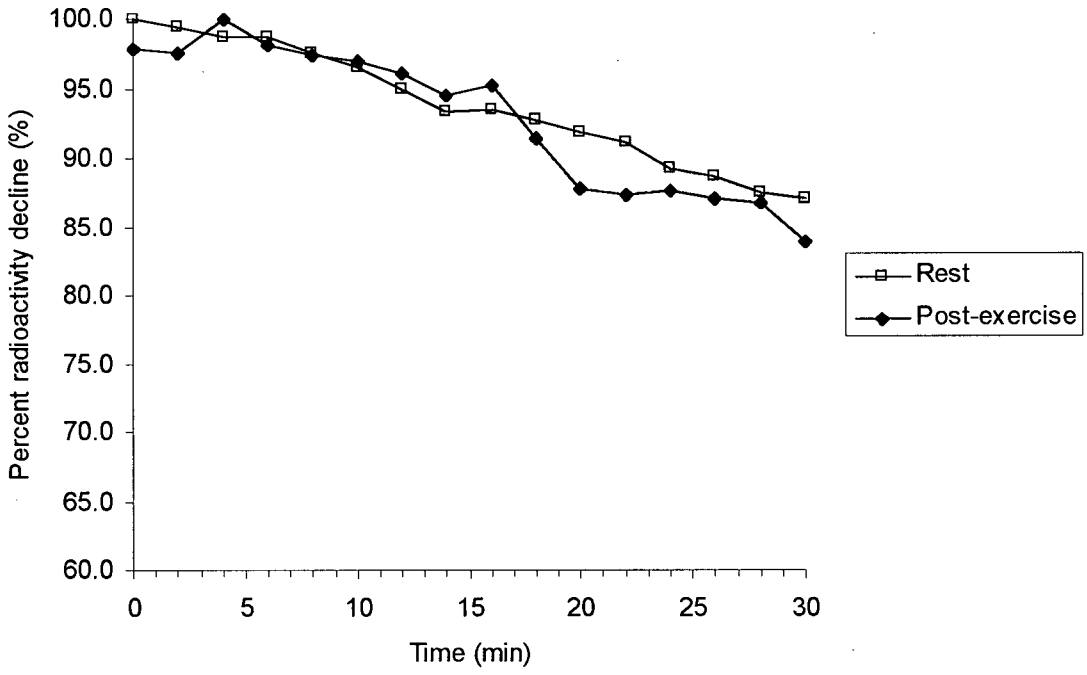

Figure 32. Subject RR pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

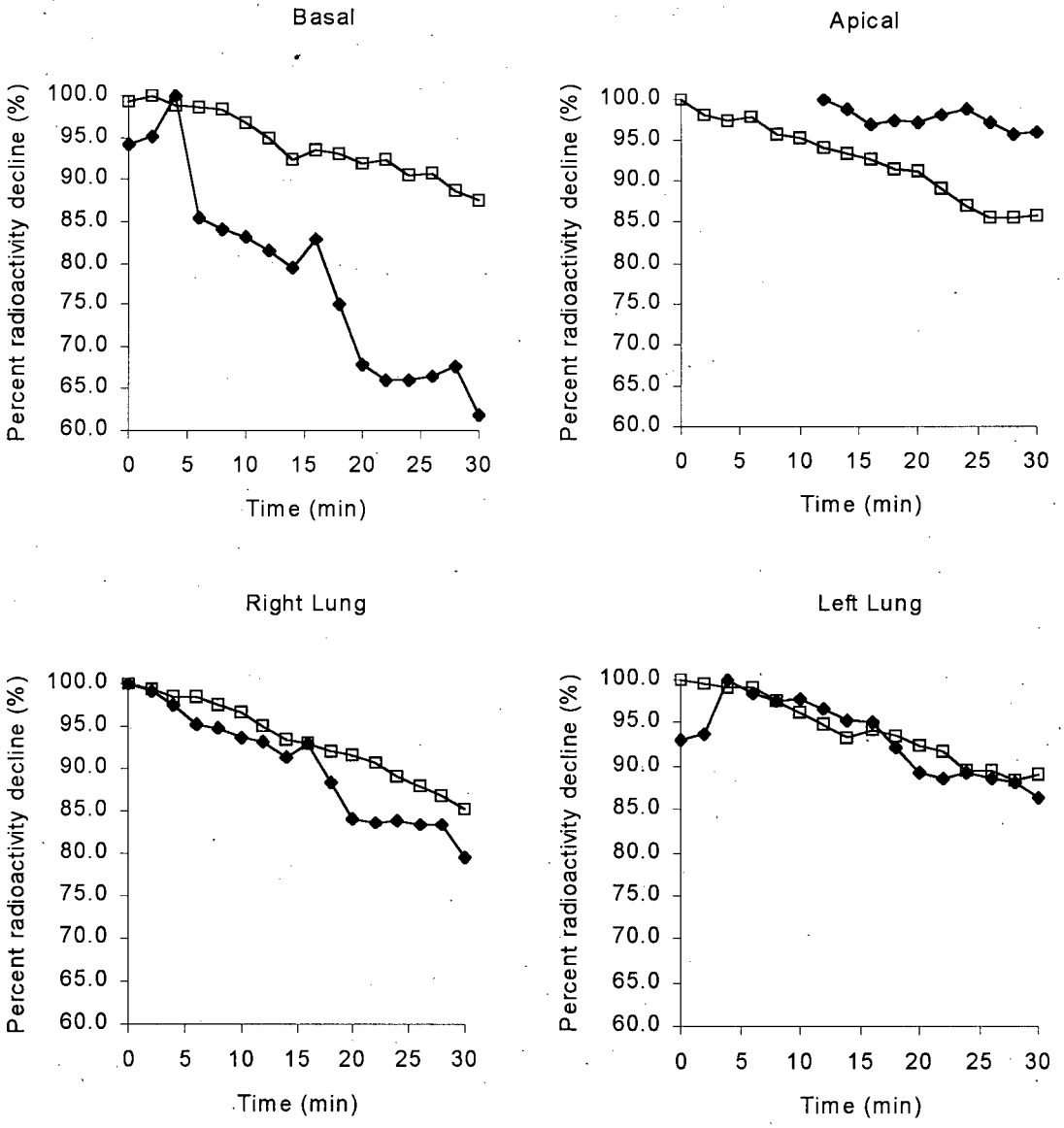

Figure 33. Subject RR pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

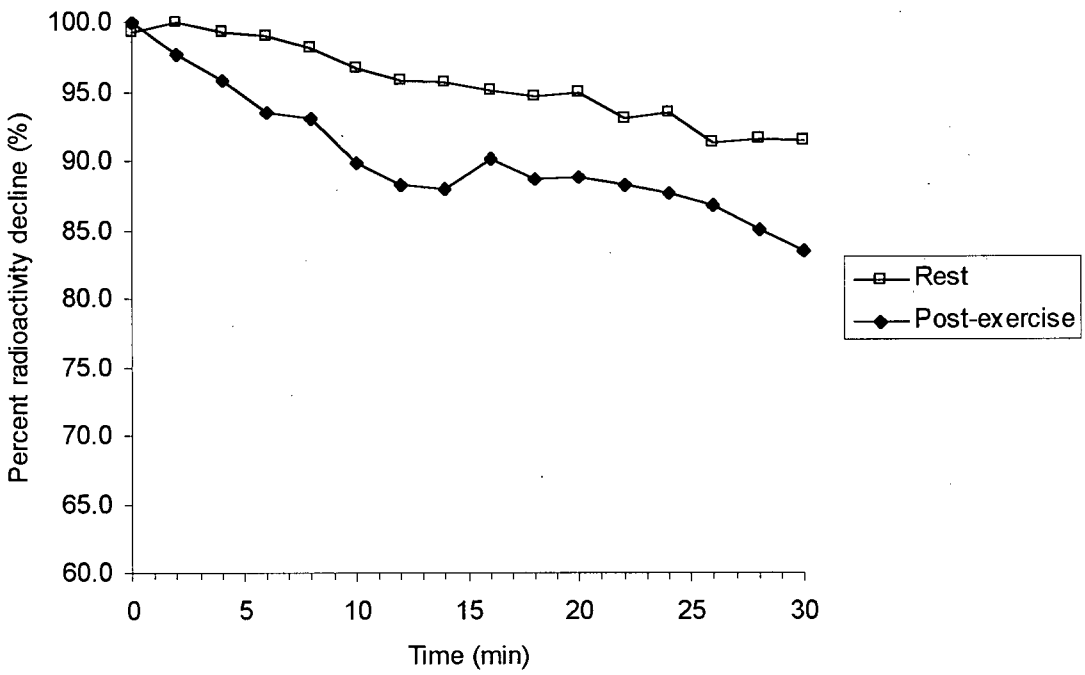

Figure 34. Subject NS pulmonary clearance time-activity curves for total lung acquired for 30 minutes at rest and post-exercise. The radioactive counts at rest and post-exercise were scaled to 100% of highest value obtained during the 30 minute acquisition period

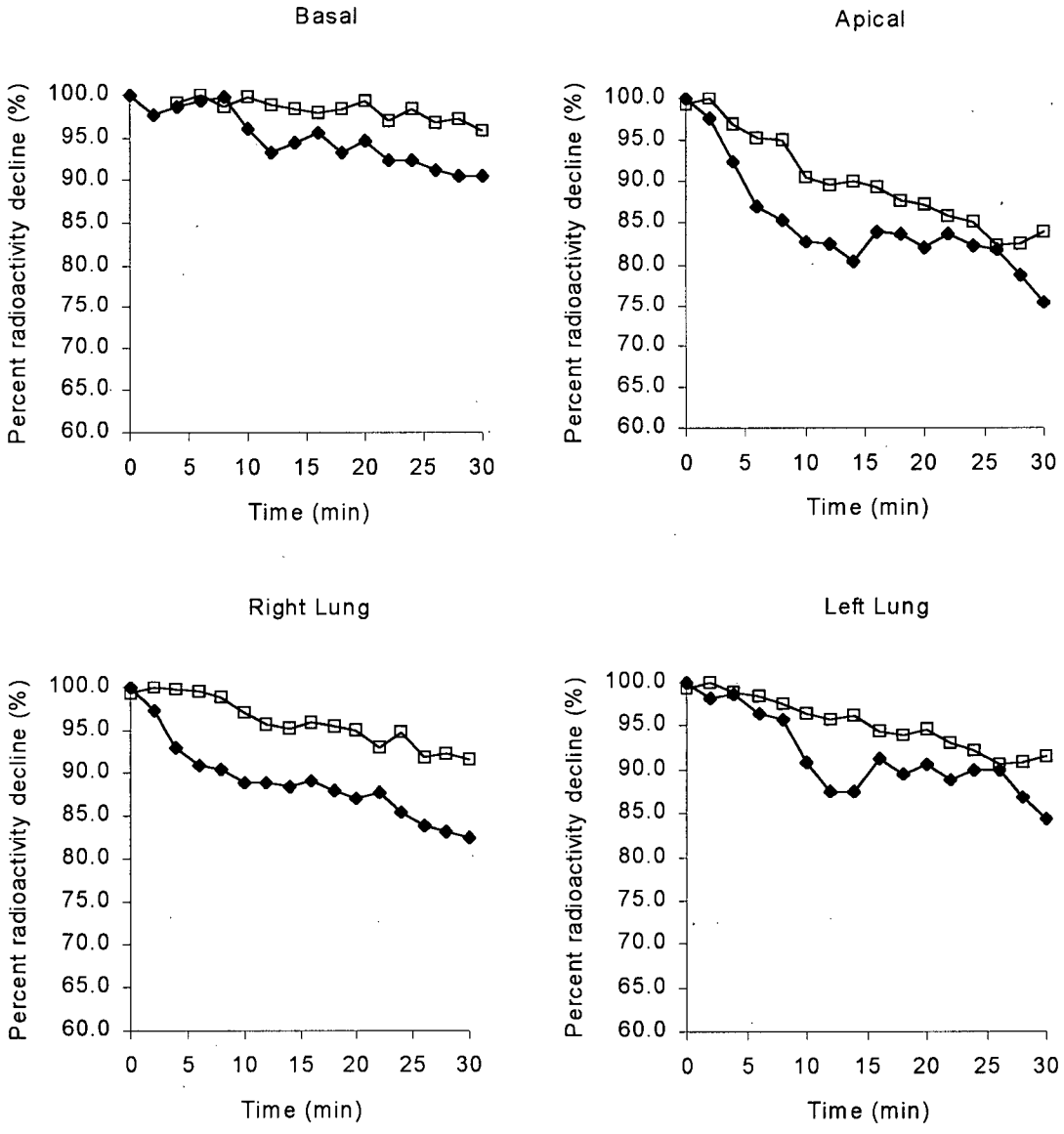

Figure 35. Subject NS pulmonary clearance time-activity curves for basal, apical, right, and left regions acquired for 30 minutes at rest (Y) and post-exercise (♦). See figure 14 for detailed description of chart.

Appenxix IV. Pilot study: Effect of hyperventilation (without exercise) on pulmonary clearance of Tc-99m DTPA.

A pilot study was designed to assess the possibility that high ventilation rates associated with exercise were responsible for the altered clearance rates found in the study by Lorino et al., (1989). Five moderately active subjects were recruited. Each subject completed a resting pulmonary clearance test. At least 48 hours following the resting clearance test, subjects performed a $\dot{V}O_2$max test. $\dot{V}O_2$max and lung clearance tests followed the protocol as previously described. Tidal volume (Tv) and breathing frequency (RR) was recorded every 15 seconds during the exercise test. At least 48 hours later, ventilation patterns without exercising were replicated by simulating the Tv and RR values obtained during the $\dot{V}O_2$max test. A ventilatory apparatus for isocapneic hyperpnea was used for this purpose and is described in detail elsewhere (Fairbarn et al., 1991). Briefly, a pump provided a variable flow rate that matched the subject's ventilation rate. Carbon dioxide (100%) was added to this airflow to maintain end-tidal CO_2 ($P_{ET}CO_2$) at each subject's predetermined resting level. A visual display for target Tv and RR was provided to subjects in order to maintain an assigned minute ventilation (from $\dot{V}O_2$max test). Shortly following the hyperventilation test, post pulmonary clearance was determined.