Equine Exercise Physiology 3: 299–304, 1991

Effects of Daily Exercise on Muscle Glycogen in the Thoroughbred Racehorse

D. H. SNOW and R. C. HARRIS

Department of Comparative Physiology, Animal Health Trust, Newmarket, Suffolk, England

ABSTRACT. Six Thoroughbred horses were studied over a period of 11 days in order to determine the fluctuation in muscle glycogen during a 12 day period of training with 1 600 m gallops every Tuesday and 1 000 m gallops each Friday. On intervening days horses were given either slow canter exercise (Week 1) or 10 to 20 min walking (Week 2). Horses consumed a normal diet of pelleted feed and hay providing an average daily intake of 108.6 MJ of digestible energy. Exercise gallops resulted in a 19 to 25% fall in the muscle glycogen content which was restored fully within 2 to 3 days. Walking or cantering exercise on intervening days did not affect repletion rates. The results do not support a cumulative decline in muscle glycogen content with a training protocol typical of that used in many British training yards

Key words. Muscle glycogen; horses; training; nutrition.

INTRODUCTION

Endurance capacity in man at work intensities of 60 to 85% VO_2 max is directly related to the glycogen content of the working muscles.[3,8] It seems reasonable to suppose that this may also be the case in the competing horse. With prolonged exercise almost total depletion of the glycogen in the muscle fibre bed can occur.[19] The importance of muscle glycogen during sustained near maximal exercise, as for example during flat racing in Thoroughbreds is more controversial.

It would appear that in man, with a glycogen content of <40 mmol glucosyl units kg^{-1} wet muscle (i.e. ~160 mmol glucosyl units kg^{-1} dry muscle (dm)) the muscles' capacity to produce lactate is compromised and exercise performance reduced.[12] A likely explanation is that glycogen is limiting to phosphorylase at these low muscle contents, though the apparent intracellular concentration (40 mmol kg^{-1} wet muscle corresponds to 13 mmol l^{-1} intracellular water) is still several times above the reported Michaelis constant (km)[4] of 2 mmol glucosyl units l^{-1}. However, for a complex molecule such as glycogen, reduction in the number of terminal glucosyl units available to phosphorylase may be more important than the absolute content measured.

In the normal Thoroughbred horse the muscle glycogen content at rest is already elevated above that found in normal man (550–600 mmol kg^{-1} dm compared to 300–400 mmol kg^{-1} dm),[6,11,17] though dietary and exercise management can increase the latter still further.[2] Despite the obvious requirement for muscle glycogen and rapid utilisation during galloping, total utilisation over flat race distances amounts to only 25 to 33% of the resting content.[7] The onset of metabolic acidosis, through the accumulation of lactate, is believed more likely to be the primary cause of fatigue during flat racing than the depletion of muscle glycogen. Glycogen depletion, however, could be a significant factor in National Hunt racing over 2 or more miles.

Concern has been voiced amongst trainers that a lowering in the resting glycogen content could occur during pre-race training due to inadequate resynthesis in between training sessions, and that this could adversely affect subsequent race performance. This

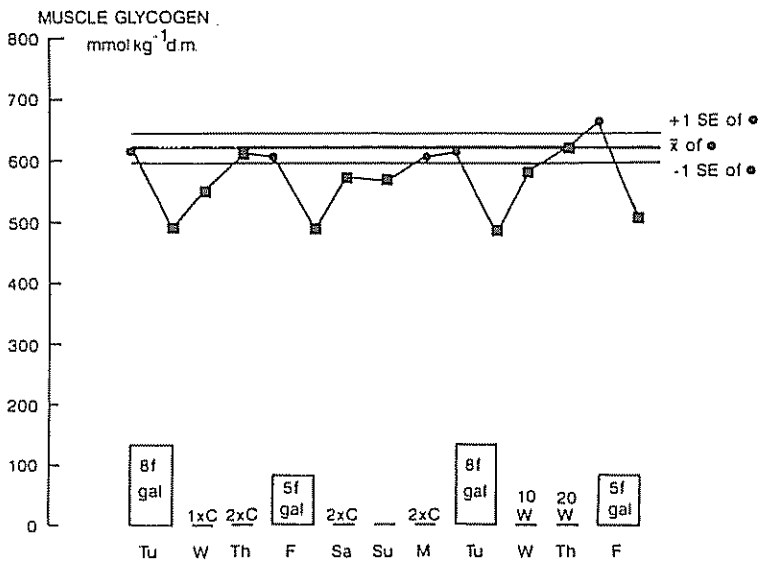

Fig 1 Mean changes in the glycogen content of the middle gluteal muscle during 2 weeks of training ●, Mean of glycogen contents on the 3rd or 4th day following training exercise of 5f or 8f. Points correspond to boxed data in Table 1. SE of ● = estimated standard error of individual means. SE of ● = (Sp(bswh)) 2/6 where Sp(bswh) is the pooled estimate of the square root of variance in the glycogen content at rest between sampling sites (× time), within horses from Table 1. gal = gallop, 8f = 1 600 m, 5f = 1 000 m; c = canter; w = walk

has prompted, in some training yards, the addition of carbohydrates supplements such as glucose and complex carbohydrates to the normal feed. The aim of the present study was to examine the pattern of glycogen depletion and repletion occurring during 2 weeks of training using a programme of exercise typical of that followed in many British training yards. This consists of 8f (1 600 m) and 5f (1 000 m) gallops on Tuesdays and Fridays of each week with light work on the remaining days (Sundays excepted).

MATERIALS AND METHODS

Six fit mature Thoroughbred horses were used in the study to examine the variation in glycogen content of the middle gluteal muscle over an 11 day training period. The study began on a Tuesday with a warm-up canter of 1 000 m followed by a timed gallop of 1 600 m on an all-weather track.

Details of the subsequent training programme are given in Fig. 1. On Day 2 one canter was given, whilst on Days 3, 5 and 7, two canters were performed. Each canter was approximately 1 000 m and was performed at a moderate pace with heart rate (HR) between 160 to 180 beats min^{-1}. On Days 9 and 10, horses were walked instead of cantering for 10 and 20 min. No exercise was performed on the Sunday, Day 6. The 1 600 m working gallops consisted of a 1 200 m swinging canter (i.e. horses were held on the bridle with HR around 200 beats min^{-1}), followed by a 400 m flat-out gallop run at maximal pace. The 1 000 m gallops were run at maximal pace throughout.

All horses were fed a mixed diet of pelleted feed (Spillers Racehorse cubes) and hay. Each day the horses were offered a total of 8.1 kg of pelleted feed divided into 3 rations of 1.8 kg (morning), 2.7 kg (midday) and 3.6 kg (evening). Hay was offered in the morning (2 kg) and the evening (2 kg). Feed left from the preceding day was collected and weighed. From this the mega-joules (MJ) of digestible energy (DE) fed per day was calculated.

Muscle biopsies were taken prior to the morning feed on each day from either the left or right middle gluteal with a 6 mm Bergström-Stille biopsy needle.[1] A second biopsy was taken on the days of galloping exercise shortly after the return of each horse to its box. This was approximately 45 min

after the end of the exercise. This biopsy was taken from the same side and within 3 cm of the one taken earlier. With the exception of these paired samples other biopsies were collected in a randomized fashion (8 from one side, 7 from the other) and all from a 10 cm^2 area of the middle gluteal located one third of the distance from the tuber coxae to the head of the tail. Biopsies were taken at a depth of 6 cm as described by Snow.[16] Samples were snapfrozen and subsequently freezed-dried before powdering to remove blood and connective tissue. Muscle glycogen was hydrolysed to free glucose with 1 mol l^{-1} HCl at 100°C for one hour and assayed in duplicate using hexokinase. Values are presented as mmol glucosyl units kg^{-1} dm.

A 3 ml blood sample was drawn from the jugular vein at the end of each 1 600 m and 1 000 m gallop, and deproteinized with 5 ml 1 mol l^{-1} perchloric acid. Lactate was determined in 20 µl of the acid extract by the method of Hohorst.[10]

HR was monitored during the gallops by means of an onboard HR meter (Hippocard PEH-3000). All results are presented ± standard deviation (SD).

RESULTS

Gallop times for 1 000 m averaged approximately 70 s with peak HR in excess of 200 beats min^{-1}. Post-exercise blood lactate concentrations over this distance averaged 19.6 ± 3.7 mmol l^{-1}. Gallop times for 1 600 m ranged from 113 to 122 s with peak HR again in excess of 200 beats min^{-1}, and post-exercise blood lactate concentrations averaged 21.6 ± 4.7 mmol l^{-1}.

Individual values for muscle glycogen over the 11 days of the study are given in Table 1 and mean values are shown in Fig. 1.

Gallops of 1 000 and 1 600 m resulted in a similar loss of muscle glycogen, approximating to 19–25% of the resting content. Following gallops at either distance glycogen contents were restored to normal limits within 2 to 3 days. Walking or cantering exercise on intervening days did not affect repletion rates.

From analysis of variance for repeated measures, there was no significant difference ($p > 0.05$) in the glycogen contents of biopsies taken on the third day after each gallop, i.e. those taken on days 4 (pre-exercise), 7, 8 (pre-exercise) and 11 (pre-exercise), and the content determined at the start of the study. Prior to the start of the study on the Tuesday, the horses had not been exercised since the previous Friday. These contents have, therefore, been assumed to be equally representative of that at rest. Means of these 5 samples from each horse ranged from 544 (Sa) to 724 (Br), with an overall mean of 623 mmol glucosyl units kg^{-1} dm. One-way analysis of variance indicated a difference between horses ($p < 0.001$). The estimated variance between sampling sites (×times), within horses—Sp(bswh)—was 57 mmol glucosyl units kg^{-1} dm.

With the 1 000 m gallop the change in muscle glycogen varied from +15 (an increase) to −265 mmol glucosyl units kg^{-1} dm. Mean changes for the two 1 000 m gallops were −115 and −157 mmol glucosyl units kg^{-1} dm. Variance beween horses within gallops in the response to exercise—Sp(bhwg)—was 97 mmol glucosyl units kg^{-1} dm; variance between gallops within horses—Sp(bgwh)—was 103 mmol glucosyl units kg^{-1} dm.

Variation in the response to exercise was relatively less at 1 600 m, the change in glycogen content varying from −47 to −207 mmol glucosyl units kg^{-1} dm. Means for the two 1 600 m gallops were −125 and −129 mmol glucosyl units kg^{-1} dm. Sp(bhwg) and Sp(bgwh) were 50 and 41 mmol glucosyl units kg^{-1} dm, respectively.

Details of the feed consumed are given in Table 2 with details of the composition of the feed in Table 3. Amounts of pelleted feed consumed varied considerably between and within horses and were as a low 4.9 kg on Day 9 for horse Mr. Estimated DE consumed per day was on average lowest in Mr,

Table 1. *Muscle glycogen contents (mmol glucosyl units kg^{-1} dm) of the 6 horses over the 11 days of the study*

Values considered representative of the content at rest are boxed

Biopsy	Day no.	Day	Horses						
			Mr	Bu	To	Cp	Sa	Br	$\bar{x}$
Pre 1 600 m	1	Tu	592	628	630	562	504	789	618
Post 1 600 m	1	Tu	483	526	505	353	418	673	493
	2	W	–	670	581	495	349	745	553
	3	Th	550	749	649	443	668	611	612
Pre 1 000 m	4	F	548	672	646	505	524	737	606
Post 1 000 m	4	F	352	685	533	441	442	489	490
	5	Sa	480	660	615	527	466	683	572
	6	Su	644	651	595	384	451	683	568
	7	M	624	702	594	613	495	613	607
Pre 1 600 m	8	Tu	517	623	608	632	608	710	616
Post 1 600 m	8	Tu	320	546	444	478	473	663	487
	9	W	571	673	464	591	475	720	582
	10	Th	584	530	640	646	572	779	625
Pre 1 000 m	11	F	520	761	648	711	590	771	667
Post 1 000 m	11	Fr	428	560	417	446	605	606	510
$\bar{x}$ of ☐ samples			560	677	625	605	544	724	623
SD			47	57	24	77	52	69	
Sp(bswh)									57

Sp(bswh) = Pooled estimate of the square root of variance between sampling sites (×time), within horses.

Table 2. *Daily feed consumption during the 11 days of the experiment*

		Horses					
		Mr	Bu	To	Cp	Sa	Br
kg pelleted feed	$\bar{x}$	6.0	7.5	7.6	6.3	7.1	7.7
	SD	0.8	0.6	0.9	0.9	0.6	0.6
kg hay	$\bar{x}$	3.2	2.3	3.3	3.0	3.7	3.2
	SD	0.7	0.8	0.8	0.6	0.5	0.6
Estimated DE (MJ day^{-1})		97.1	107.4	116.8	99.0	114.2	117.2
Body weight (kg)		416	418	424	462	448	474
DE (MJ day^{-1} kg^{-1} bwt)		0.233	0.257	0.275	0.214	0.255	0.247

Table 3. *Chemical composition of the concentrate and hay fed to horses*

	Pelleted feed	Hay[a]
Moisture (%)	12	15
% dry matter (DM)		
Crude protein	14	9.5
Fiber	9.5	35.9
Ash	7.5	–
Oil	3.3	–
Starch/sugar	38.5	–
Calculated DE (MJ kg^{-1} DM)	13.4	9.7

[a] Average figures for Timothy — Rye grass for 1987.

though expressed per kg body weight was lowest in horse Cp.

DISCUSSION

A frequent problem encountered in studies of muscle glycogen is the variance between different sampling sites. In a previous study this amounted to 12.6 mmol glucosyl units kg^{-1} dm when 4 sites within the same muscle were sampled at 3 to 6 hour intervals on the same day.[18] The higher estimate of between-site variance of 57 mmol glucosyl units kg^{-1} dm found in the present study includes variance in the resting content which is re-established 3 or more days after exercise, and the variance between equivalent sites on the left and right middle gluteal muscles. Specific variance due to site can probably be attributed to fibre variation[5,13]; type 1 fibres possibly having a lower glycogen content than type II's.[20] Variance between sites used for the pre- and post-exercise samples undoubtedly contributed to the marked variability between horses in their response to exercise. As seems reasonable this was notably less following the 1 600 m gallop.

The principal finding of this study was the return to normal resting glycogen content by the third day after exercise. Judged on the two sessions using gallops of 1 600 m, this occurred equally whether horses were cantered or given walking exercise over the following days.

It was originally intended to provide horses with approximately 136 MJ of DE per day, comparable to that given as the normal diet in our previous study.[18] In practice, however, horses consumed an average of only 108.6 MJ per day. The lowest recorded was 80.8 MJ by horse Mr on Day 9. These intakes are substantially below a reported range of 147 to 209 MJ of DE per day given to American and English Thoroughbreds in training,[9,14] though the mean is close to the 1989 NRC recommendation[15] of 110 to 125 MJ per day for horses (weighing 400–450 kg) in active training and racing. Despite some variance between horses in the MJ of DE consumed, the feed taken was clearly adequate in all cases in supporting full restitution of the muscle glycogen content within 3 days.

Mean rates of glycogen synthesis during the first 24 hours were 62 (SD 70), 82 (SD 77) and 95 (SD 91) mmol glucosyl units kg^{-1} dm day^{-1} following the 1 600 m gallops. These rates compare to a rate of 94 mmol glucosyl units kg^{-1} dm day^{-1} in our earlier study when horses were fed a low carbohydrate diet comprising 87 MJ of DE and consisting mainly of hay, but fall below rates of 155 mmol glucosyl units kg^{-1} dm day^{-1} observed with a normal and high carbohydrate diet of 136 MJ day^{-1} or more.[18] It is notable, however, that in the previous study which utilised an exercise model of two maximal gallops of 900 m, the initial depletion in glycogen content (mean 187 mmol glucosyl units kg^{-1} dm) was greater than in the present study. This may have contributed to the faster resynthesis rate observed in this earlier study with horses maintained on a normal diet of race cubes and hay (as in the present study).

It would appear from this and the earlier study,[18] that following glycogen depletion with hard exercise, the content returns quickly to high baseline values. Addition of

further carbohydrate to the feed, above that required to meet NRC energy requirements[15] does not appear to result in any "overshoot" in glycogen synthesis, as has been described for man.[2] This is further supported by a preliminary study in which the feeding of a complex carbohydrate, in addition to the normal diet, again failed to cause any "overshoot" in glycogen synthesis (Snow and Harris, unpublished data).

ACKNOWLEDGEMENTS

The authors gratefully acknowledge support for this study from the Horserace Betting Levy Board, London.

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