

# **Advances in Equine Nutrition**

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# RECENT RESEARCH DEVELOPMENTS FROM KENTUCKY EQUINE RESEARCH

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Significant advances have been made over the past 30 years in the understanding of equine nutrition. Still, many questions remain unanswered about how to feed and manage horses. Over the past 12 years, Kentucky Equine Research has conducted numerous research studies that have addressed questions relevant to the horse feed industry. This paper will review five of the most recent studies.

## Hay Intake and Performance

The amount of energy that a horse needs to run is directly related to the weight being moved (horse, rider, and tack) and the speed of running. Addition of weight increases the energy cost of locomotion. Diet can have a marked effect on body weight. Specifically, high-roughage diets increase the mass of ingesta in the equine large intestine, the result of greater water consumption and the ability of fiber to bind to water. High-fiber diets are desirable for horses engaged in endurance sports because the larger reservoir of fluid and electrolytes in the hindgut may lessen the severity of dehydration during prolonged exercise when compared with a low-fiber diet. On the other hand, the increase in body weight when horses consume a high-fiber diet will mandate an increase in energy expenditure at any given running speed and may be detrimental to performance, particularly during high-intensity exertion.

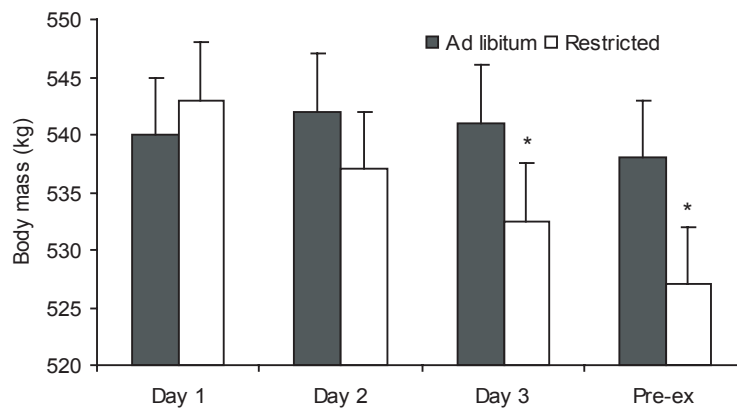
Traditionally, racehorse trainers restrict the amount of hay fed to horses before racing in an attempt to improve performance. However, no study has examined the effect of restricted hay intake on body weight, nor the effects of this dietary manipulation on exercise performance. Therefore, KER conducted a study to determine the effects of restricted hay intake on the metabolic responses of horses to high-intensity exercise (Rice et al., 2001). We hypothesized that, compared to ad libitum hay intake, a regimen of restricted hay feeding starting 3 days before a standardized exercise test would decrease body weight and reduce energy expenditure during running.

Four conditioned Thoroughbred horses were studied in a 2 x 2 crossover design. Initially, the length of time required for adaptation to ad libitum intake of grass hay was determined. Thereafter, the metabolic responses to sprint exercise (SPR)

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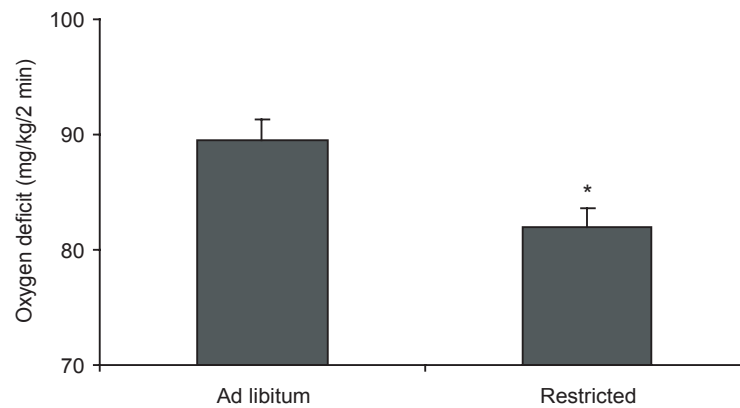
were examined in two dietary periods, each 5 days in duration: 1) Ad libitum (AL), where horses had free-choice access to hay; and 2) Restricted (RES), where hay intake was restricted (~1% of body weight) for 3 days before the exercise test. Feed and water were removed 4 h before the exercise test.

After measurement of body weight, horses completed a warm-up followed by 2 min at 115% of maximum oxygen uptake, then a 10-min walking recovery (REC). During the 3 days before SPR, hay intake in AL averaged ( $\pm$  SE)  $10.1 \pm 0.9$  kg, whereas intake during RES was  $4.3 \pm 0.2$  kg. Pre-exercise body weight was significantly lower in RES ( $528 \pm 5$  kg) than in AL ( $539 \pm 4$  kg). During SPR, total mass-specific  $\text{VO}_2$  was higher ( $P=0.02$ ) in RES ( $243 \pm 8$  ml/kg/2 min) than in AL ( $233 \pm 10$  ml/kg/2 min). Conversely, accumulated oxygen deficit was higher ( $P<0.01$ ) in AL ( $89.4 \pm 2.2$  ml  $\text{O}_2$ /kg) than in RES ( $82.4 \pm 1.7$  ml  $\text{O}_2$ /kg). Peak plasma lactate was also higher in AL ( $22.2 \pm 1.2$  mM) than in RES ( $19.1 \pm 2.1$  mM), and  $\text{VO}_2$  during recovery was 10% higher ( $P=0.12$ ) in AL.



**Figure 1.** Body weight in the ad libitum and restricted hay intake groups for the 3-day period preceding the exercise test. Pre-ex = Body weight measured 5 min before the exercise test. \*Significant ( $P<0.05$ ) difference ad libitum vs. restricted.

The main findings of this study were: 1) compared to ad libitum hay feeding, 3 days of restricted (1% of body weight) hay intake was associated with an approximately 2% decrease in body weight; and 2) the reduction in body weight associated with restricted hay feeding resulted in an increase in the mass-specific rate of oxygen consumption during sprint exercise, with a corresponding decrease in anaerobic energy expenditure. The anaerobic contribution to energy expenditure during exercise was lower in RES than in AL as evidenced by lower values for accumulated oxygen deficit (Figure 2) and peak plasma lactate concentrations.



**Figure 2.** Accumulated oxygen deficit during 2 min of exercise at 115% of maximal oxygen uptake in the ad libitum and restricted treatments. \* $P < 0.05$  restricted vs. ad libitum.

Currently, it is recommended that performance horses receive hay at a minimum of 1% of body weight per day to satisfy requirements for long stem fiber and minimize digestive upsets. In this context, relative to the restriction protocol used in this study, more severe or longer-term restrictions of hay intake are not recommended. Nonetheless, on the basis of our results, further studies that examine the relationship between fiber intake, body weight, and exercise metabolism and performance are warranted.

### Arabians vs. Thoroughbreds

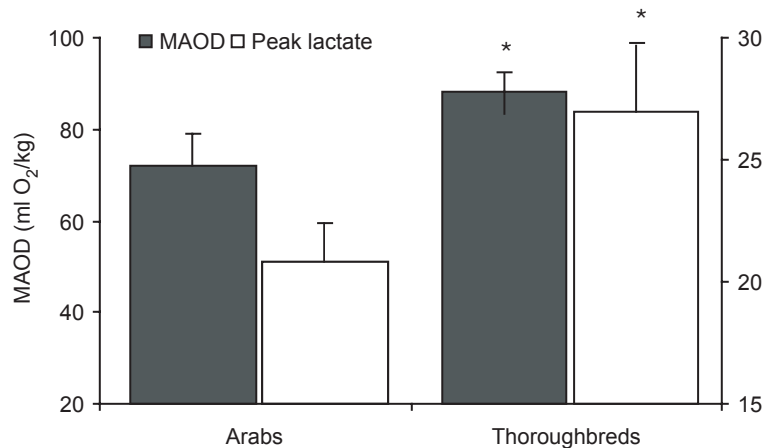
Few studies have formally compared indices of athletic performance in different breeds of horses. Rose and colleagues (1995) described indices of exercise capacity in Thoroughbred and Standardbred racehorses that were examined because of poor performance. These researchers reported that aerobic capacity and total run time, measured during an incremental exercise test, were significantly greater in the Thoroughbreds compared to the Standardbreds. However, as these horses were examined for performance problems, it is difficult to apply the findings to normal racehorses.

In a recent KER study (Prince et al., 2001), we examined selected measures of exercise capacity and metabolism in a small group of Thoroughbred and Arabian horses of similar age, training background, and diet. Both breeds of horses are used for several athletic disciplines, ranging from sprint racing to endurance events. However, anecdotal evidence indicates that Thoroughbreds have superior high-intensity exercise capacity, whereas Arabian horses are regarded as superior performers during endurance exercise. We hypothesized that the facility of Thoroughbred horses for high-intensity exercise would be reflected in greater aerobic and anaerobic capacities when compared to the Arabian horses. We also

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hypothesized that respiratory exchange ratio (RER) would be lower in the Arabians during low-intensity exercise, reflecting a greater use of fat for energy.

The metabolic responses to low- and high-intensity exercise were compared in five Arabian (AR) and five Thoroughbred (Tb) horses. For 2 months before the study, horses were fed an identical diet and undertook a similar exercise training program. Horses then completed three treadmill (3° incline) trials: 1) an incremental test (MAX) for determination of aerobic capacity,  $V_{LA4}$ , and lactate threshold (LT; the percentage of  $VO_{2max}$  when plasma lactate = 4 mM); 2) a sprint test (SPR) for estimation of maximal accumulated oxygen deficit (MAOD) in which horses ran until fatigue at 115%  $VO_{2max}$ ; and 3) a 90-min test at 35%  $VO_{2max}$  (LO). There was a minimum of 7 days between tests and the order of the SPR and LO trials was randomized. For all tests,  $VO_2$ ,  $VCO_2$ , and RER were measured throughout exercise. For MAX, horses ran for 90 s at 4 and 6 m/s, with subsequent increases of 1 m/s every 60 s until fatigue. Blood samples were obtained during the last 10 s of each speed increment. In SPR, MAOD was calculated by subtracting actual  $VO$  from estimated  $O_2$  demand. In LO, samples for measurement of plasma glucose and free fatty acids (FFA) were obtained at 0, 5, 15, 30, 45, 60, 75, and 90 min of exercise. Data were analyzed using Student's t-test or 2-way repeated measures ANOVA.



**Figure 3.** Maximum accumulated oxygen deficit and peak lactate concentrations for the Arabian and Thoroughbred horses during a single high-speed exercise test at 115% of maximum oxygen uptake.

$VO_{2max}$  ( $P<0.001$ ) and running speed ( $P<0.05$ ) at  $VO_{2max}$  were higher in Tb ( $154 \pm 3$  ml/kg/min at  $12.9 \pm 0.5$  m/s) than in AR ( $129 \pm 2.5$  ml/kg/min at  $11.8 \pm 0.2$  m/s). Run time to fatigue during MAX was greater ( $P<0.05$ ) in Tb ( $10.5 \pm 0.5$  min) than in AR ( $9.3 \pm 0.3$  min). However,  $V_{LA4}$  and LT were not different between groups. Run time during SPR (Tb  $149 \pm 16$ ; AR  $109 \pm 11$  s) and MAOD

(Tb  $88 \pm 4$ ; AR  $70 \pm 6$  ml O<sub>2</sub>/kg) were higher ( $P < 0.05$ ) in the Tb group (Figure 3). During LO, FFA were higher ( $P < 0.05$ ) in AR than in Tb between 60 and 90 min, while RER was lower ( $P < 0.05$ ) from 60 to 90 min of exercise. The higher aerobic and anaerobic capacity of the Tb horses likely contributed to their superior high- intensity exercise performance. Conversely, the AR may be better adapted for endurance exercise as evidenced by the greater use of fat. These metabolic differences may reflect breed variation in muscle fiber types.

### Glycemic Response with Beet Pulp

Previous studies in our laboratory demonstrated a marked glycemic response when horses were fed a fiber mix consisting of equal parts rice bran, soy hulls, wheat bran, and soaked beet pulp (Pagan et al., 1999a). We speculated that, in part, the beet pulp portion of this fiber mix was responsible for the increase in plasma glucose concentrations after meal ingestion. We further hypothesized that the magnitude of the glycemic response to beet pulp would depend on how the beet pulp was prepared. Therefore, KER conducted a study to determine how different preparations of shredded beet pulp affect glycemic response in Thoroughbred horses (Groff et al., 2001). In a 4 x 4 Latin square design, four mature geldings (mean age 12 yr, body weight 568 kg) were fed: 1) 0.75 kg rinsed beet pulp (Rinse); 2) 0.75 kg hydrated beet pulp (Hydrate); 3) 0.675 kg dry beet pulp and 0.075 kg molasses (BP/Molasses); and 4) 0.75 kg whole oats (Oats). Water was added to both the rinsed and hydrated beet pulp, which were then allowed to stand overnight. In the Rinse treatment, excess water was drained and the beet pulp was washed repeatedly until the concentration of glucose in the wash water was  $< 1$  mg/dl. In the Hydrate treatment, water was not removed before feeding the beet pulp. Each treatment period was 7 days. In each period, the diet consisted of the treatment meal (0700h), 2 kg of whole oats (1600h), and 6.8 kg alfalfa hay cubes, divided into three equal feedings, at 0700, 1600, and 2200. Horses were given access to free exercise on pasture during the day, although they were not allowed to graze. The glycemic response trials were performed on day 7 of each period after an overnight fast (10 h). The test meal was fed at 0700 h. Blood samples were taken before feeding to determine baseline glucose values and at 30-min intervals following feeding for 480 min. The morning allotment of alfalfa cubes was fed after completion of sample collection. Measurements included area under the curve, mean glucose (mg/dl), peak glucose (mg/dl), and time to peak glucose (min). Plasma glucose concentrations were statistically analyzed by the general linear model procedure for analysis of variance, with period, horse, and treatment included in the model. Statistical significance was set at  $P < 0.05$ . Using area under the curve for whole oats as a standard of reference, a glycemic index was determined from area under the curve for the other treatments.

Ingestion of rinsed beet pulp resulted in significantly lower area under the curve, mean glucose, peak glucose, and time to peak glucose when compared to

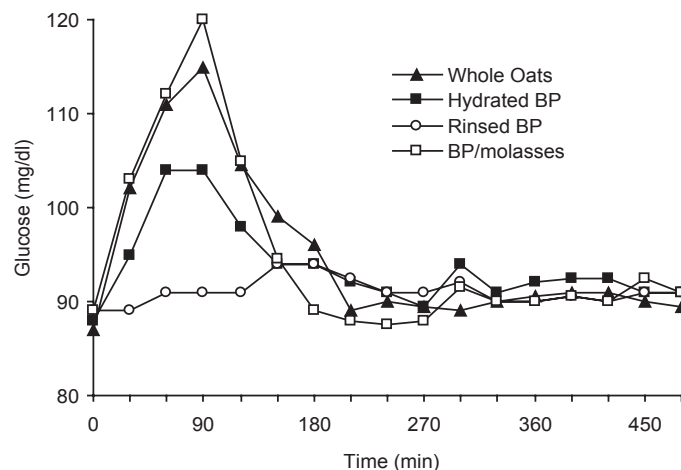
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**Table 1.** Area under the curve, mean glucose, peak glucose, and time to peak glucose concentration for all dietary treatments.

	<i>Area under curve</i>	<i>MG (mg/dl)</i>	<i>PG (mg/dl)</i>	<i>TTP (min)</i>	<i>GI</i>
Rinsed	1422 <sup>a</sup>	91.2 <sup>a</sup>	94.5 <sup>a</sup>	195 <sup>b</sup>	34.1
Hydrated	3017 <sup>b</sup>	94.4 <sup>b</sup>	105.4 <sup>b</sup>	75 <sup>a</sup>	72.2
Dry + molasses	3834 <sup>b</sup>	95.1 <sup>b</sup>	119.0 <sup>c</sup>	83 <sup>a</sup>	94.8
Whole oats	4175 <sup>b</sup>	94.8 <sup>b</sup>	114.6 <sup>c</sup>	83 <sup>a</sup>	100
SEM	406	0.41	2.72	37	
Statistical significance	0.01	0.05	0.01	0.01	

<sup>abc</sup> Treatments lacking a common superscript differ ( $P < 0.05$ ). MG, mean glucose; PG, peak glucose; TTP, time to peak glucose; GI, glycemic index

the other treatments (Table 1). Ingestion of Oats and BP/Molasses resulted in the highest glycemic response (Figure 4), while the plasma glucose response after Hydrate was intermediate between the Oats and BP/Molasses treatments and the Rinse treatment (Table 1, Figure 4). The estimated glycemic index was substantially lower for Rinse when compared to the other treatments (Table 1). The results of this study demonstrate that the glycemic response to a meal of beet pulp is markedly affected by preparation method. Removal of residual sugars by hydrating and rinsing dry beet pulp results in a negligible glycemic response, whereas the addition of 10% molasses to dry beet pulp results in a plasma glucose response that is indistinguishable from that observed after a meal of whole oats. These findings have important implications in the design of diets and feeding methods for horses that require diets low in hydrolyzable carbohydrate (e.g., horses with recurrent exertional rhabdomyolysis or polysaccharide storage myopathy).



**Figure 4.** Plasma glucose concentrations after ingestion of whole oats, hydrated beet pulp, rinsed beet, and beet pulp and molasses in four horses.

## Trace Mineral Requirements

Very little research has been conducted to determine the trace mineral requirements for athletic horses. In a previous KER study evaluating different forms of selenium (Se), horses supplemented with inorganic Se demonstrated a significant increase in urinary Se excretion after a single bout of exercise (Pagan et al., 1999b). These findings suggest that exercise increases the requirement for Se. Are the requirements for other microminerals also affected by exercise and training? To answer these questions, KER conducted a study (Hudson et al., 2001) to:

1. Determine the digestibility and retention of copper (Cu), zinc (Zn), and manganese (Mn) over four different levels of intake (basal, 50% of NRC added, 100% of NRC added, and 200% of NRC added);
2. Evaluate how regular exercise and training alters the requirements of these trace minerals.

Six mature Thoroughbred geldings [three sedentary (SED) and three horses in regular exercise training (EX)] were studied in a 16-week longitudinal experiment that consisted of 4 periods, each with a 23-day adaptation period followed by a 5-day complete collection digestion trial. In period 1, horses were fed an unfortified diet (basal intake – no supplementation), while in periods 2, 3 and 4 respectively, the diet included a supplement that provided 50, 100 and 200% of the NRC (1989) requirements for Cu, Zn, and Mn. For the basal diet, horses were fed unfortified sweet feed and timothy hay. This diet provided approximately 85%, 160% and 65% of the NRC recommendations for Cu, Mn and Zn, respectively. During the last week of this period, horses underwent a 5-day complete digestion trial. For the EX horses, a standardized exercise test was completed on the third day of the trial.

**Table 2.** Comparison of the true digestibility, endogenous loss, and estimated daily requirements for copper (Cu), zinc (Zn), and manganese (Mn) in sedentary (SED) horses (n = 3) and physically conditioned (EX) horses (n = 3). Data are mean  $\pm$  SD.

		<i>Cu</i>	<i>Zn</i>	<i>Mn</i>
Digestibility (%)	SED	41.8 $\pm$ 17.6	25.4 $\pm$ 11.4	57.9 $\pm$ 10.0
	EX	54.5 $\pm$ 11.7	14.3 $\pm$ 3.9**	40.2 $\pm$ 11.8**
Endogenous loss (mg/day)	SED	15.7 $\pm$ 1.6	65.2 $\pm$ 25.6	304.8 $\pm$ 95.1
	EX	20.3 $\pm$ 18.0	69.6 $\pm$ 35.9	163.6 $\pm$ 67.9
Requirement (mg/day)	SED	44.2 $\pm$ 23.6	274.4 $\pm$ 87.5	528.6 $\pm$ 168.6
	EX	35.0 $\pm$ 24.6	461.3 $\pm$ 133.2*	408.3 $\pm$ 107.1

\* P < 0.05 EX vs. SED

\*\* P < 0.1 EX vs. SED

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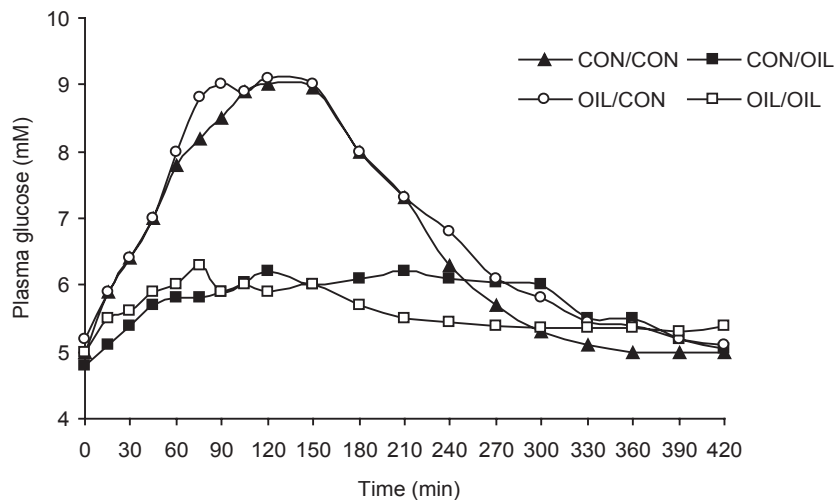
There was a significantly higher Zn requirement in EX than in SED horses (Table 2). Although there were no other significant differences between SED and EX, there were trends for lower true digestibility of Mn ( $P=0.059$ ) and Zn ( $P=0.09$ ) in EX. Results of this study suggest that exercise training results in a higher requirement for Zn but does not affect the true digestibility and maintenance requirements of Cu and Mn in mature Thoroughbred horses.

### **Corn Oil Affects Gastric Emptying**

The  $^{13}\text{C}$ -octanoic acid breath (or blood) test was recently developed as a noninvasive method for measuring the rate of solid-phase gastric emptying (GE). We used this method to test the hypothesis that GE is delayed following ingestion of a grain plus corn oil meal compared to a meal of grain alone (Geor et al., 2001). Four mature (10-12 yr) Arabian horses were studied in a 2 x 2 factorial design study. Factor A was the habitual diet, either a control (CON; hay plus sweet feed [SWF]) or an isocaloric fat-supplemented diet (FAT; hay, SWF, and corn oil). Factor B was the type of meal consumed for the GE test (SWF, 2 g/kg body weight vs. SWF 2 g/kg body weight plus 10% corn oil [OIL]). Each diet period lasted 10 weeks, with 6 weeks in between. GE studies were performed during the 4<sup>th</sup> and 8<sup>th</sup> weeks of each period. Within each dietary period, and in random order, horses were tested in both the SWF and OIL conditions with the following four treatment combinations: CON/SWF, CON/OIL, FAT/SWF, and FAT/OIL. For assessment of solid-phase GE, the test meals were labeled with 1 g of  $^{13}\text{C}$ -octanoic acid. Blood samples for measurement of plasma glucose concentration and  $^{13}\text{C}$ -enrichment were collected at 30 min and immediately before ingestion of the test meal and at frequent intervals thereafter for 7 h. Three indices of blood  $^{13}\text{C}$ -enrichment were calculated: half-dose recovery time ( $t_{1/2}$ ), the time to peak blood  $^{13}\text{C}$ -enrichment ( $t(\text{max})$ ), and the gastric emptying coefficient (GEC).

The glycemic response was markedly decreased in the OIL compared to the SWF trials (Figure 5). This effect of corn oil was not altered by habitual diet. The blood  $^{13}\text{C}$  vs. time curve was altered such that it was not possible to calculate  $t_{1/2}$  and  $t(\text{max})$  for one horse in both the CON/OIL and FAT/OIL trials. Excluding data from this horse, addition of corn oil to the meal of SWF was associated with a significant decrease in GEC and a significant increase in  $t_{1/2}$  and  $t(\text{max})$ , as shown (mean  $\pm$  s.d.) in Table 3.

Based on this study, it was concluded that: 1) the addition of corn oil to a meal of sweet feed results in a delay in solid-phase GE; 2) the effect of oil on GE is not affected by short-term adaptation to a fat-supplemented diet; and 3) the slowing of GE may contribute to the blunted glycemic response following a grain meal containing corn oil. The delayed GE may be due to a direct effect of oil on motility or the resultant increased energy density of the test meal.



**Figure 5.** Glycemic response with and without added corn oil.

**Table 3.** Effect of adding corn oil on the gastric emptying coefficient (GEC), half-dose recovery time ( $t_{1/2}$ ), and the time to peak blood  $^{13}\text{C}$ -enrichment ( $t(\text{max})$ ).

<i>Treatment</i>	<i>GEC</i>	<i>t<sub>1/2</sub> (h)</i>	<i>t(max) (h)</i>
CON/SWF	$2.96 \pm 0.15$	$2.25 \pm 0.55$	$1.20 \pm 0.21$
CON/OIL	$2.10 \pm 0.14$	$3.87 \pm 0.39$	$2.08 \pm 0.30$
FAT/SWF	$3.02 \pm 0.09$	$2.21 \pm 0.45$	$1.24 \pm 0.37$
FAT/OIL	$2.05 \pm 0.21$	$4.11 \pm 0.66$	$2.14 \pm 0.28$

## Acknowledgments

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## References

- Geor RJ, Harris PA, Hoekstra KE, Pagan JD. 2001. Effect of corn oil on solid-phase gastric emptying in horses. J Vet Int Med 2001 (abstract submitted).
- Groff L, Pagan J, Hoekstra K, Gardner S, Rice O, Roose K, Geor R. 2001. Effect of preparation method on the glycemic response to ingestion of beet pulp in Thoroughbred horses. Proc. 17<sup>th</sup> Equine Nutr. and Physiol. Soc. Symp.

## 10 *Recent Research from Kentucky Equine Research*

- Hudson C, Pagan J, Hoekstra K, Prince A, Gardner S, Geor R. 2001. Effects of exercise training on the digestibility and requirements of copper, zinc and manganese in Thoroughbred horses. Proc. 17<sup>th</sup> Equine Nutr. and Physiol. Soc. Symp.
- Pagan JD, Harris PA, Kennedy MAP, Davidson N, Hoekstra KE. 1999a. Feed type and intake affects glycemic response in Thoroughbred horses. Proc. 16<sup>th</sup> Equine Nutr. And Phys. Soc. Symp.149-150.
- Pagan JD, Karnezos P, Kennedy MAP, Currier T, Hoekstra KE. 1999b. Effect of selenium source on selenium digestibility and retention in exercised Thoroughbreds. Proc. 16<sup>th</sup> Equine Nutr. and Physiol. Soc. Symp. 135-140.
- Prince A, Geor R, Harris P, Hoekstra K, Gardner S, Hudson C, Pagan J. 2001. Comparison of the metabolic responses of trained Arabian and Thoroughbred horses during high and low intensity exercise Proc. 17<sup>th</sup> Equine Nutr. and Physiol. Soc. Symp.
- Rice O, Geor R, Harris P, Hoekstra K, Gardner S, Pagan J. 2001. Effects of restricted hay intake on body weight and metabolic responses to high-intensity exercise in Thoroughbred horses. Proc. 17<sup>th</sup> Equine Nutr. and Physiol. Soc. Symp.
- Rose RJ, King CM, Evans DL, Tyler CM, Hodgson DR. 1995. Indices of exercise capacity in horses presented for poor racing performance. Equine Vet J Suppl. 18:418-421.