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Energy Use and Cardiorespiratory Responses to Prolonged Submaximal Exercise

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ABSTRACT. Four Thoroughbred horses were given 75 min exercise on a treadmill inclined at 6° at an intensity equivalent to 50% of maximum oxygen uptake (VO_2max). Before exercise and at 5, 15, 30, 45, 60 and 75 min exercise, arterial and mixed venous blood samples were collected for blood gas analyses, determination of oxygen content, and acid base values. Mixed venous blood samples were used to measure plasma concentrations of lactate. During exercise, heart rate (HR) was measured using telemetry ECG, and $\dot{V}O_2$ and $\dot{V}CO_2$ were determined using an open flow system. Cardiac output ($\dot{Q}$) and stroke volume (Vs) were measured using the direct Fick method. The respiratory exchange ratio (R) decreased ($p<0.05$) from a mean value of 0.86 at 5 min to a lowest value of 0.74 at 45 min exercise. Plasma lactate and standard bicarbonate did not change during exercise. From a 5 min mean value of 170 bpm, the HR decreased by 7% at 15 min but was unchanged thereafter. The $\dot{Q}$ was unchanged from mean values of 163 l min^{-1} until 60 min where there was a 20% decrease ($p<0.01$). This was due to a decrease in Vs of 22%. There was a progressive decrease in mean $PaCO_2$ from 46.9 Torr (rest) to 33.8 Torr (75 min exercise). During this time the mean PaO_2 increased ($p<0.05$) from 103 Torr (rest) to 113 Torr (75 min exercise). Blood temperature showed a progressive rise from a mean of 37.5°C (rest) to 40.9°C (75 min) and mean haematocrit and total plasma protein values increased by 8 and 14%, respectively. The calculated mean energy expenditure during exercise was 45 megajoules (MJ) which translated to around 79 MJ of DE, representing an increase over maintenance of 112%.

Key words: Energy; blood gas; respiratory quotient; oxygen uptake; cardiac output; horses.

INTRODUCTION

Studies of prolonged submaximal exercise in the horse have been largely confined to investigations at competitive endurance rides.[3,6,9,10,15] These studies have indicated that substantial fluid and electrolyte losses occur together with variable plasma biochemical disturbances. While estimation of energy expenditure during this type of exercise has been made,[2] there is little information on metabolic changes during prolonged exercise where energy costs have been measured by oxygen uptake and carbon dioxide production.[13,14] The only study of energy expenditure during prolonged exercise is that of Pagan et al.[14] where horses were exercised at exercise intensities around 25–30% VO_2max. With the advent of newer training techniques more emphasis has been placed upon slow, long-distance training in Thoroughbred racehorses.[8] It is, therefore, important to understand some of the physiological responses to prolonged exercise in the Thoroughbred horse and to estimate the possible energy expenditure.

Given this background, our study had two main aims: 1) to examine metabolic and cardiorespiratory changes during prolonged submaximal exercise, and 2) to provide information on energy use during prolonged exercise at speeds equivalent to 50% VO_2max.

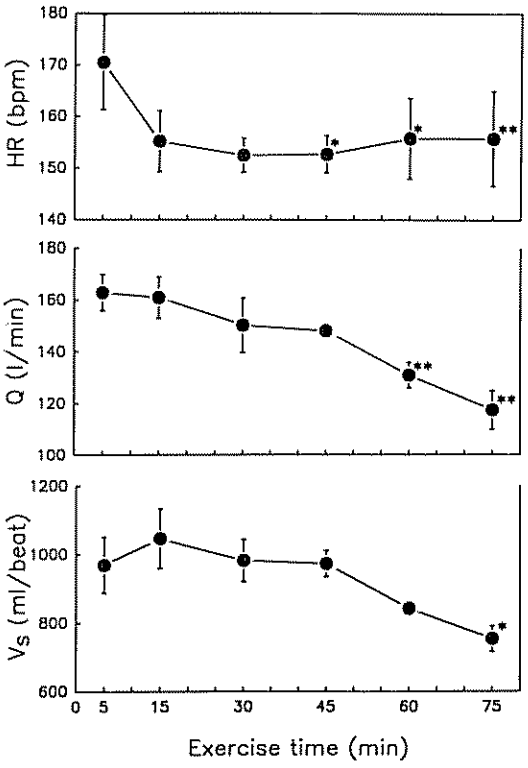

Fig 1 Heart rate (HR), cardiac output ($\dot{Q}$) and stroke volume (Vs) values (mean ± SEM) in 4 Thoroughbred horses during 75 min of exercise at 50% $\dot{V}O_2$max * $p<0.05$, ** $p<0.01$—significant differences from values at 5 min exercise.

MATERIALS AND METHODS

Four Thoroughbred horses aged between 4 and 7 years were used in the study. Bodyweights ranged from 440–480 kg. The horses had undergone regular treadmill exercise but were not in active training. Prior to the experiment, two other Thoroughbred horses were used to evaluate the exercise capacity of horses at speeds that were equivalent to approximately 50% of maximal oxygen uptake ($\dot{V}O_2$max). These studies indicated that unfit Thoroughbreds could maintain this exercise intensity for around 75 min.

Prior to the experiment, the horses were given a total of three incremental exercise tests on a treadmill (Beltalong, Euroa, Victoria) set at a +10% slope. During these tests, oxygen uptake ($\dot{V}O_2$) and carbon dioxide production ($\dot{V}CO_2$) were measured using an open flow mask system described previously.[18] The test commenced at 4 m s^{-1} for 3 min, followed by 90 s at 6 m s^{-1}, and 60 s at 8, 10, 11 and 12 m s^{-1}. Not all horses completed the exercise test, which was terminated when a plateau in $\dot{V}O_2$ was apparent. From the tests, $\dot{V}O_2$ versus speed regression equations were determined over the linear part of the data (4–10 m s^{-1}). By using the individual horse's $\dot{V}O_2$max value, the speed was calculated at which 50% $\dot{V}O_2$max was reached. This calculated speed was used for each horse during the 75 min exercise.

Before commencing the prolonged exercise, catheterisation of the carotid artery, previously relocated to a subcutaneous position, was performed using an 18 g catheter. The pulmonary artery was catheterised using a 110 cm pig tailed catheter inserted via the jugular vein. Both catheters had extension tubes attached so that blood samples could be withdrawn while the horse was exercising. The position of the pulmonary artery catheter was verified by its connection to a transducer and oscilloscope so that the characteristic change in trace could be seen as the catheter moved from the right ventricle to the pulmonary artery. A further catheter was placed into the jugular vein to allow the introduction of a thermistor to measure blood temperature at the level of the right atrium as described previously.[17]

After placing the horse on the treadmill, resting arterial and mixed venous blood samples (2 ml) were collected for blood gas analysis and measurement of acid base values using a blood gas machine (ABL 3, Radiometer, Copenhagen). On these samples, the haematocrit and total plasma protein were determined using the microhaematocrit method and refractometry, respectively. Mixed venous blood samples were also collected for measurement of plasma lactate concentrations using an automated method (YSI-23LM, Yellow Springs Instruments, Columbus, OH).

After application of a respiratory gas col-

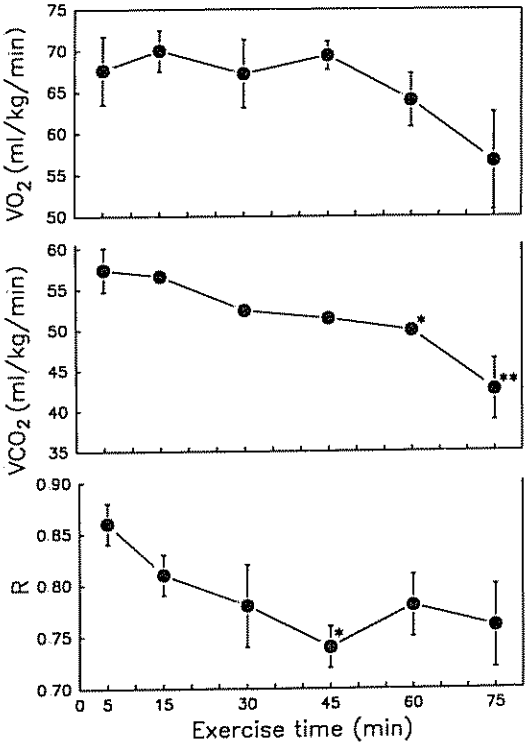

Fig 2 Oxygen uptake ($\dot{V}O_2$), carbon dioxide production ($\dot{V}CO_2$) and respiratory exchange ratio (R) values (mean ± SEM) in 4 Thoroughbred horses during 75 min of exercise at 50% $\dot{V}O_2$max. * $p<0.05$, ** $p<0.01$—significant differences from values at 5 min exercise.

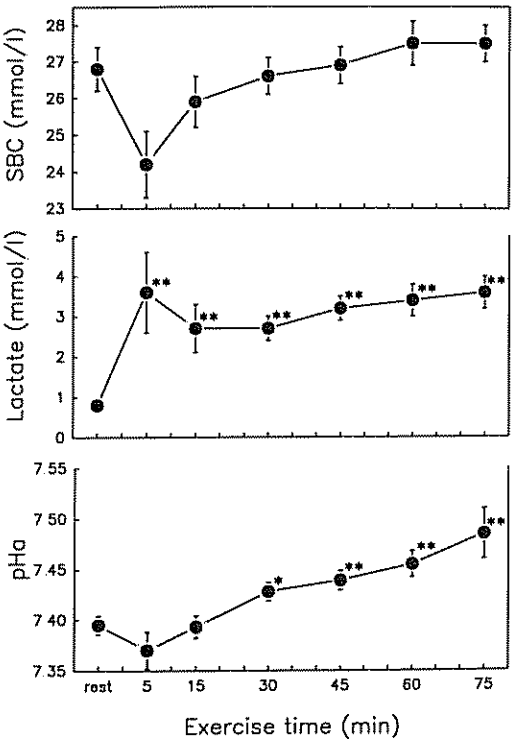

Fig 3 Arterial blood standard bicarbonate (SBC), arterial pH (pHa) and plasma lactate values (mean ± SEM) in 4 Thoroughbred horses before and during 75 min of exercise at 50% $\dot{V}O_2$max. * $p<0.05$, ** $p<0.01$—significant differences from values at rest.

lection mask, exercise was commenced with the horse being exercised at the speed appropriate to produce about 50% of $\dot{V}O_2$max. During the test, a 120 W fan was placed in front of the treadmill to assist in cooling the horses. At 5, 15, 30, 45, 60 and 75 min exercise, arterial and mixed venous blood samples were obtained for blood gas and acid base analyses while the $\dot{V}O_2$ and $\dot{V}CO_2$ and respiratory exchange ratio (R) were measured. The oxygen contents of arterial and mixed venous blood were also determined at each of the measurement times using the machine's inbuilt computer. This technique had previously been verified using directly measured oxygen contents according to the method of Tucker.[21]

Heart rate (HR) during exercise was measured using telemetry electrocardiography as previously described.[4] Stroke volume (Vs) was determined using the direct Fick technique.

RESULTS

The ambient temperature during the 75 min exercise varied from 14 to 19°C. All horses completed the exercise bout, but in two cases there was great difficulty in continuing exercise after 65 min and required additional traction by the person holding the lead rope.

Heart rate during exercise was higher ($p<0.01$) at 5 min than throughout the rest of the exercise but was unchanged from 15

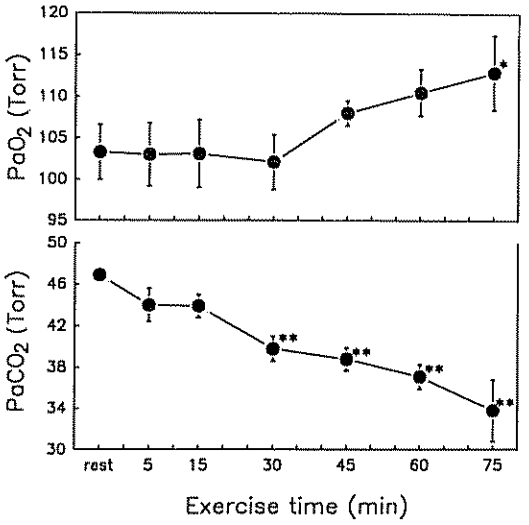

Fig. 4. Arterial oxygen (PaO_2) and carbon dioxide ($PaCO_2$) tensions (mean ± SEM) in 4 Thoroughbred horses before and during 75 min of exercise at 50% $\dot{V}O_2$max. * $p<0.05$, ** $p<0.01$—significant differences from values at rest.

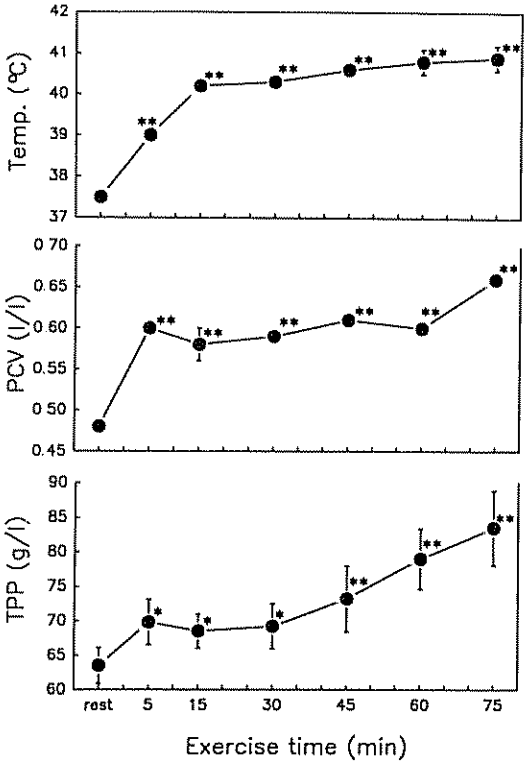

Fig. 5. Mixed venous blood temperature (temp), arterial haematocrit (PCV) and total plasma protein (TPP) in 4 Thoroughbred horses before and during 75 min of exercise at 50% $\dot{V}O_2$max. * $p<0.05$, ** $p<0.01$—significant differences from values at rest.

min until the end (Fig. 1). The cardiac output (Q) was lower ($p<0.05$) from 60 min until the end of exercise, due to a decrease in Vs (Fig. 1). The $\dot{V}O_2$ showed no change throughout exercise although the VCO_2 was lower ($p<0.05$) at 60 and 75 min exercise than at 5 min exercise. This resulted in a decrease in R from 45 min exercise (Fig. 2).

A progressive respiratory alkalosis contributed to a mean increase in pH of 0.09 units from rest to the end of exercise ($p<0.01$). There was an increase in standard bicarbonate ($p<0.05$) but no change in plasma lactate from 5 min of exercise until the end. These results are shown in Fig. 3. An increase arterial oxygen tension ($p<0.05$) over both the resting and 5 min exercise values was evident at 75 min exercise (Fig. 4). This was coincident with a significant ($p<0.01$) hypocapnia resulting from a 27% decrease in the arterial carbon dioxide tension from a mean pre-exercise value of 46.9 Torr, to a mean value of 33.8 Torr at 75 min exercise (Fig. 4). These changes were accompanied by significant increases in blood temperature ($p<0.01$), haematocrit ($p<0.01$) and total plasma protein ($p<0.05$) after 5 min of exercise (Fig. 5).

DISCUSSION

The exercise test used in the study was based on previous assessment of the ability of untrained Thoroughbreds to exercise at intensities around 50% $\dot{V}O_2$max. Two of the horses completed the test without difficulty but the other two horses needed considerable encouragement to exercise for the final 10–15 min. While there was extensive sweat loss apparent from the early stages of exercise, the most substantial change in plasma protein increased by 14%, indicating substantial

loss of fluid from the extracellular fluid. In contrast, the changes in haematocrit during this period were more modest, there being only an 8% increase in PCV. The extensive fluid losses by the horses in the study are probably larger than that in the field, as effective evaporative cooling is difficult to achieve in the laboratory setting. It seems likely that fluid loss was responsible for the progressive decrease in Vs during exercise. While the mean HR during exercise did not change, two horses showed an increase in HR during the final 15 min exercise, indicating difficulty in maintaining Q.

Although a fan was used to help cool the horses during exercise, the extent of evaporative cooling possible was rather limited. Whether fluid loss from the central vascular compartment was involved in those horses that showed fatigue or whether ineffective thermoregulation produced fatigue from the resulting high temperature,[11] was not clear. Hodgson et al.[7] noted that during short term submaximal exercise and high intensity exercise at $\dot{V}O_2$max, the blood temperature at the point of fatigue was similar. Of other possible factors, it is unlikely that decreased muscle glycogen reserves were involved in the fatigue as has been found in longer duration exercise in the horse.[5,20] Plasma lactate concentrations remained relatively constant throughout exercise and mean values were always below the onset of blood lactate accumulation (OBLA).

In contrast to a study in Standardbred horses exercising at a lower intensity for 90 min on a treadmill,[16] there was evidence of alveolar hyperventilation in horses in the present study. Over the 75 min exercise there was a 25% decrease in $PaCO_2$, with the most pronounced changes between 15 and 30 min, and 60 and 75 min exercise. As there was no change in standard bicarbonate or plasma lactate during the exercise period, the hyperventilation is unlikely to have been due to chemoreceptor stimulation. It seems likely that the hyperventilation was a response to the high temperatures during exercise, resulting in a respiratory alkalosis. An increasing minute ventilation was found throughout exercise, in the study of Rose and Evans,[16] although there was no change in $PaCO_2$ or PaO_2. However, in that study horses exercised while wearing a respiratory gas collection mask with valves. Such a mask was shown to inhibit respiratory frequency and result in higher $PaCO_2$ and lower PaO_2 tensions than when exercise was undertaken without a mask.[5] In the present study, a "flow through" system of gas collection was utilised, which we had shown did not affect arterial blood gas tensions when flow rates in excess of 6000 l min^{-1} were used (Knight and Rose, unpublished data). The mean decrease in $PaCO_2$ of about 10 Torr from the beginning to the end of exercise, coincided with a similar increase in PaO_2.

From calculations using the ideal gas equation, there was a similar increase in alveolar oxygen tension. Hodgson et al.[7] also reported an elevation of PaO_2 in Thoroughbred horses exercising for a shorter duration at an exercise intensity of 65% $\dot{V}O_2$max.

While there was no significant change in $\dot{V}O_2$ throughout exercise, the non-significant decrease in $\dot{V}O_2$ at 75 min may have been due to the extra support that the horses received at this stage. Two of the horses required traction on the lead rope over the last 5 min exercise to permit completion of the exercise test with the likely result of a decrease in $\dot{V}O_2$. The decrease in $\dot{V}CO_2$ contributed to a decrease in R values, which reached their lowest mean value at 45 min exercise. At the beginning of exercise, the R values indicated mixed carbohydrate and fat metabolism, with fats playing a more important role as exercise progressed. This progressive increase in fat utilisation during sustained exercise is similar to findings in man[1] and horses exercising at lower exercise intensities.[14] Pagan et al.[14] found that the decrease in R values during submaximal exercise was dietary dependant and that the maximum decrease in R was found after 60 min exercise. In their study, horses receiving a high fat or high protein diet showed a much more substantial decrease in R values than those

on the control diet during 90 min exercise at about 25–30% VO_2max. The current study did not attempt to investigate the effect of diet, but our horses received a diet containing about 12% crude protein, which is similar to the control diet in the study of Pagan et al.[14] The interrelationship between diet, exercise intensity and changes in R values awaits further investigation.

Using the VO_2 and VCO_2 to calculate energy utilisation in megajoules (MJ) and ignoring the probable small loss of urinary and sweat nitrogen, the following energy expenditure can be estimated from the equation: Total work (kJ) = 15.8 $\dot{V}O_2$ + 4.86 $\dot{V}CO_2$, where the $\dot{V}O_2$ and $\dot{V}CO_2$ are in l min^{-1}.[19] When this equation is applied to each period of exercise, the average energy utilisation per minute is about 600 KJ, which is a 40-fold increase over resting energy use. The total energy use during the 75 min exercise is, therefore, about 78.9 MJ of digestible energy (DE), assuming an efficiency of utilization of DE of 57%.[13] When this is translated into National Research Council (NRC) nutritional requirements[12] and the maintenance energy guidelines (70 MJ DE per 500 kg horse) are added, there is a total daily DE requirement of 148.9 MJ DE per horse, an increase over maintenance of 112%.

In summary, horses undergoing prolonged submaximal exercise utilise substantial energy reserves, which after 75 min exercise at 50% VO_2max represent 112% increase over daily maintenance DE requirements. A progressive increase in fat utilisation was found in the current study, with R values reaching their lowest mean values at 45 min exercise, which contrasts with the lowest R values in the study of Pagan et al.[14] at 60 min exercise. While hypercapnia is found at high exercise intensities, hypocapnia is evident during submaximal exercise, due to the probable involvement of the respiratory system in thermoregulation. Associated with substantial fluid losses during submaximal exercise and movements of fluid from the central vascular compartment, there was a decrease in Vs towards the end of exercise. It appears that the major limiting factor to short term submaximal endurance exercise may be associated with plasma volume depletion and difficulties in maintaining thermoregulation.

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Influence of Diet on Substrate Metabolism during Exercise

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ABSTRACT. Six horses were fed a normal (N-diet), a fat rich (Fat-diet) or a carbohydrate rich (CHO-diet) diet, each for 5 weeks. The horses performed a standardised exercise tolerance test (SET) and a submaximal exercise test to fatigue (SEF) on a treadmill during the last week on each diet. Blood samples were taken in connection with SET and SEF and muscle biopsies in connection with SEF. The speeds producing a blood lactate value of 4 mmol l^{-1} (V_{La4}) and a heart rate of 200 bpm (V_{200}) were within the normal ranges on all diets during SET. Diet had no influence on the duration of SEF. The average durations of exercise of horses fed N-, CHO-, and Fat-diets were 56, 51 and 52 min, respectively. Muscle triglycerides varied 5-fold among horses irrespective of diet. Resting muscle glycogen was 12% higher in CHO-diet compared to N- and Fat-diets. In all diets, glycogen decreased during SEF with a marked depletion in type I and IIA fibres at fatigue. Muscle lactate and glucose-6-phosphate levels were higher at the end of exercise in CHO- and Fat-diet than in N-diet period. Plasma concentrations of free fatty acids (FFA), glucose, lactate, ammonia, alanine, branched-chain amino acids, tyrosine and tryptophan increased during SEF. Higher concentrations of glucose, lactate, ammonia, alanine and branched-chain amino acids were seen at end of exercise in CHO- and Fat-diet than in N-diet period. At the beginning of exercise FFA concentration was higher in CHO-diet compared with N-diet. The results indicated that dietary manipulations could affect glycogen storage and change the substrate utilisation during submaximal exercise. These changes did not appear to affect time to fatigue during this type of exercise.

Key words. Horses; diet; exercise test; muscle glycogen; plasma metabolites

INTRODUCTION

Carbohydrates, especially glycogen, in skeletal muscle of man and horse have been shown to be an important energy source during various intensities of exercise.[4,10,14,26] During submaximal exercise in man, fatigue is associated with glycogen depletion and a close relationship is observed between initial glycogen content and work time.[10] A shorter time to fatigue during submaximal exercise has been observed in horses as in humans when the glycogen level in muscle has been markedly lowered.[25] Lipids also contribute to energy utilization during submaximal exercise.[4,6] From a study on rats, it was suggested that an increased availability of free fatty acids (FFA) in blood may have a glycogen sparing effect, thus delaying the time to fatigue during prolonged running.[8] Protein, as an energy source during exercise, is known to be of minor importance. It has been estimated that under normal conditions the metabolism of protein and amino acids contributes less than 10% to the total energy yield during exercise in human subjects.[13] Protein and amino acid metabolism has been suggested to have other important functions, e.g. to serve as a metabolic link