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Cardio-Respiratory and Muscle Metabolic Responses to Draught Work on a Treadmill in Standardbred Horses

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Summary

Cardio-respiratory and muscle metabolic responses to standardized draught work on a treadmill were studied at a slow trotting speed (4.8 m/sec) until the horses could no longer lift weights from the ground. Ten horses performed incremental draught work in which weights were added every two minutes to a rope which ran over a pulley and was connected horizontally to the harness. Heart rates increased linearly to a mean of 209 bpm and blood lactates exponentially to a mean of 10.5 mmol/l with increasing weight loads. Lactate levels increased significantly and glycogen and creatine phosphate levels decreased in both m. gluteus and semitendinosus, and adenosine triphosphate (ATP) levels decreased in m. semitendinosus only. The ATP level in the semitendinosus muscle at the end of exercise was significantly negatively correlated to the percentage of type II fibers. In conclusion, for slow trotting of short duration, glycolysis with lactate accumulation and phosphagen breakdown seem to be important pathways for energy supply in muscles that are involved during exercise with incremental draught loading.

Lactate; adenosine triphosphate; creatine phosphate.

Introduction

Draught work is used as a method of training Standardbred racehorses in the Nordic countries. Many trainers are of the opinion that the strength of horses can be increased by use of draught work in their training program. Draught work is also often used after horses have had injuries or surgery when conventional training at higher speeds has to be avoided due to stress on joints, ligaments and tendons. The specific metabolic response induced by pulling against increasing resistance and the training effects gained from this are poorly understood. It was therefore of interest to study cardio-respiratory and muscle metabolic responses using a model in which draught work was performed under standardized, measurable and reproducible conditions as close as possible to the way it is performed on the track.

Material and Methods

The 10 Standardbred trotters (4 mares, 1 stallion and 5 geldings), aged between 4 and 13 years, had previously been used as racehorses, but had no intensive training during the last months preceding the investigation. Before the test each horse was examined and all were found to be clinically healthy. Furthermore, before the test, 8 of the horses were subjected to a standardized exercise tolerance test on a treadmill (Sikob, Sollentuna, Sweden) at a 6.25% slope with consecutively increased velocities from 6–9 m/sec for two minutes on each. The total red cell volume (CV) was determined by the technique of Persson (1967). This was used as an indicator for the oxygen transport capacity of the circulatory system. Based on this assessment none of the horses showed reduced work performance (Persson 1967, 1983.)

The treadmill draught loading test. In each test incremental draught loading was induced on a flat treadmill by addition of weights to a rope which was horizontally connected via a pulley to the harness. The pulley was mounted on a support behind the treadmill. The treadmill was run at a mean speed of 4.8 m/sec, and the draught resistance of 4.1 kp at the start of the experiment was incrementally increased by 20.6 kp every 2 minutes until the horse could no longer be motivated to lift the weights from the ground. If a horse lasted less than 30 sec with the heaviest weight load it was considered to have completed only the previous weight loading period. However, if it lasted for more than 30 sec it was considered to have completed that weight load. Before the test the draught resistance was measured by means of a force transducer fastened between the harness and the rope. The force transducer (strain gauge, type KRG-4, Bofors Electronics, Sweden) was coupled to a transducer indicator (type BKI-1, Hewlett-Packard, Moseley Division) from which the output signal was scaled, low-pass filtered, and recorded by stripchart recorder (Moseley Model 680 M, Hewlett-Packard).

Electrocardiogram, blood samples, respiratory rates and stride lengths. Heart rate (HR), blood lactate (LA) and respiratory rate (RR) was recorded at rest, after 2 minutes at each weight load and 0, 2, 5, and 10 minutes after the cessation of work. The HR was monitored by the use of a bipolar electrocardiogram (Mingograph type 804, Siemens Elena, Solna, Sweden). The venous blood lactate concentration was determined enzymatically (Boehringer Mannheim, Combination No. 124842). The time taken by the horse to run 50 paces was estimated at the end of each weight load in order to calculate the mean stride length. Creatine kinase (CK) was estimated from a blood sample taken before the test and 24 hours after. The electrocardiogram (ECG) recordings were made 24 hours before and after each experiment.

Muscle samples. Muscle biopsies were taken before and within 10 sec of each test through the same incision from the right gluteus medius in all horses and from the left semitendinosus in 8 of the horses by the technique of Lindholm and Piel (1974). M. gluteus was biopsied about 10 cm dorso-caudally to trochanter major on tuber coxae, m. semitendinosus at a point midway between the tuber ischii and the flexor aspect of the stifle in the midline of the muscle. The muscle samples for biochemical analysis were immediately frozen in liquid nitrogen while those for histochemistry were rolled in talcum powder before they were frozen. The samples were stored at −80°C until analyzed.

Histochemistry. Transverse serial sections were cut in a cryostat microtome at −20°C to produce cross sections of 10 μm. These were stained for myofibrillar ATPase after alkaline (pH 10.3) and acid (pH 4.3 and 4.6) preincubation to identify the fiber type (Brooke and Kaiser, 1970). The fiber composition of each biopsy was established by counting at least 200 fibers.

Biochemistry. The muscle biopsies were freeze-dried. Blood and connective tissue were removed under a dissection microscope and the samples were weighed. Analysis was made of glycogen by boiling part of the muscle tissue for 2 hours in 1 ml of 1 M HCL in order to hydrolyze the glycogen into glucose residues. Glucose, and after perchloric acid extraction lactate, creatine phosphate (CP), glucose-6-phosphate (G-6-P) and adenosine triphosphate (ATP) concentrations were determined fluorometrically by the technique of Lowry and Passonneau (1973). The weighed samples for enzyme analysis were homogenized with an ultrasonic disintegrator in cooled 0.1 M potassium phosphate buffer (pH 7.3). The activities of citrate synthase (CS), as a measure of citric acid cycle activity, lactate dehydrogenase (LDH) as a measure of glycolytic capacity and 3-hydroxy-acyl-CoA dehydrogenase (HAD) as a measure of the capacity for lipid oxidation were determined at 25°C using fluorometric techniques. The assay mixtures described by Essén *et al.*, (1980) and by Essén-Gustavsson *et al.*, (1984) were used.

Statistical analysis. The data obtained were analyzed by the t-test for paired observations, by linear and exponential regression and by analysis of variance, using a randomized block design.

Results

The maximal weight load lifted varied between the horses (65.9–107.1 kp). Heart rates increased linearly with increasing weight loads and reached a mean of 209 ± 13 bpm (Fig. 1). The weight loading producing a HR = 200 bpm (W 200) was on average 75 ± 11.6 kp and was attained after 8–10 minutes of treadmill exercise. The respiratory rates did not change further after the initial rise to a mean of 92 ± 14. The blood lactate increased exponentially with increasing weight loads (Fig. 1). The blood lactate varied between 5.2 and 16 mmol/l immediately after exercise.

The red cell volume determined in connection with the standardized treadmill exercise tolerance test averaged 30.5 ± 5.91 1. The cell volume was significantly correlated to W200 ($r = 0.75$) and also to the weight load producing a lactate concentration of 4 mmol/l (WLA4), ($r = 0.73$). The WLA4 indicates the weight load inducing a blood lactate accumulation of 4 mmol/l and was on average 64.6 ± 14.2 kp. The stride length did not change significantly, and after the first 2 minutes was 2.85 ± 0.29, and at exhaustion, 2.76 ± 0.27 meters. The CK increased significantly ($P < 0.02$) from a mean of 3.7 ± 1.2 μkat/l before the test to a mean of 5.9 ± 1.5 μkat/l 24 hours after the test. No effect of draught work on ECG could be detected.

Muscle fiber distribution. The fiber distribution for the two different muscles and the enzyme levels are shown in Table 1. There were no differences in fiber distribution, LDH, or HAD activity between the two muscles but the CS activity was 37% higher, on average, in m. gluteus than in m. semitendinosus.

Substrate and metabolites. The substrates and metabolites in the muscles are shown in Fig. 2. Glycogen was significantly decreased by a mean of 84.2 mmol/kg in gluteus and by 126.6 mmol/kg in semitendinosus. Lactate increased significantly by a mean of 48.8 mmol/kg in m. gluteus and by 73.9 mmol/kg in m. semitendinosus. The lactate level at the end of work was significantly higher ($P < 0.02$) in m. semitendinosus than

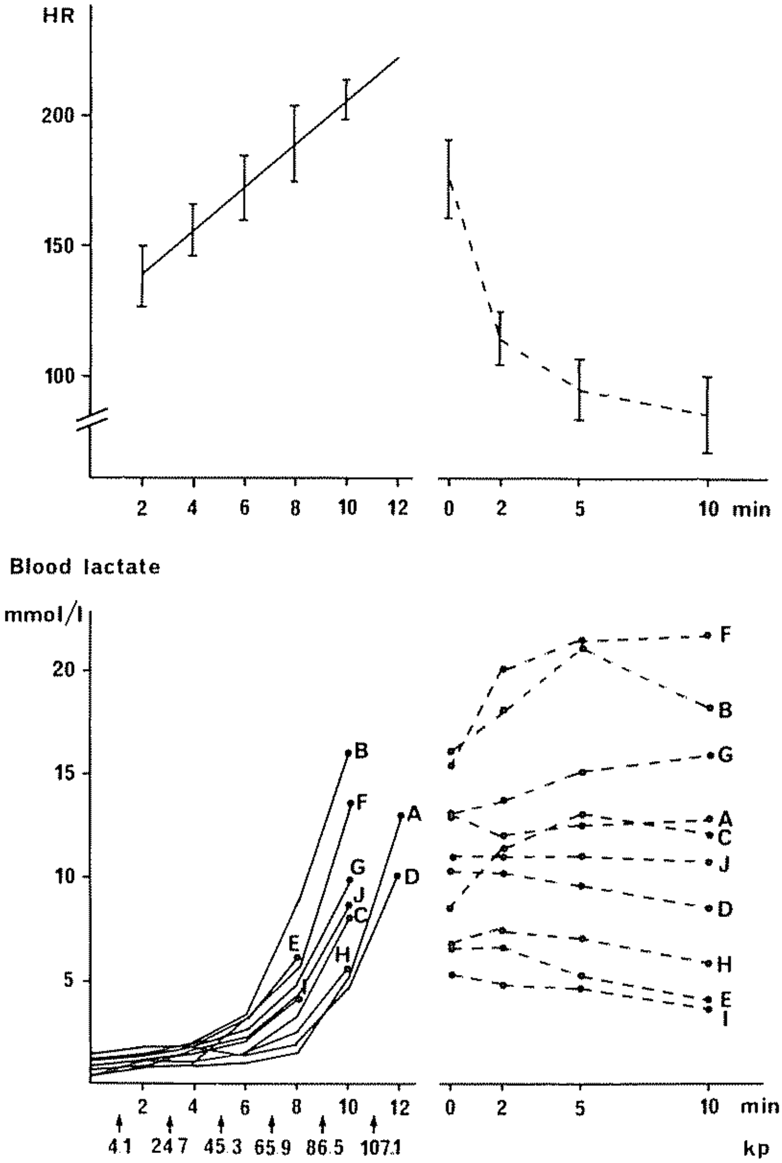

FIGURE 1 The heart rate and blood lactate response to increasing weight resistance. n = 8 at 10 minutes, n = 2 at 12 minutes. HR = heart rate, kp = kiloponds.

in m. gluteus. The highest recorded muscle lactate value was 123 mmol/kg in gluteus, and 201 mmol/kg in semitendinosus and the lowest 34 and 36 mmol/kg, respectively. There were significant correlations between the blood lactate at the cessation of work and the muscle lactates produced by semitendinosus and gluteus (r = 0.80 and r = 0.88, respectively).

The G-6-P increased significantly in both muscles with a mean of 7.6 mmol/kg in

TABLE 1. Fiber composition and enzyme activities in m. gluteus and m. semitendinosus

	Fiber composition (%)			Enzyme activities (mol/g d.w./min)		
	I	IIA	IIB	CS	HAD	LDH
m. gluteus (n = 10)						
mean	17	47	36	35*	20	1188
± SD	5	11	13	12	5	491
m. semitendinosus (n = 8)						
mean	13	47	40	22	16	1126
± SD	4	8	10	6	6	446

CS = citrate synthase; HAD = 3-hydroxyacyl-CoA; LDH = lactate dehydrogenase.
* = significant difference between m. gluteus and m. semitendinosus ($P < 0.05$).

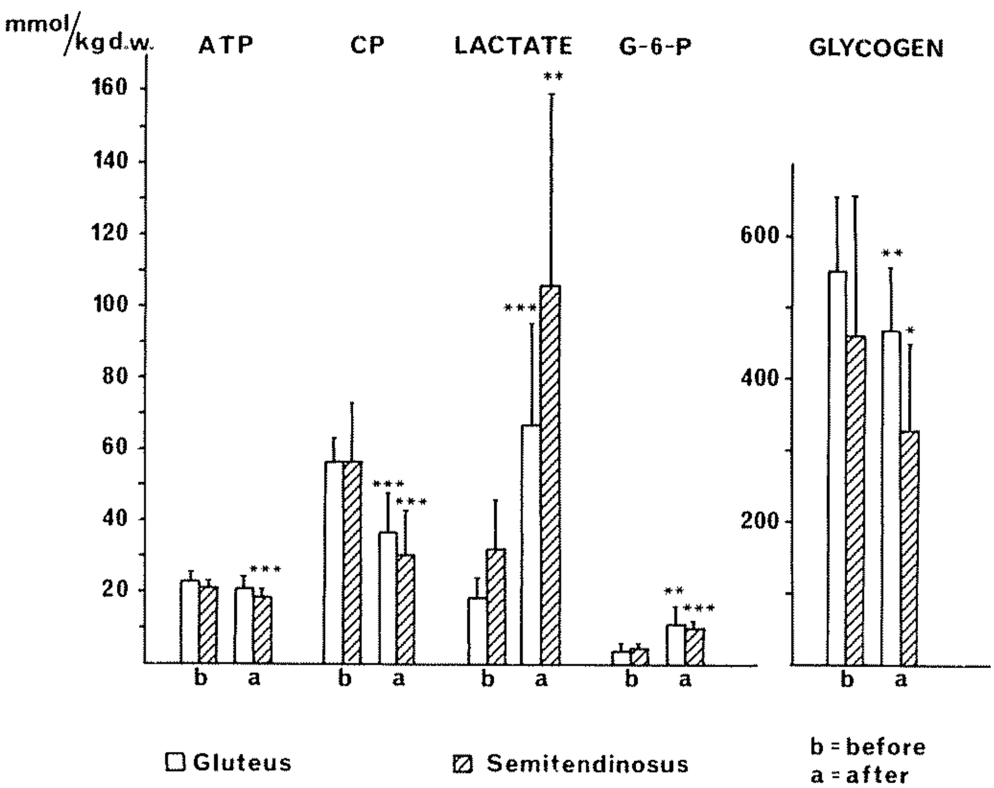

FIGURE 2. Substrates and metabolites in m. gluteus and m. semitendinosus before and after exercise. * = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$. ATP = adenosine triphosphate, CP = creatine phosphate, G-6-P = glucose-6-phosphate

gluteus and 5.6 mmol/kg in semitendinosus. The CP level was significantly decreased in both muscles with a mean of 19.7 mmol/kg in gluteus and 26.4 mmol/kg in semitendinosus. The ATP level was significantly decreased in m. semitendinosus only with a mean decrease of 2.88 mmol/kg. The ATP level in the semitendinosus muscle at the end of exercise was significantly negatively correlated to the percentage of type II fibers ($r = 0.73$) (Fig. 3). The mean increase in G-6-P and lactate and the mean decrease in glycogen and CP in the gluteus and semitendinosus muscles did not significantly differ. ATP levels were significantly lower in the semitendinosus muscle than in m. gluteus.

Discussion

This study shows that the energy contribution is provided not only through oxidative processes but also to a great extent through glycolysis with lactate formation and phosphagen breakdown during incremental draught-loading at a slow trotting speed. In contrast, when similar treadmill work is performed, but without the addition of weights, only small increases in HR and blood lactate responses are seen, indicating that energy is mainly provided by oxidative processes (unpublished observation). Weight-loaded treadmill exercise is not readily understood from an ergonometric point of view. The horses were trotting at a constant speed pulling the rope and thereby lifting the weights from the ground. This might have caused a slight change in locomotion pattern compared with ordinary draught work on the track. The horses stopped exercise when it was no longer possible to stimulate them to lift the weights from the ground. It was notable that the horses pulled different weight loads at the end of exercise and showed

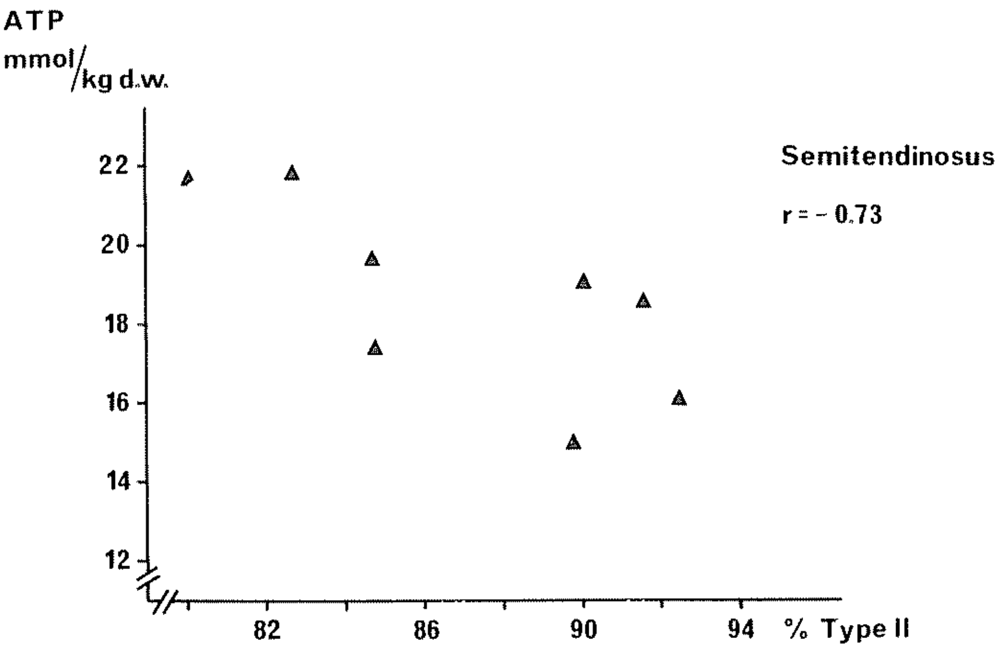

FIGURE 3. Adenosine triphosphate after exercise in relation to the percentage of type II fibers in the semitendinosus muscle. ATP = adenosine triphosphate.

different metabolic responses in blood and muscle. It is important in this connection to remember the psychological element which, in combination with physiological factors, determine the limit of maximal performance. Varying ambition probably accounted for some of the differences in weight lifting ability.

The red cell volume is considered to be an expression of the oxygen transport capacity of the cardiorespiratory system of the horse (Persson, 1967). This means that the positive relationship between red cell volume on one hand and W200 and WLA4 on the other indicates that oxygen transport capacity is of significance also for draught work.

Although the blood lactate responses differed between the horses, the mean level found after exercise was high and similar to what has been found without draught load at a speed of 10 m/s in untrained Standardbred trotters (unpublished observation) or with incrementally increased speeds on a 6.25% sloped treadmill (Thornton *et al.*, 1983). In most of the horses the draught work produced high lactate and low phosphagen levels in both gluteus and semitendinosus muscles. Some horses showed very high muscle lactate and low phosphagen levels similar to what can be found after racing in Standardbred trotters (Valberg, S. Personal communication). The lactate content was significantly higher in m. semitendinosus than it was in m. gluteus, although the percentage of the different fiber types did not differ between the two muscles. Therefore, different fiber recruitment or different metabolic capacities within the muscle fibers may explain the different metabolic responses. The CS activity was 37% higher in the gluteus than the semitendinosus muscle. This may indicate that energy was supplied to a greater extent through oxidative processes than by glycolysis with lactate production or phosphagen breakdown in the gluteus muscle. It was also evident that a significant decrease in ATP occurred in the semitendinosus muscle. In a recent study on man low muscle ATP and high lactate levels were also found after heavy-resistance exercise (Tesch *et al.*, 1986). Although the CK values were slightly increased after the draught work there was no indication of muscle damage as the increases were within the normal limits.

From investigation of glycogen depletion patterns it has been shown that mainly type I and a small portion of type IIA fibers are recruited at lower trotting speeds and that the fibers are recruited in the sequence type I, IIA, and IIB with increasing trotting speeds (Valberg, 1986). In spite of the low speed in this study the addition of weights gives rise to an increased demand for force in order to sustain the draught load. It is thus likely that fast-contracting type II fibers were recruited in both the gluteus and semitendinosus muscles. This is supported by the high muscle lactate and low phosphagen levels found after the exercise. Type II fibers have a higher glycolytic capacity than type I fibers in the horse (Snow, 1983, Valberg and Essén-Gustavsson, 1985) and this is also found in other animals. Furthermore, the correlation found between a low muscle ATP, on one hand, and a high percentage of type II fibers in m. semitendinosus, on the other, indicate that type II fibers have been recruited. This is in agreement with a recent study where a similar relation was found between type II fibers and ATP levels in m. gluteus after racing (Essén-Gustavsson and Valberg, 1987) and a further study which found the lowest muscle ATP levels post race in pools of type II fibers (Valberg and Essén-Gustavsson, 1987). The ATP depletion and the metabolic acidosis following high lactate levels are often suggested as causes of fatigue (Harris, 1985). As the metabolite analyses were made on a biopsy of whole muscle they do not show the content of metabolites in the individual muscle fibers. The lactate content may therefore have been very high in some fibers and ATP very low or even depleted.

In conclusion, glycolysis with lactate accumulation and phosphagen breakdown seem to be important pathways for energy supply in muscles that are involved during exercise with incremental draught loading. In spite of pulling the same weight the horses showed different metabolic responses. Further knowledge is therefore needed about muscle fiber recruitment patterns and metabolic responses during draught work. As seen from the results, it is possible that weight-loaded exercise is a method by which all fiber types may be recruited at a low velocity.

Acknowledgments

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