

NITROGEN UTILIZATION AND METABOLIC RESPONSES OF HORSES
TO INTENSE ANAEROBIC EXERCISE

by

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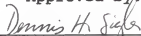
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CHAPTER 1
INTRODUCTION

Nutrient requirements of race and performance horses are currently of interest to many people, yet research to date on the protein requirements of working horses is very limited. Furthermore, the current recommendations regarding protein intake for exercising horses are based on conclusions from a small number of experiments. The National Research Council (NRC, 1978) has recommended that the exercising horse does not require additional dietary protein above the maintenance level of .63 kg of crude protein/d based on studies done in draft horses (Harvey et al., 1939) and in endurance trained horses (Slade et al., 1975). However, recent studies done by Freeman et al. (1985a) indicate that nitrogen retention tends to increase during strenuous exercise. If this is true, increasing protein intake in horses doing strenuous work might enhance athletic performance.

In order to maximize the horse's potential to perform a thorough understanding of the relationship of nutrition to exercise is needed. Research in human exercise physiology has shown that muscle mass and strength increase as a result

Citations in this thesis follow the style of the Journal of Animal Science.

of anaerobic weight training due to increased protein synthesis and muscle fiber hypertrophy (Gollnick et al., 1973; Consalazio et al., 1975; Fox et al., 1988). If this is true in the horse, increased protein intake might increase nitrogen retention and be utilized as muscle building material.

Because so many horses are used in cutting, roping, racing and other anaerobic-type events it is important that the nutrient requirements of the horse are being met and to know the best training strategies for maximizing performance. Therefore, it is necessary to study protein utilization during exercise and during different training regimens.

The objectives of this study were to investigate the effect of intense anaerobic exercise on nitrogen metabolism in horses, and to determine the metabolic adaptations to this exercise.

Chapter 2

REVIEW OF LITERATURE

Heart Rate in Response to Exercise

Heart rate is used as an indicator of physical condition in both humans (Karpovich, 1965; Morehouse, 1976; Astrand and Rodahl, 1977; McArdle et al., 1986; Fox et al., 1988) and in horses (Asheim, 1970; Rodiek, 1982; Erickson et al., 1985). A reduced resting heart rate due to training is rarely observed in the horse (Cardinet et al., 1963; Sigler, 1981; Erickson et al., 1985; Sexton et al., 1985; Drozd et al., 1987; Foreman et al., 1987). However, Stewart (1972) found non-significant trends toward lower resting heart rates in Thoroughbred racehorses after a training regime. Likewise, Burke et al. (1981) found a nonsignificant trend in reduced resting heart rates for 2 and 3 yr old Quarter Horse fillies in training for 30 d. Although resting heart rates are not usually affected by conditioning, exercising heart rates are. Untrained individuals have higher heart rates than trained athletes (Karpovich, 1965; Morehouse, 1976; McArdle et al., 1986; Fox et al., 1988).

Maximum heart rate in the horse appears to be around 240 beats/min (Lindholm and Saltin, 1974) which is much higher than that observed in other species (Asheim, 1970; Lindholm and Saltin, 1974). According to Morehouse (1976) and McArdle et al. (1986), the average heart rates of humans during

maximal exercise decreases as a human ages. Asheim (1970) found large variations in maximum heart rates of horses but found no relationship between maximum heart rates and age or size of the horses.

Heart rate can be affected by many factors including age, sex, level of fitness, temperature and humidity, as well as the duration and intensity of exercise (Karpovich, 1965; Morehouse, 1976; McArdle et al., 1986; Fox et al., 1988). Heart rate, in the horse, is increased during different types of work (Asheim, 1970; Lindholm and Saltin, 1974; Thomas and Fregin, 1981; Miller et al., 1985; Topliff et al., 1985) and is affected by both the intensity and duration of exercise (Lindholm and Saltin, 1974). Lowered heart rates during exercise can be accomplished by submaximal training in humans (Karpovich, 1971; McArdle et al., 1986; Fox et al., 1988) and in horses (Asheim, 1970; Erickson et al., 1985; Sexton et al., 1985; Drozd et al., 1987).

In horses a clear pattern of heart rate response to submaximal exercise is often observed. There is an immediate rise in heart rate followed by a plateau if the workload remains constant (Lindholm and Saltin, 1974; Thomas and Fregin, 1981). However, a reduction in the height of the plateau can be accomplished by conditioning in humans (Clausen, 1970) as well as in horses (Asheim, 1970).

The time required for heart rates to return to normal depends on the workload of the exercise period, the duration

and intensity of the exercise and the physical condition of the subject. In persons of good physical condition, recovery occurs more rapidly than in poorly trained subjects (Karpovich, 1965; Morehouse, 1976; Fox et al., 1988). Banister and Purvis (1968) discovered that recovery heart rates did not indicate the level of fitness in horses doing less strenuous work. In addition, Witherington (1971) found that the climate had an effect on the recovery rates of horses. However, in competitive trail horses Cardinet et al. (1963) found that heart rate recovery was faster in conditioned than unconditioned horses. Similarly, Sexton et al. (1985) found that ponies subjected to an 8 wk training regimen had significantly faster recovery rates after a 25 min treadmill test. Furthermore, Erickson et al. (1985) found that there was a significant difference in recovery rates after a 174 d training program in 2 and 3 yr old Quarter Horses. However, Milne et al. (1977) found no significant difference in heart rate recovery values in horses who had undergone a 92 d conditioning program.

Respiration Rate in Response to Exercise

Respiration rate is affected by many factors, including climate, lactic acid concentration in the blood, and tidal volume (Cardinet et al., 1963; Fox et al., 1988). Respiration rate increases when exercising in humans (Fox et al., 1988) and in horses (Milne, 1976; Snow and MacKenzie, 1977a;

Campbell et al., 1985; Topliff et al., 1985; Drozd et al., 1987). In inefficient, untrained human subjects the frequency of respiration is higher the trained subjects (Karpovich, 1971; McArdle et al., 1896; Fox et al., 1988). However, Cardinet et al. (1963) has pointed out that endurance horses after a ride had much variability in the rate and depth of respiration but breathing became more shallow as the respiration rates increased. It was also noted that for horses able to complete the endurance ride there were greater variations in respiration rates when compared to the heart rates. In addition, Hinton (1978) found that in endurance horses it is more practical to measure respiration rate 20 to 30 min after exercise in order to determine exhaustion. However, Kelly (1977) found that heart rate during recovery is a more appropriate measure of assessing fitness because respiration rate was greatly varied when ambient temperature was high. In addition, Stewart (1972) noted that in Thoroughbred racehorses after 10 to 20 min of recovery from a galloping test, respiration rates became very irregular. Snow and MacKenzie (1977a), Sigler (1981), Drozd et al. (1987) and also reported variations in respiration rate during exercise and recovery, and found no significant changes in respiration rates in horses that had been conditioned. Therefore, because respiration rates are affected by such a wide variety of factors they are not a reliable indicator of fitness (Cardinet et al., 1963; Johnson et al., 1987).

Blood Lactic Acid Concentration in Response to Exercise

Both anaerobic and aerobic energy metabolism systems contribute energy during exercise in humans. Their relative roles are dependent upon type of exercise performed, state of training and diet of the athlete (Fox et al., 1988). Energy for short duration, high intensity exercise is supplied by the anaerobic system, while energy for low intensity, long duration exercise is supplied by aerobic metabolism (Morehouse, 1976; Astrand and Rodahl, 1977; Fox et al., 1988). During the first few minutes of moderate intensity exercise, anaerobic metabolism supplies energy until aerobic metabolism can take over and cover the energy demand (Astrand and Rodahl, 1977; Fox et al., 1988).

Blood lactic acid is an end product of anaerobic metabolism (Morehouse, 1976; Astrand and Rodahl, 1977; Fox et al., 1988). In the first few minutes of low or moderate intensity exercise, blood lactate levels rise and then reach a plateau which will remain constant for the duration of exercise (Astrand and Rodahl, 1977; Fox et al., 1988).

Blood lactic acid in response to exercise is used as an indicator of fitness in humans (Karpovich, 1965; Karlsson and Saltin, 1970; Donovan and Brooks, 1983; Fox et al., 1988) and in horses (Asheim, 1970; Lindholm and Saltin, 1974; Snow and MacKenzie, 1977b; Keenan, 1979; Thomas and Fregin, 1981;

Erickson et al., 1985; Sexton et al., 1985; Topliff et al., 1985).

Blood lactic acid is dependent upon the intensity and duration of the exercise (Karpovich, 1965; Astrand and Rodahl, 1977; Fox et al., 1988). As the intensity of exercise increases lactic acid builds up in the blood and the muscle (Morehouse, 1976; Fox et al., 1988). In addition to the accumulation of blood lactic acid as the exercise intensity increases, oxygen consumption increases as well. Yet the level of blood lactate does not begin to accumulate until 55% of the healthy, untrained persons's maximal capacity for aerobic metabolism (McArdle et al., 1986; Fox et al., 1988). At this point, the blood lactate concentration becomes nonlinear with oxygen consumption; this point is called the anaerobic threshold. The anaerobic threshold is defined by Fox et al. (1988) as the workload intensity or oxygen consumption at which anaerobic metabolism is accelerated. The anaerobic threshold is better defined, however, by McArdle et al. (1986) as being the point at which the formation of lactic acid exceeds its rate of removal via oxidation in the Krebs' Cycle or its resynthesis to pyruvate in the liver.

Most research in humans reveals that blood lactate concentrations during or after an exercise bout are decreased due to training (McArdle et al., 1986; Fox et al., 1988). Drozd et al. (1987) found that Quarter Horses which were aerobically trained for 84 d had lower blood lactate

concentrations during and after a standard exercise tolerance test on a treadmill. Similar results were discovered by Webb et al. (1985) in Quarter Horse mares. Erickson et al. (1985) and Sexton et al. (1985) found similar results indicating that blood lactate in response to standard exercise tolerance tests on a treadmill is lower during exercise and recovery in conditioned versus the unconditioned horse. It must be noted, however, that Erickson et al. (1985) and Drozd et al. (1987) noted no significant changes in resting blood lactate concentration between conditioned and unconditioned horses. In humans it also has been suggested that resting blood lactate concentration does not change in response to conditioning (McArdle et al., 1986; Fox et al., 1988).

It also has been reported by Snow and MacKenzie (1977) after a 10 wk conditioning program on Thoroughbreds and heavy hunters that there were no changes in blood lactate values after the conditioning. Similar results were discovered by Johnson et al. (1987) on ponies conditioned by pulling a weighted sled for 6 wk and then subjected to a standard exercise tolerance test on a treadmill.

Protein Metabolism in Response to Exercise

Protein has many functions in the body including building and maintaining structural units of the body (bone, cartilage, connective tissue, blood vessels, muscle, hair and hooves); utilization in the formation of enzymes, hormones, immune

antibodies and hereditary material (Cunha, 1980; Brooks and Fahey, 1984); and it is used as an energy source when fats and carbohydrates are in short supply (Stegemann, 1981; Fox et al., 1988).

Reseachers have tried to determine whether the working horse requires higher levels of dietary protein than sedentary horse. Often, equine exercise physiologists look to human exercise physiology for answers to their questions. Briggs and Calloway (1979) state that since exercise tends to lead to increased strength and muscle size, additional protein may be required to aid in the building of muscle tissue. However, Bogert et al. (1973) observed that while muscular work is the largest single factor in determining energy needs, it has no appreciable effect on protein requirements except during initial periods of training when muscular tissue is developing. Similarly, Gontzea et al. (1975) suggests that protein intake should be increased for a few days following that start of a training regimen or an increase in training intensity. However, not all researchers feel dietary protein is needed to improve the development of muscle tissue or improve athletic performance. Crampton (1964) reported that increasing protein levels above maintenance as work increases had no beneficial effect on fitness of performance. Furthermore, Consalazio et al. (1975) found that additional body protein did not enhance exercise performance but did increase muscle mass and body protein stores.

Because protein is an integral part of many foods, protein intake generally increases with greater total food consumption for both humans and horses. A survey conducted by Winter and Hintz (1981) it was indicated that Thoroughbred racehorses consume approximately 2 1/2 times more crude protein than the National Research Council (1978) recommendation, due to their increased high protein feed. Hinkle et al. (1981) point out that many horse owners routinely feed much higher levels of protein to working horses than what is required for maintenance. Cunha (1980) states that most excess protein is deaminated through the urea cycle and is excreted while some serves as an energy source or is converted to and stored as fat through complex body mechanisms.

Researchers have been concerned about the loss of nitrogenous compounds in sweat during exercise. Bogert et al. (1973) and Brooks and Fahey (1984) point out that nitrogenous compounds are lost in urine, feces, sweat, semen, menses, phlegm, hair and nail clippings. These types of nitrogenous losses are often not accounted for in nitrogen balance studies. Consalazio et al. (1975) believe that the loss of nutrients through sweat is relevant in determining requirements, particularly when sweating is increased; therefore, decreasing the accuracy of human metabolic studies. Furthermore, Calloway et al. (1971) conducted nitrogen balance studies in sedentary and exercising men and found that values

based on only urine and fecal collections were 45% lower than when nitrogen lost in sweat, desquamated cells, nails and hair was accounted for. Yet, some researchers feel that nitrogen losses in the sweat are low and have little effect on nitrogen balance (Bogert et al., 1973; Meyer, 1980). It is known that the horse has an extremely high sweat protein concentration in comparison with other species (Jenkinson et al., 1974; Kerr et al., 1983), and this may effect nitrogen requirements. Hintz (1982) and Slade (1979) realize that nitrogen containing material is lost through sweat in the exercising horse, yet they point out that the increased feed intake needed to supply energy for work generally provides more than an adequate amount of dietary protein.

Protein status of the animal body is determined through the use of nitrogen balance trials. These trials indicate whether the amount of protein metabolized in the body is equal to, less than or greater than the amount of protein in the feed (Bogert et al., 1973; Brooks and Fahey, 1984). Urine and feces are collected and analyzed for nitrogen content. Fecal and urinary nitrogen are added together and subtracted from nitrogen content of the feed to determine the nitrogen status. Bogert et al. (1973) points out that when nitrogen intake and output are approximately equal, the body is in a state of nitrogen equilibrium. A positive nitrogen balance occurs when intake is greater than output, and indicates that new tissue is being synthesized along with nitrogen retention. Nitrogen

deposition occurs when protein intake is very high (Slade et al., 1970; Consalazio et al., 1975; Oodoye and Margen, 1979; Hinkle et al., 1981; Lin and Huang, 1982). Negative nitrogen balance results if output of nitrogen is greater than intake. This may occur if protein intake is less than that needed to maintain body tissues or if the body must burn protein because the diet fails to provide sufficient carbohydrate and fat to meet the energy needs. Bogert et al. (1973) states that a prolonged negative nitrogen balance is detrimental to both growth and/or performance. However, Consalazio et al. (1975) found that persons consuming the recommended amount of dietary protein went into negative nitrogen balance when they began a weight training program, but nitrogen balance was restored within a few days to a week.

The National Research Council (1978) states the recommended level of dietary protein for the horse to be .63 kg crude protein/d regardless of the exercise level. This recommendation is based on research done with endurance trained horses (Slade et al., 1975) and draft horses (Harvey et al., 1939). Slade et al. (1975) found that endurance trained horses fed a diet containing twice the level of dietary protein exhibited profuse sweating and increased heart and respiration rates following a 35 to 40 mile endurance ride. When low and high protein diets were compared it was found that the high protein ration was the least effective in developing stamina. Similarly, Harvey et al. (1939) found

that two draft geldings maintained a positive nitrogen balance when fed a limited amount of oats and timothy hay while doing heavy work. Therefore, it was concluded that the working horse required no additional protein above the maintenance level.

In more recent years, studies concerning the protein needs of exercising horses have been conducted. Hintz et al. (1980) found that feeding 12% and 24% crude protein had no effect on the performance of endurance horses, but was costly and led to a slight decrease in feed efficiency. Thus, the feeding of additional dietary protein was not recommended. Patterson et al. (1985) fed 18 mature horses on three levels of crude protein (5.5, 7.0 and 8.5%) and three levels of physical activity (maintenance, medium and intense work). It was determined that 1.9 g DP/w^{.75} was an adequate minimum protein requirement for the horse regardless of workload provided the horse's digestible energy needs were met. This is much lower than the National Research Council's (1978) recommendation of 2.7 g DP/w^{.75}. Yet, all the research does not support the current National Research Council's protein recommendations. Freeman et al. (1981) and Hinkle et al. (1981) found that in mature stock type geldings, conditioning and nitrogen intake increased nitrogen retention. Freeman et al. (1985a) found that mature horses working for 135 d at a workload of 2800 kg-km/d and a galloping speed of .45 km/min tended to increase nitrogen balance. Slade et al. (1970)

demonstrated increased nitrogen balance resulting from increased intake in unconditioned horses as well.

Chapter 3

NITROGEN UTILIZATION AND METABOLIC RESPONSES OF HORSES TO INTENSE ANAEROBIC EXERCISE

ABSTRACT

Four 3-yr-old Quarter Horse geldings were used to study the effects of intense anaerobic exercise on nitrogen utilization and metabolic responses to three standard exercise tolerance tests: a loaded treadmill test; a sled test; and a 182.4 m acceleration test. Conditioning consisted of pulling a weighted sled for 10 min/d, twice a day, 5 d/wk for 8 wk. Weight was adjusted accordingly to maintain heart rates of 170 to 180 beats/min throughout each work session. All horses were subjected to a 15 min loaded standard exercise tolerance treadmill test (LSET) on 0, 28 and 56 d of conditioning and after 28 d of detraining. On days 0 and 56, horses were subjected to the 10 min sled test and the 182.4 m acceleration test. Parameters measured included heart rate, blood lactate concentration and respiration rate. Split times were recorded for the acceleration test at 91.0 m and 182.4 m. The LSET appears to be an appropriate means of accessing fitness for this type of anaerobic exercise. Blood lactate concentration appears to be an accurate indicator of fitness for the LSET as blood lactate concentration was lower ($P < .05$) on day 56 of conditioning than day 0 at 5, 10 and 15 min of

the test. In addition, horses appeared to maintain cardiovascular fitness through 28 d of detraining. Data from the acceleration test indicate that it is not a good measure of fitness as it was a completely different type of exercise, much shorter in duration and less strenuous than the other standard exercise tolerance tests. However, it must be noted that the anaerobic conditioning did increase ($P<.05$) speed for the first 91.0 m of the acceleration test and tended ($P=.07$) to increase speed from start to the 182.4 m finishing point. Five day nitrogen balance trials were conducted prior to conditioning, at 1 to 5 d of conditioning, 52 to 56 d of conditioning and again for 5 d after 28 d of detraining. Fecal nitrogen excretion increased ($P<.05$) from the first to the third collection period and urinary nitrogen excretion decreased ($P<.05$) in the second collection period and then increased ($P<.05$) in the third collection period. Nitrogen balance was lower ($P<.05$) in the third and fourth collection periods than in the first. Nitrogen absorption did not differ significantly. Nitrogen intake was highly correlated with nitrogen balance. It appears then that nitrogen balance is affected by both nitrogen intake and intense anaerobic training. Since negative nitrogen balance values were seen, horses undergoing intense anaerobic exercise may have an increased need for dietary protein.

INTRODUCTION

A number of horses in today's industry are being used for performance events, therefore an understanding of the relationship of nutrition to exercise is important. Cardiovascular fitness is enhanced through various types of conditioning programs (Sigler, 1981; Erickson et al., 1985; Sexton et al., 1985; Drozd et al., 1987). Generally, as the intensity or duration of the exercise increases, the energy requirements of the horse also increase (National Research Council, 1978). A point that is more controversial, however, is whether or not the horse's protein requirements increase as well. The National Research Council (1978) recommends that the exercising horse does not require additional dietary protein above the maintenance level of .63 kg of cp/d, but research done by Freeman et al. (1985a) indicate that nitrogen retention may be increased in horses performing long term, low intensity exercise. The objectives of this study, then, were to investigate the effect of intense anaerobic exercise on nitrogen utilization in horses and to determine cardiovascular adaptations to this exercise.

EXPERIMENTAL PROCEDURES

Experimental Animals

Four 3-yr-old Quarter Horse geldings with an average weight of 481 ± 32 kg (Appendix A) were conditioned to determine the effects of intense anaerobic work on nitrogen utilization and on metabolic responses to three standard exercise tolerance tests (SET): a loaded treadmill test; a sled test; and an acceleration test. Horses were housed individually in 3.7 x 4.6 m steel pipe pens, except during nitrogen balance trials when they remained tied to prevent coprophagy and to keep movement to a minimum. Horses were conditioned by pulling a weighted sled (Appendix B) across a dirt/sand surface for 10 min/d twice a day, 5 d/wk for 8 wk. Heart rate was monitored throughout the conditioning period by a heart rate monitor (Uniq Heartwatch, Computer Instrument Corp., Hempstead, NY). Weight on the sled was adjusted whenever necessary to maintain a heart rate of 170 to 180 beats/min throughout each 10 min work period.

Feeding and Sample Collection

Horses were fed a complete pelleted diet (Table 1). Horses were weighed weekly and feed intake was adjusted to maintain constant body weight during conditioning. However, feed intake was kept exactly the same for each individual

TABLE 1. COMPOSITION OF EXPERIMENTAL DIETS

Ingredient	Base Diet	Chromic Oxide Diet
	%	
Corn	55	55
Sun cured alfalfa	37	37
Molasses, liquid	7	7
Salt	1	1
Chromic oxide	-	.24

horse throughout the nitrogen balance trials. Chromic oxide was used as an external indicator and was incorporated into the pellet at a level of .24%. Horses were fed twice daily at 12 hr intervals and were allowed ad libitum access to water. After precollection adjustment periods of 10 d, 5 d nitrogen balance trials were conducted prior to conditioning, at 1 to 5 d of conditioning, at 52 to 56 d of conditioning, and again for 5 d after 4 wk detraining. Feed samples were taken at random and stored in plastic freezer bags for later analysis.

Fecal grab samples were collected once daily for each day of the nitrogen balance trials. Daily samples were collected at different pre-determined times during the day to reduce diurnal variation. Samples were stored in plastic freezer bags and frozen for later analysis of chromium and proximate components.

Total urine was collected for 5 d in urine harnesses. Harnesses were cleansed with water once each day to keep urease activity to a minimum. Urine was measured every 3 hr and a 1% subsample was acidified with 6 M Hydrochloric acid and then frozen in an airtight container for later analyses. Horses were tied and remained standing throughout the nitrogen balance trials to prevent spillage of urine and coprophagy. When horses were not being conditioned, they were hand-walked for 10 min/d to prevent stiffness.

Preparation of Samples and Laboratory Analysis

Individual fecal samples were dried at 50 C in a draft oven and were then composited on an equal weight basis into one sample per horse per period. Feed samples were composited in a similar manner. Both feed and fecal samples were ground through a 1 mm screen in a Wiley Mill (model no. 3, Arthur H. Thomas Co., Philadelphia, PA) and stored for later analyses in airtight containers.

Dry matter was determined gravimetrically after drying in a vacuum oven at 102 C for 24 h.

Chromium content was determined by wet ashing followed by atomic absorption spectroscopy (AOAC, 1984).

Ether extract was determined by the Goldfish method as outlined by the AOAC (1984).

The Kjeldahl method of acid digestion followed by titration was used to determine the crude protein content of all samples. AOAC (1984) procedures were followed for the acid digestion, however boric acid was used in the distillation.

Mineral matter was determined by ashing samples in a muffle furnace (AOAC, 1984).

Samples were analyzed in quadruplicate for all constituents measured. Digestion coefficients for dry matter, ether extract, ash and crude protein were determined. Nitrogen balance and nitrogen absorption values were also calculated.

Loaded Standard Exercise Tolerance Treadmill Test

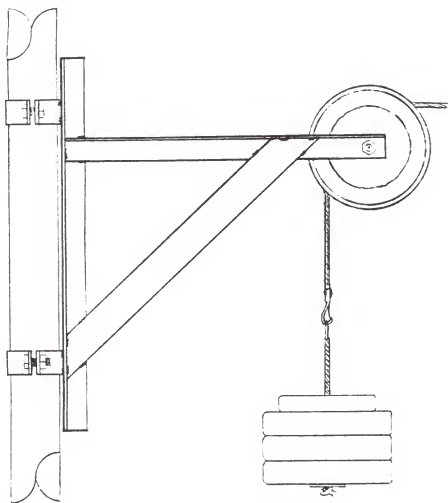
All horses were subjected to a loaded standard exercise tolerance treadmill test on days 0, 28 and 56 of conditioning, as well as day 28 post-conditioning. The test consisted of working the horses on a loaded equine treadmill (Anamill, MLR Design, Sundusky, OH) at a 7 degree incline for 15 min. While on the treadmill, horses wore a collar and hames. Attached to each side of the hames by a snap was a cable leading back to the pulley system in diagram 1. This enabled horses to pull weight while running on the treadmill. Horses were worked at a speed of 1.0 m/sec for 10 min followed by 5 min at a speed of 2.8 m/sec.

Heart rates were measured by a heart rate monitor (Uniq Heartwatch, Computer Instrument Corp., Hempstead, NY) and were recorded for every minute of the test and at 1 through 5 and 10 min of recovery (Appendix C).

Respiration rates were measured at rest and immediately following the cessation of the test by observing the nostrils.

Jugular venous blood samples (5 ml) were taken through an indwelling catheter (Abbocath-T, Abbott Hospitals, Inc., North Chicago, IL) at rest, 5 and 10 min of exercise and immediately following the test for determination of lactic acid concentration. Diluted heparin was injected into the catheters to prevent clotting. Following centrifugation, lactic acid concentration of each sample was determined with

DIAGRAM 1. PULLEY FOR LOADED STANDARD EXERCISE TOLERANCE
TREADMILL TESTS



A MOUNTED SIDE VIEW W/ WEIGHTS
SCALE 1" = 6.0"

ROB GEORGE 3/27/89

a YSI 23L Lactate Analyzer (Yellow Springs Instrumentation Co., Yellow Springs, OH).

Standard Exercise Tolerance Sled Test

All horses were subjected to a 10 min sled test at day 0 and day 56 of conditioning. Horses were allowed a 2 min warm-up pulling an empty (90.9 kg) sled. Weight was added progressively throughout the remaining 8 min (20.5 kg at 2 min, 40.9 kg at 4 min, 45.5 kg at 6 min and 9.1 kg at 7 min) until the horses were pulling a total of 206.8 kg.

Heart rate was measured by a heart rate monitor and was recorded every minute throughout the test (Appendix D).

Respiration rate was taken at rest and immediately after the test by observing the nostrils.

Blood samples (5 ml) were taken at rest and at the cessation of the test for blood lactate analysis.

Standard Exercise Tolerance Acceleration Test

Horses were subjected to a 182.4 m acceleration test at day 0 and 56 of conditioning. The test was conducted on a 805 m oval dirt track. Horses were allowed a warm-up which consisted of 805 m at a walk and 402 m at a long trot. Horses were then loped for 91 m until reaching the starting point at which time they were sprinted for 182.4 m.

Heart rates were recorded by a heart rate monitor at rest, 91.0 m and immediately after crossing the 182.4 m finishing line.

Blood samples (5 ml) were taken at rest and at the cessation of the test for blood lactate analysis.

Split times were recorded at 91 m and 182.4 m of the sprint test by individual stop watches.

Statistical Analysis

Data of all experiments were analyzed by analysis of variance and means were separated by LS Means Method using the Statistical Analysis Systems Package (SAS, 1985).

RESULTS AND DISCUSSION

Physiological Responses to the Loaded Standard Exercise Tolerance Treadmill Tests

Heart rate responses (beats/min) to the loaded standard exercise tolerance treadmill tests (LSET) are shown in Table 2. Pre-test heart rate averaged 37.8 beats/min over all test days and there was no significant change over time. This agrees with data from other researchers who found no reduction in resting heart rate due to conditioning (Cardinet et al., 1963; Erickson et al., 1985; Sexton et al., 1985; Drozd et al., 1987; Foreman et al., 1987).

Heart rates during the LSET did not vary with conditioning. Other researchers (Sigler, 1981; Erickson et al., 1985; Sexton et al., 1985; Drozd et al., 1987) have reported a decrease in exercising heart rate on the treadmill

TABLE 2. MEAN HEART RATE RESPONSE (beats/min) TO LOADED STANDARD EXERCISE TOLERANCE TREADMILL TESTS

Time	Day 0	Day 28	Day 56	Day 28 detraining
Pre-test	38.3	35.0	38.8	39.0
5 min	118.0 ^a	128.8 ^{ab}	116.0 ^a	156.3 ^b
10 min	113.3	124.0	109.8	128.8
15 min	215.3	218.5	206.0	201.8
Recovery				
5 min	95.3 ^c	89.3 ^{bc}	80.3 ^a	86.5 ^{ab}
10 min	88.8 ^c	80.0 ^{bc}	65.8 ^a	76.3 ^b

^{abc} Means in the same row with different superscripts differ significantly (P<.05).

following training; however, no such change was observed in this study. This is in agreement with work done by Milne et al. (1977) who observed no decrease in heart rates of Standardbred horses after training and Johnson et al. (1987) who found no changes in the exercising heart rates of ponies after 6 wk of anaerobic conditioning. Perhaps no differences were observed during the LSET because horses were nearing maximal heart rates, which are not greatly affected by training (Sexton et al., 1985).

Differences ($P < .05$) were observed in the 5 min recovery rates which dropped from 95.3 beats/min on day 0 to 80.3 beats/min on day 56. Recovery heart rates at 10 min also were lower ($P < .05$) at the end of the 56 d conditioning period going from 88.8 beats/min on day 0 to 65.8 beats/min on day 56. This agrees with work done by Cardinet et al. (1963), Erickson et al. (1985) and Sexton et al. (1985). During all LSET horses recovered very rapidly as heart rates dropped from 210.4 beats/min to 87.9 beats/min at 5 min and 77.7 beats/min at 10 min of recovery. This rapid recovery rate has been reported by other researchers in Quarter Horses (Cardinet et al., 1963; Sigler, 1981; Erickson et al., 1985) and in ponies (Sexton et al., 1985). Milne (1977), however, found no significant difference in heart rate recovery values in horses who had undergone a 92 d conditioning program.

After 28 d of detraining no differences were observed in resting heart rate when compared to day 0, 28 and 56 of

conditioning. However, at 5 min of exercise during the LSET heart rates were higher ($P<.05$) than at day 0 and 56 of conditioning. At 5 and 10 min of recovery, heart rates were lower ($P<.05$) than day 0 indicating horses were still more conditioned after 28 d of rest.

Respiration rates (exhalations/min) before and after the LSET are shown in Table 3. A change was observed in pre-test respiration rates decreasing ($P<.05$) from 40.0 exhalations/min on day 0 to 23.5 exhalations/min on day 56.

Post-test respiration rates averaged 91.5 exhalations/min over both test days and no changes were observed due to conditioning. Snow and MacKenzie (1977a) and Sigler (1981) also found no significant changes in respiration rates as a result of conditioning in Standardbreds and Quarter Horses. Similar results were reported by Drozd et al. (1987). Campbell et al. (1985), however, did see a significant decrease in respiration rate in Quarter horses after cutting training.

After 28 d of detraining a significant decrease ($P<.05$) was seen in the pre-test respiration rate when compared to day 0 of conditioning. However, no significant changes were seen in the post-test respiration rates.

Respiration rates are affected by a wide variety of factors and therefore are not useful as a reliable indicator of fitness (Cardinet et al., 1963; Johnson et al., 1987).

Blood lactic acid concentrations (mmol/l) for the LSET are seen in Table 4. Pre-test blood lactate concentration did

TABLE 3. MEAN RESPIRATION RESPONSE (exhalations/min) TO
LOADED STANDARD EXERCISE TOLERANCE TREADMILL TESTS

Time	Day 0	Day 28	Day 56	Day 28 detraining
Pre-test	40.0 ^c	30.0 ^b	23.5 ^{ab}	17.5 ^a
Post-test	102.0	87.3	87.5	89.3

^{abc} Means in the same row with different superscripts differ significantly ($P < .05$).

TABLE 4. MEAN BLOOD LACTATE RESPONSE (mmol/l) TO LOADED STANDARD EXERCISE TOLERANCE TREADMILL TESTS

Time	Day 0	Day 28	Day 56	Day 28 detraining
Pre-test	.6	1.0	.9	.6
5 min	3.0 ^b	1.7 ^a	1.1 ^a	1.5 ^a
10 min	4.5 ^b	2.0 ^a	1.5 ^a	2.3 ^a
15 min	19.8 ^b	10.4 ^a	10.4 ^a	9.9 ^a

^{ab} Means in the same row with different with different superscripts differ significantly ($P < .05$).

not differ significantly over all test days and averaged .8 mmol/l. These data agree with other researchers (Lindholm and Saltin, 1974; Mullen et al., 1979; Erickson et al., 1985; Sexton et al., 1985; Drozd et al., 1987; Johnson et al., 1987) who also reported no change in resting blood lactate concentration due to conditioning.

Blood lactate concentration during and immediately after the LSET was significantly lower ($P<.05$) after 28 and 56 d of conditioning. This indicates that there was a significant conditioning response to the anaerobic training. This agrees with several other researchers who have observed decreased concentrations of blood lactate as a result of conditioning (Sigler, 1981; Erickson et al., 1985; Sexton et al., 1985; Shelle et al., 1985; Webb et al., 1987; Drozd et al., 1987).

After 28 d of detraining it was also found that blood lactate concentration was significantly lower ($P<.05$) than day 0 at 5, 10 and 15 min of conditioning, indicating there was still a conditioning effect after 28 d of detraining.

Physiological Responses to Standard Exercise Tolerance Sled Tests

Heart rate responses (beats/min) to the exercise tolerance sled tests are seen in Table 5. Pre-test heart rates decreased ($P<.05$) over time from 40.8 beats/min on day 0 to 34.8 beats/min on day 56. However, a reduced resting heart rate is rarely observed (Cardinet et al., 1963; Sigler,

TABLE 5. MEAN HEART RATE RESPONSE (beats/min) STANDARD EXERCISE TOLERANCE SLED TESTS

Time	Day 0	Day 56
Pre-test	40.8 ^b	34.8 ^a
5 min	131.8	106.5
10 min	172.3	153.8

^{ab} Means in the same row with different superscripts differ significantly ($P < .05$).

1981; Erickson et al., 1985; Sexton et al., 1985; Drozd et al., 1987; Foreman et al., 1987).

Heart rates during and after the exercise tolerance sled tests averaged 119.5 and 163.5 beats/min and did not vary with conditioning.

Respiration rates (exhalations/min) before and after the exercise tolerance sled tests are shown in Table 6. Pre-test respiration rate decreased ($P < .05$) from 33.0 exhalations/min on day 0 to 18.5 exhalations/min on day 56. Post-test respiration rates also decreased ($P < .05$) from 96.0 exhalations/min on day 0 to 40.0 exhalations/min on day 56.

Blood lactic acid concentrations (mmol/l) before and after the exercise tolerance sled test are shown in Table 7. Pre-test blood lactate concentration averaged .7 mmol/l over both test days and did not differ significantly due to conditioning.

Blood lactate concentration following the exercise tolerance sled tests did show a significant decrease ($P < .05$) from 4.3 mmol/l on day 0 to 1.3 mmol/l on day 56.

Physiological Responses to Standard Exercise Tolerance Acceleration Tests

Heart rate responses (beats/min) to the sprint acceleration tests are shown in Table 8. Pre-test heart rate over both days averaged 35.9 beats/min and did not show a significant decrease due to training. Post-test heart rates averaged 158 beats/min at 91.0 m and 196.3 beats/min at the

TABLE 6. MEAN RESPIRATION RESPONSE (exhalations/min) TO STANDARD EXERCISE TOLERANCE SLED TESTS

Time	Day 0	Day 56
Pre-test	33.0 ^b	18.5 ^a
Post-test	96.0 ^b	40.0 ^a

^{ab} Means in the same row with different superscripts differ significantly ($P < .05$).

TABLE 7. MEAN BLOOD LACTATE RESPONSE (mmol/l) TO STANDARD EXERCISE TOLERANCE SLED TESTS

Time	Day 0	Day 56
Pre-test	.7	.6
Post-test	4.3 ^b	1.3 ^a

^{ab} Means in the same row with different superscripts differ significantly ($P < .05$).

TABLE 8. MEAN HEART RATE RESPONSE (beats/min) TO STANDARD EXERCISE TOLERANCE ACCELERATION TESTS

Distance	Day 0	Day 56
Pre-test	37.0	34.8
91.2 m	153.0	163.0
182.4 m	196.5	196.0

finish of the test and showed no changes due to conditioning.

Blood lactate concentration (mmol/l) before and after the exercise tolerance acceleration test are shown in Table 9. Pre-test blood lactate concentration averaged .7 mmol/l over both test days and did not differ significantly due to conditioning.

Blood lactate concentration after the sprint acceleration test averaged 8.6 mmol/l over both test days and did not differ significantly due to conditioning.

Sprint Time Response to the Exercise Tolerance Acceleration Tests

Sprint times (sec) before and after conditioning are shown in Table 10. Time at 91 m showed a significant decrease ($P < .05$) from 8.9 sec on day 0 to 6.8 sec on day 56. Time at 182.4 m averaged 15.0 sec over both test days and did not show a significant decrease as a result of conditioning. However, there was a trend ($P = .07$) towards a faster time at the 182.4 m finishing line.

Nutrient Digestibility

As was stated previously, feed intake was kept exactly the same for each individual horse throughout the nitrogen balance trials.

Digestibilities of proximate components are shown in Table 11. Dry matter digestibility averaged 54.4% over all

TABLE 9. MEAN BLOOD LACTATE RESPONSE (mmol/l) TO STANDARD EXERCISE TOLERANCE ACCELERATION TESTS

Time	Day 0	Day 56
Pre-test	.5	.9
Post-test	9.2	8.0

TABLE 10. MEAN SPRINT TIMES (seconds) FOR STANDARD EXERCISE TOLERANCE ACCELERATION TESTS

Distance	Day 0	Day 56
91.2 m	8.9 ^b	6.8 ^a
182.4 m	16.4	13.6

^{ab} Means in the same row with different superscripts differ significantly ($P < .05$).

TABLE 11. DIGESTIBILITY OF PROXIMATE COMPONENTS

Variable	Period 1	Period 2	Period 3	Period 4
Dry matter digestibility, %	55.9	54.1	53.4	54.3
Ether extract digestibility, %	14.8 ^b	22.8 ^c	-7.1 ^a	11.9 ^b
Ash digestibility, %	26.2 ^b	10.4 ^a	1.9 ^a	2.1 ^a
Crude protein digestibility, %	33.3	30.4	31.0	42.2

^{ab} Means in the same row with different superscripts differ significantly ($P < .05$).

collection periods and did not differ from one collection period to the next.

Ether extract digestibilities were significantly lower ($P < .05$) during the third collection period and there was no significant difference observed between the first and fourth collection periods. Schneider and Flatt (1975) point out the ether extract digestibility may be biased by ether soluble substances in the feces, and that errors are often present, particularly when fat content of the diet is low. A large variation in ether extract digestibility as well as negative values were also reported by McNally (1979) in grade geldings consuming alfalfa, brome and prairie hay. Similar findings were reported by Ott (1985) in Thoroughbred and Quarter Horse geldings consuming rations of coastal bermudagrass hay or coastal bermudagrass hay plus cracked corn.

Ash digestibility was significantly lower ($P < .05$) during the second, third and fourth collection periods when compared to the first collection period. Schneider and Flatt (1975) point out that errors may be present in ash digestibility since fecal mineral matter may include large amounts of mineral substances excreted from the blood into the digestive tract, and therefore much of the ash in the feces may not be undigested feed ash.

Crude protein digestibility averaged 34.2% over all collection periods and no significant differences were observed. In any digestion trial, some errors in the

calculation of proximate digestibilities are inherent. Schneider and Ellenberger (1927) found that in dairy cows, the greatest error encountered when calculating digestion coefficients was due to irregularities in the amount of fecal excretion, especially at low levels of feed intake. In this study chromic oxide was fed as an external indicator and total feces were not collected. Schurg, (1985) found that there was no significant difference between total fecal collection and chromic oxide in determining digestion coefficients for mature ponies and horses consuming hay, hay-grain or whole corn plant diets.

Digestibility may have been affected by pelleting the feed. Schneider and Flatt (1975) point out that pelleting may reduce digestibility somewhat due to a faster rate of passage through the gastrointestinal tract. Hintz and Loy (1966) reported that pelleting increased ether extract digestibility, but did not affect the digestibility of other proximate components in Thoroughbred and Quarter Horse fillies. They concluded that feeding the same levels of pelleted versus nonpelleted diets did not affect the apparent efficiency of feed utilization.

Nitrogen Utilization in Response to Conditioning

Nitrogen balance data are shown in Table 12. Nitrogen intake decreased ($P < .05$) from the first collection period to the third collection period. Nitrogen intake was also significantly lower ($P < .05$) in the fourth collection period when compared to the first collection period.

Fecal nitrogen excretion increased ($P < .05$) from the first to the third collection period. Fecal nitrogen excretion in the fourth collection period was not significantly different than the first collection period but was significantly lower ($P < .05$) than the second and third collection periods. It appears then, that fecal nitrogen is not related to nitrogen intake.

Urinary nitrogen excretion was significantly lower ($P < .05$) during the second collection period when compared to the first; however, there was no significant difference observed between the first and third collection periods. The fourth collection period also was not significantly different from the first collection period. Urinary nitrogen excretion in the second collection period was significantly different ($P < .05$) from all other collection periods.

Nitrogen absorbed was higher in period 1 than all other collection periods. Period 2 and 4 were not significantly different, while period 3 was the lowest. Freeman et al. (1985a) suggests that nitrogen metabolism may be affected by exercise in the horse; however, nitrogen absorption was

TABLE 12. NITROGEN VALUES

Variable	Period 1	Period 2	Period 3	Period 4
N-intake, g/d	162.8 ^d	156.3 ^c	139.0 ^a	145.9 ^b
Fecal N excretion, g/d	88.4 ^a	95.2 ^b	95.9 ^b	84.3 ^a
Urine N excretion, g/d	67.6 ^{bc}	61.0 ^a	69.3 ^c	65.2 ^b
N-balance, g/d	6.8 ^c	.1 ^{bc}	-26.2 ^a	-3.6 ^b
N-absorbed, g/d	74.4 ^c	61.1 ^b	43.1 ^a	61.6 ^b

abcd Means in the same row with different superscripts differ significantly ($P < .05$).

related to intake and in the third collection period nitrogen intake and nitrogen absorbed were both lower than the first and second collection periods.

Nitrogen balance values were significantly lower ($P < .05$) in the third collection period when compared to all others. Nitrogen balance in the fourth collection period was significantly lower ($P < .05$) than the first collection period and was higher ($P < .05$) than the third collection period. Nitrogen balance, then, was higher at higher levels of nitrogen intake. This has been reported by Slade et al. (1970) and Harper and Vander Noot (1974) in sedentary horses and Freeman et al. (1981) and Freeman et al. (1985b) in conditioned horses.

Correlations between nitrogen intake and other nitrogen values are shown in Table 13. Nitrogen intake was highly correlated with nitrogen balance.

Nitrogen balance and nitrogen absorbed as a percentage of intake are shown in Table 14. Nitrogen balance as a percentage of intake in the third collection period was significantly lower ($P < .05$) than the first and second collection periods. The fourth collection period was also significantly lower than the first collection period. Nitrogen absorption as a percent of intake was significantly lower ($P < .05$) in the third collection period than any other period. Furthermore, nitrogen absorption as a percent of

TABLE 13. CORRELATIONS BETWEEN NITROGEN INTAKE AND OTHER NITROGEN VALUES

Variable	Correlation coefficient
Fecal nitrogen	-.26
Urinary nitrogen	-.22
Nitrogen balance	.89*
Nitrogen absorbed	.32

*Significant correlation ($P < .0001$).

TABLE 14. NITROGEN BALANCE AND NITROGEN ABSORPTION AS A PERCENTAGE OF INTAKE

Variable	Day 0	Day 28	Day 56	Day 28 of detraining
N balance as % of intake	4.2 ^c	.06 ^{bc}	-18.9 ^a	-2.6 ^b
N absorbed as % of intake	45.7 ^c	39.1 ^b	31.0 ^a	42.1 ^{bc}

abc means in the same row with different superscripts differ significantly ($P < .05$)

intake in the second collection period was significantly lower ($P < .05$) than in the first period.

Because differences were seen in nitrogen balance, it appears that nitrogen balance is affected by both nitrogen intake and intense anaerobic exercise. Therefore, it may be that working horses require additional protein above the National Research Council's maintenance requirement.

One problem encountered in nitrogen balance trials is that nitrogen is lost through the sweat (Calloway et al., 1971; Jenkinson et al., 1974; Snow et al., 1982; Kerr et al., 1983; Freeman et al., 1985a; Johnson et al., 1987). Nitrogen balance values, then, may be higher than true balance values. Consalazio et al. (1975) point out that nitrogen balance may be biased particularly when sweating is profuse. Thus nitrogen lost in the sweat may have been a source of error in calculating nitrogen balance.

SUMMARY

The LSET appears to be an appropriate means of accessing fitness for this type of anaerobic exercise and data indicate that different standard exercise tolerance tests are needed for different types of conditioning. Blood lactate concentration appears to be an accurate indicator of fitness for the LSET as blood lactate concentration was lower ($P < .05$) on day 56 of conditioning than day 0 at 5, 10 and 15 min of

the test. Furthermore, horses appeared to maintain cardiovascular fitness through 28 d of detraining. Data from the acceleration tests indicate that it is not a good measure of fitness for this type of conditioning, as it was a completely different type of exercise, much shorter in duration and less strenuous than the other standard exercise tolerance tests. However, it must be noted that the anaerobic conditioning did increase ($P<.05$) speed for the first 91.0 m of the acceleration test and tended ($P=.07$) to increase speed from the start to the 182.4 m finishing point.

Nitrogen intake was highly correlated with nitrogen balance. From this study, it appears that nitrogen balance is affected by both nitrogen intake and intense anaerobic conditioning. Since negative nitrogen balance values were observed, working horses may have a need for increased dietary protein above the recommended requirement.

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APPENDICES

APPENDIX A. PRE AND POST-TRIAL BODY WEIGHTS OF
ANAEROBICALLY EXERCISE HORSES

Horse Number	Pre-trial wt, kg	Post-trial wt, kg
1	470.5	475.0
2	522.7	522.7
3	484.1	484.1
4	447.7	450.0
avg	481.3	483.0

APPENDIX B. AVERAGE WEIGHT PULLED EACH WEEK

Horse Number	week							
	1	2	3	4	5	6	7	8
	wt pulled, kg							
1	206.8	272.7	286.4	343.2	343.2	353.4	353.4	365.9
2	206.8	272.7	302.3	343.2	343.2	363.6	363.6	372.9
3	206.8	272.7	275.0	343.2	343.2	347.3	347.3	372.7
4	206.8	272.7	275.0	290.9	310.0	318.2	318.2	338.1
avg	206.8	272.7	284.7	330.1	334.9	345.6	345.6	362.4

APPENDIX C. MEAN HEART RATE RESPONSE (beats/min)
 FOR EACH MINUTE OF THE LOADED STANDARD
 EXERCISE TOLERANCE TREADMILL TESTS

Time (min)	Day 0	Day 28	Day 56	Day 28 of detraining
1	38.3	35.0	38.8	39.0
2	143.3	123.3	113.3	136.8
3	152.8	135.3	118.3	141.0
4	130.8	131.8	111.0	140.5
5	118.0	128.8	91.0	156.3
6	107.8	114.3	109.3	137.0
7	121.5	117.3	110.3	128.5
8	121.0	115.5	110.0	126.0
9	120.3	113.5	106.3	130.8
10	113.3	124.0	109.8	128.8
11	172.5	192.0	183.0	158.8
12	196.0	207.0	198.0	195.3
13	214.8	214.5	194.0	199.3
14	193.0	212.0	196.3	199.3
15	197.5	218.5	206.0	201.8
Recovery				
1	138.3	117.8	106.0	121.3
2	106.3	111.8	96.0	100.5
3	102.0	93.0	91.0	92.3
4	97.5	90.5	87.5	88.8
5	95.3	89.3	80.3	86.5
10	88.8	80.0	65.8	76.3

APPENDIX D. MEAN HEART RATE RESPONSE (beats/min) FOR EACH MINUTE OF THE STANDARD EXERCISE TOLERANCE SLED TESTS

Time (min)	Day 0	Day 56
1	138.5	83.3
2	125.3	81.5
3	128.0	85.8
4	126.8	97.0
5	131.8	106.5
6	134.0	124.3
7	156.3	142.5
8	163.5	155.0
9	169.0	143.3
10	172.3	153.8

NITROGEN UTILIZATION AND METABOLIC RESPONSES OF HORSES
TO INTENSE ANAEROBIC EXERCISE

by

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Four 3-yr-old Quarter Horse geldings were used to study the effects of intense anaerobic exercise on nitrogen utilization and metabolic responses to three standard exercise tolerance tests: a loaded treadmill test; a sled test; and a 182.4 m acceleration test. Conditioning consisted of pulling a weighted sled for 10 min/d, twice a day, 5 d/wk for 8 wk. Weight was adjusted accordingly to maintain heart rates of 170 to 180 beats/min throughout each work session. All horses were subjected to a 15 min loaded standard exercise tolerance treadmill test (LSET) on 0, 28 and 56 d of conditioning and after 28 d of detraining. On days 0 and 56, horses were subjected to the 10 min sled test and the 182.4 m acceleration test. Parameters measured included heart rate, blood lactate concentration and respiration rate. Split times were recorded for the acceleration test at 91.0 m and 182.4 m. The LSET appears to be an appropriate means of accessing fitness for this type of anaerobic exercise. Blood lactate concentration appears to be an accurate indicator of fitness for the LSET as blood lactate concentration was lower ($P < .05$) on day 56 of conditioning than day 0 at 5, 10 and 15 min of the test. In addition, it horses appeared to maintain cardiovascular fitness through 28 d of detraining. Data from the acceleration test indicate that it is not a good measure of fitness as it was a completely different type of exercise, much shorter in duration and less strenuous than the other standard exercise tolerance tests. However, it must be noted that the anaerobic conditioning did increase ($P < .05$) speed for

the first 91.0 m of the acceleration test and tended ($P=.07$) to increase speed from the start to the 182.4 m finishing point. Five day nitrogen balance trials were conducted prior to conditioning, at 1 to 5 d of conditioning, 52 to 56 d of conditioning and again for 5 d after 28 d of detraining. Fecal nitrogen excretion increased ($P<.05$) from the first to the third collection period and urinary nitrogen excretion decreased ($P<.05$) in the second collection period and then increased ($P<.05$) in the third collection period. Nitrogen balance was lower ($P<.05$) in the third and fourth collection periods than in the first. Nitrogen absorption did not differ significantly. Nitrogen intake was highly correlated with nitrogen balance. It appears then that nitrogen balance is affected by both nitrogen intake and intense anaerobic training. Since negative nitrogen balance values were seen, horses undergoing intense anaerobic exercise may have an increased need for dietary protein.