From the Department of Clinical Biochemistry, Royal Veterinary College, the Department of Physiology, Gymnastik- och idrottshögskolan, Stockholm, and the Equine Hospital, Solvalla, Stockholm, Sweden.

THE PHYSIOLOGICAL AND BIOCHEMICAL RESPONSE OF STANDARDBRED HORSES TO EXERCISE OF VARYING SPEED AND DURATION*

$\mathbf{B}\mathbf{y}$ Arne Lindholm and Bengt Saltin

LINDHOLM, ARNE and BENGT SALTIN: The physiological and biochemical response of standardbred horses to exercise of varying speed and duration. Acta vet. scand. 1974, 15, 310—324. — Well-trained standardbred horses were studied to examine the metabolic response to excercise of various speeds and duration. Comparisons between interval (400, 700, 1,000 and 2,000 m) and continuous trotting

(1 hr., 2 hrs.) and racing were made.

Muscle and rectal temperatures were recorded before and immediately after each work bout. Heart rate was linearly related to trotting speed, and maximal heart rate (240 beats × min.-1) was achieved when trotting at least 700 m at close to maximal speed (12.0—

12.5 m \times sec.⁻¹).

Biopsy specimens from the gluteus medius muscle and venous blood were obtained before and after each work bout.

Muscle and blood lactate values were markedly increased first at speeds close to maximal speed (11.4—12.5 m×sec.-1). Trotting 6×700 m at 12.5 m \times sec. $^{-1}$ produced as high muscle and blood lactate values as 23.7 and 19.0 mmol \times kg $^{-1}$ wet weight and l $^{-1}$, respectively. Corresponding values after a race were about 15 mmol \times kg $^{-1}$ (muscle) and l $^{-1}$ (blood).

Glycogen utilization was related to work intensity and was most pronounced during the first work bouts. At a speed of 12 m×sec.⁻¹ and trotting 2000 m, there was a glycogen utilization of near 12 mmol glucose units × kg⁻¹ × min.⁻¹ wet muscle.

It is concluded that interval training over a distance of 700—

1000 m repeated 4—6 times with a trotting speed close to maximal speed (11.4—12.5 m \times sec.⁻¹) appears to be optimal.

ATP; CP; blood lactate; glycogen utilization; heart rate; horse skeletal muscle; muscle lactate; racing training.

^{*} This study was supported by grants from the Swedish Medical Research Council and the Wallenberg Foundation.

Blood lactate values have been found to be fairly low in trotting horses, even at exercise intensities eliciting a marked increase in heart rate (Asheim et al. 1970, Krzywanek et al. 1972, Engelhardt et al. 1973). Blood lactate concentrations of the same magnitude as after races were not observed until the speed was great enough to produce near-maximal heart rate response (Asheim et al.).

If the aim of a training program is to mimic the physiological stresses involved in racing, very high trotting speeds for extended periods of time appear to be called for. The present study was undertaken in an attempt to shed further light on critical speed levels and the extent to which interval training programs can be used. In addition to heart rate, temperature and blood lactate concentration as parameters in quantifying exercise stress, glycogen utilization and the muscular concentration of lactate, adenosine triphosphate (ATP) and creatine phosphate (CP) were also determined.

MATERIAL, PROCEDURE AND METHODS

The material comprised a total of 44 standardbred horses (25 males and 19 females) from 4 to 8 years. All horses studied were clinically healthy with regard to cardiopulmonary function and were in good training condition. They were regularly raced and able to trot 2100 m at a speed of 11.8 m×sec.⁻¹ (1 min. 25 sec.×km⁻¹) or better. The horses were rested in the stable 24 hrs. prior to the experiments and were fed in the morning, as usual, prior to the investigations. No special diet regimen was proposed, and all studies were performed between 9 and 12 p.m.

Trotting intensities from 3.3 m×sec.⁻¹ up to 12.5 m×sec.⁻¹ were employed in order to investigate different training and racing conditions. Interval training with exercise sessions of varying length (400, 700, 1000 and 2000 m) and intensity plus 1 or 2 hrs. of long-distance trotting were also investigated. All studies were performed at Solvalla race track in Stockholm, Sweden. The track has a length of 1000 m. The test room was situated in the Solvalla Horse Hospital which is located very close (< 10 m) to the track. The experiments were only performed under good weather and track conditions but with varying outdoor temperatures, as the experiments comprised studies both in winter and summer.

Biopsies and venous blood samples were taken from the horse at rest, in the test room and immediately after each work bout as soon as the horse entered the test room. At this time measurements of muscle and rectal temperatures were also made. Each work bout ended at the part of the track closest to the test room and it took an average of 20 sec. from the end of exercise before sampling could be started. The biopsies, blood samples and temperature measurements were made within 20 sec. to 2 min. after the end of each work bout. Between work bouts, the horse rested for 5 min. in the test room. The biopsies were taken from the deep part of the gluteus medius muscle as described by Lindholm & Piehl (1974). After careful shaving and disinfection, 2-3 ml of 2 % Xylocaine (Astra, Sweden) was injected s.c. 10 cm dorso-caudal to the trochanter major of the tuber coxae. A 5 mm incision was made with a scalpel including both skin and fascia, and a biopsy needle (Bergström 1962) with an external diameter of 5 mm was used. Muscle samples weighing 20-40 mg were obtained when the inner cylinder was pushed back and forth 1-2 times. Biopsy sampling took 1-3 sec. Specimens were immediately frozen in liquid nitrogen. Determinations of glycogen, adenosine triphosphate (ATP), creatine phosphate (CP), glucose-6-phosphate (G-6-P), glucose and lactate in the biopsies were made with the Lowry technique as described by Karlsson et al. (1970). Venous blood was taken from the jugular vein. Glucose determinations were made using whole blood kept in plastic tubes containing NaF (sodium fluoride) and placed in ice-cold water until subsequently analyzed on the same or next day according to a glucose oxidase method (Hjelm & de Verdier 1963). For analyses of blood lactate, 0.2 ml of whole blood was immediately pipetted into 1.0 ml of ice-cold 0.6 N-HClO4 (percloric acid) and lactate determined by a micro-modification of an enzymatic method (Scholz et al. 1959).

Muscle (gluteus medius) and rectal temperatures were determined by electro-thermometry (Electric Universal Thermometer, Type E 3). Heart rate was telemetrically recorded on a single channel recorder according to methods described by Asheim et al. (1970), using a telemetric equipment (Medinic, Örsundsbro, Sweden) with the receiver at the Horse Hospital close to the track. Two silver-plated crocodile clamps were used. One was attached 10 cm cranial to the apex of the sternum and the other to the supraorbital process of the frontal bone on the forehead.

RESULTS

Trotting 1000 and 2000 m at different speeds (Figs. 1 and 2).

At rest in the test room heart rate was usually around 40 beats \times min.⁻¹. As soon as the horse started to trot, a rapid increase in heart rate was noted. Within 30—45 sec. a new and fairly stable level was attained, as long as the speed was kept constant. Exercise heart rate averaged 140 beats \times min.⁻¹ even at a speed as low as 3.3 m \times sec.⁻¹ (5 min. \times km⁻¹). At higher speeds up to the speed the horse was able to sustain for 1000 m, a linear increase was found for the heart rate related to work intensity; maximal heart rate being around 240 beats \times min.⁻¹.

Mean muscletemperature, which was 36.8°C at rest was higher at each exercise level, averaging 41°C after the 1000 m at maximal speed. Exercise duration in all these work bouts was fairly short (1 min. 20 sec. — 5 min.). Therefore, the dura-

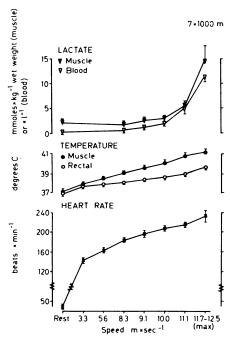


Figure 1. Mean values and s.e.m. for muscle and blood lactate concentrations (upper panel), muscle and rectal temperatures (middle panel) and heart rate (lower panel) in 6 standardbred horses when trotting 7×1000 m with increasing speed from 3.3 m×sec.-1 to maximal speed (11.7—12.5 m×sec.-1). Five min. of rest was allowed between work bouts.

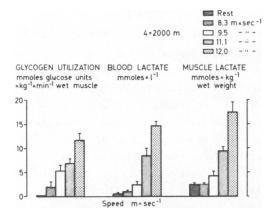


Figure 2. Mean values and s.e.m. for glycogen utilization, blood lactate and muscle lactate concentrations in 5 standardbred horses before and after trotting 2000 m at 4 different speeds (8.3—12.0 m× sec.-1). Only 1 experiment was performed on the same day.

tion may not have been long enough for the muscle temperature to reach an equilibrium at each trotting speed. This must be the case to an even greater extent for rectal temperature for which values up to 39.4°C was recorded.

At speeds up to 10 m \times sec.⁻¹ (1 min. 40 sec. \times km⁻¹) only a very minor increase in muscle and blood lactate was found. Not until a speed of 11.1 m \times sec.⁻¹ was there any further significant increase in muscle and blood lactate concentrations, which were more dramatically elevated at maximal speed (15 mmol \times kg⁻¹ and 12 mmol \times l⁻¹ for muscle and blood lactate, respectively).

Trotting 2000 m at different speeds resulted in almost the same heart rate response as found for 1000 m. At the lower speeds, muscle temperature was 0.2—0.5°C higher when trotting the longer distance. At the highest speeds, no clearcut differences were observed, and essentially the same peak muscle temperature was also found. Rectal temperature was slightly higher at the same speed trotting for twice the distance reaching 39.5°C at maximal trotting speed.

Glycogen utilization could not be accurately estimated at the 1000 m distance. Trotting 2000 m at a slow to moderate speed was found to produce — as expected — varying glycogen utilization in relation to exercise intensity (Fig. 2). At the highest speed (12 m×sec.⁻¹), almost 32 mmol×kg⁻¹ were broken down

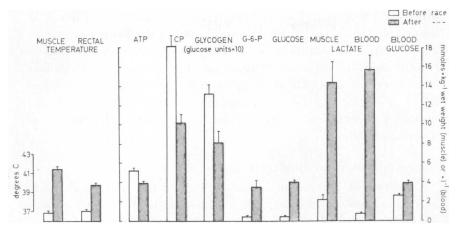


Figure 3. Mean values and s.e.m. for muscle and rectal temperatures, i.m. concentrations of ATP, CP, glycogen, G-6-P, lactate and glucose, blood lactate and glucose concentrations for 5 standardbred horses before and after a real race. Distance 2100 m. Mean speed 11.8 m×sec.⁻¹. Biopsies were obtained from the gluteus medius muscle within 2—3 min. after finish of the race. Note the different scales for temperatures and metabolites.

in 2 min. 50 sec., which is 6 times faster than at $8.3 \text{ m} \times \text{sec.}^{-1}$. Peak muscle and blood lactate levels were higher in trotting 2000 m, but almost identical lactate concentrations were found at the slower speeds.

Racing (Fig. 3)

Studies of temperature and metabolites could be performed on 4 horses in connection with races. Muscle and rectal temperatures reached 41.5 °C and 39.8 °C, respectively. In spite of the fact that the biopsies were taken 2—3 min. after the race, phosphagen levels were still down to 60—70 % of their normal values. As much as 50 mmol \times kg⁻¹ of glycogen were broken down in the gluteus medius muscle. As very little glycogen is probably utilized in the pre-race warm-up (cf. Fig. 2), most of the glycogen breakdown must have occurred during the race.

Another indication of the enhancement of glycolysis during the race was the 8 to 10-fold increase in G-6-P and glucose levels. In fact, the blood glucose level appeared to be somewhat lower than the muscle glucose level. Muscle and blood lactate concentrations were greatly elevated, and 14.4 mmol×kg⁻¹ and 15.7

mmol \times l⁻¹ were observed, respectively. The sampling time 2—3 min. after the race explains the similarity in the 2 concentrations. The lactate concentration is probably also higher in the muscle fibre than in the capillary at this time. Lactate concentration was also determined on blood samples from 38 other standardbred horses 3 min. after a race. The mean value was 15.9 \pm 0.4 (s.e.m.) mmol \times l⁻¹, which is in very close agreement with the aforementioned value.

Interval training programs (Figs. 4—6)

A series of experiments was performed in order to evaluate how closely one can attain racing values for the different variables studied in interval training of varying speeds. Six times 400 m was studied first, the horse trotting at near-maximal speed (12.2—12.4 m \times sec.⁻¹) in each work bout. Heart rate approached 220 beats \times min.⁻¹ in the first exercise period, and no further significant increase was found during the remaining periods. The same appeared to be the case for muscle lactate which amounted to 13.0 mmol \times kg⁻¹ after the second work bout and 13.8 mmol \times kg⁻¹ after the final work bout. Blood lactate increased more gradually and reached a final value of 12 mmol \times l⁻¹. Total glycogen depletion in the gluteus medius muscle amounted to 40 mmol \times kg⁻¹.

When trotting was performed for 700 m and repeated 6 times, the critical nature of speed in attaining true maximal values could be demonstrated. Although all 3 speeds used were close to maximal speed, only the highest speed, 12.5 m×sec.⁻¹, was high enough to elicit heart rate, body temperature and muscle and blood lactate responses equal to or greater than the response encountered in the race. Of further note may be the fact that glycogen utilization was most pronounced during the first and second work bouts. The increase in lactate concentration produced in muscle and blood at each work bout must then be, in part, the result of uncomplete lactate removal during rest periods.

The same general results were observed in trotting 6×1000 as in trotting 6×400 and 6×700 m. The importance of speed in eliciting a maximal physiological response was again demonstrated. A speed of 11.1 m \times sec.⁻¹ may be sufficient to stress the circulatory system, but a speed of only 11.8 m \times sec.⁻¹ caused muscle and blood lactate values to approach very high levels. This is in accordance with the findings reported in Fig. 1.

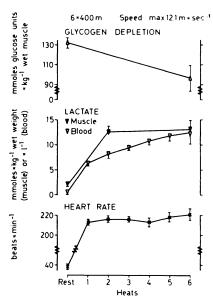


Figure 4. Mean values and s.e.m. for glycogen depletion (upper panel), muscle and blood lactate concentrations (middle panel) and heart rate (lower panel) for 4 standardbred horses before and after trotting 6×400 m at maximal speeds (average 12.1 m×sec.-1). Muscle and blood lactate concentrations and heart rate were also determined after each 400 m run. One min. of rest was allowed between work bouts when muscle and blood samplings were performed.

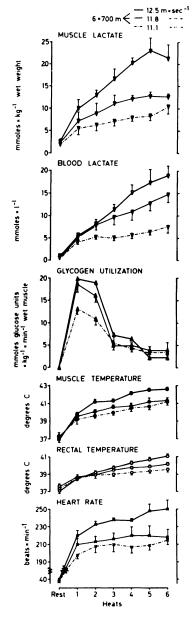


Figure 5. Mean values and s.e.m. (from top to bottom) for muscle and blood lactate concentrations, glycogen utilization, muscle and rectal temperatures for 5 standardbred horses at rest and after each work bout, trotting 6×700 m at 3 different speeds (11.1, 11.8 and 12.5 m \times sec.⁻¹). Only 1 speed was performed on the same day. Heart rate was recorded during each work bout. A 5-min. rest was allowed between work bouts.

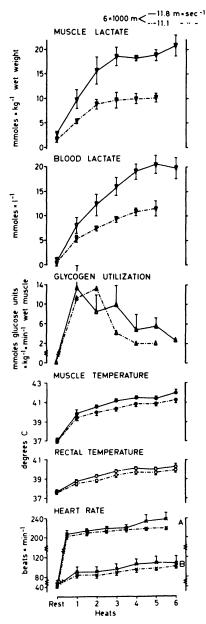


Figure 6. Mean values and s.e.m. (from top to bottom) for muscle and blood lactate concentrations, glycogen utilization, muscle and rectal temperatures and heart rate for 4 standardbred horses at rest and after each work bout, trotting 6×1000 m at 2 different speeds (11.1 and 11.8 m \times sec.⁻¹). Only 1 speed was performed on the same day. Heart rate was also recorded before (B) and during each work bout (A). A 5-min. rest was allowed between work bouts.

Traditional training programs (Figs. 7 and 8)

For purposes of comparison, studies were also made of normal training programs for the horses at this track. In addition to very slow trotting for 60 to 120 min. a day, which caused no or only a minor increase in heart rate, temperature and lactate (Fig. 8), the normal training programs also comprised some speed training 1 to 2 times a week. The usual distance was 2100 m which is first repeated twice (a rest period of a couple of min. inbetween) at a speed of around 75 % in the first and 85 % of maximal speed in the second work bout. After a rest period of 30 min. the horse then trotted a third time, this time at maximal speed. As might be expected, all the observed variables only achieved maximal or racing levels in the last work bout. This was especially true of muscle and blood lactate concentrations.

DISCUSSION

The present study confirms previous results, i.e. that the maximal heart rate in horses is very high compared to other species of the same size (Bayer 1968, Asheim et al. 1970, Hall 1972, Ehrlein et al. 1973). Both speed and duration of excercise have a decisive effect on heart rate, and a maximal heart rate is only achieved after maximal trotting for at least 700 m at top speed. With a longer excercise duration and 1000 m or more of trotting, the same heart rate can be achieved at a speed close to the maximal trotting speed.

Lactate accumulation increased strikingly at maximal speed or near-maximal speed (11.4—12.5 m×sec.⁻¹). Lactate values were then equal to or greater than the values obtained during a race. Exercise sessions covering 400 m were obviously too short, even at maximal speeds, to produce the same lactate concentration as in a race. However, repeated exercise sessions covering 700 and 1000 m were shown to be more suitable as training distances at maximal or near-maximal speeds.

A 5-min. rest between exercise episodes was too brief to permit the removal of lactate from blood and muscle. Even at distances greater than 2000 m, elevated lactate values on racing levels were only obtained at maximal speed or very near-maximal speed. Thus, traditional training with 3—4 2000-m heats with successively increased speed only produces pronounced anaerobic loading in the fastest and final heat (Fig. 8). Since a maximal lactate

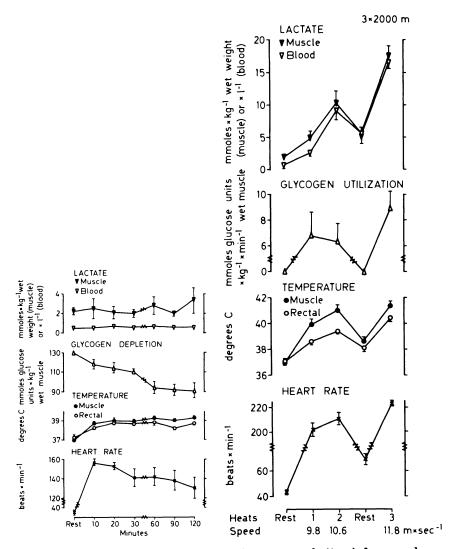


Figure 7. Mean values and s.e.m. (from top to bottom) for muscle and blood lactate concentrations, glycogen depletion, muscle and rectal temperatures and heart rate for 4 standardbred horses at rest and after various time courses when slow-trotting (average speed 3.3 m×sec.-1) for 2 hrs. The horses rested for 3 min. during the sampling procedure. Heart rate was recorded when trotting immediately before each sampling occasion.

Figure 8. Mean values and s.e.m. (from top to bottom) for muscle and blood lactate concentrations, glycogen utilization, muscle and rectal temperatures for 7 standardbred horses at rest and after trotting 2000 m at 3 separate heats (speed 9.8, 10.6 and 11.8 m×sec.⁻¹). During each heat heart rate was recorded when trotting the last 500 m. There was a 5-min. rest between heats 1 and 2 for the sampling procedure and 30 min. of rest between heats 2 and 3.

increase is only obtained at maximal or near-maximal speed, a distance even over 2000 m may be too long and too taxing for the immature skeletons of 2—4-year old horses at the speeds required. That is why an increased number of shorter training intervals would be advantageous.

Since the 400 m interval is apparently too short with a view to heart rate and lactate increases, 700 and 1000 m appear to be more ideally suited to horses. Further support for this view is found in Fig. 9, which shows the heart rate, blood and muscle lactate concentrations from 1 standardbred horse after different types of interval training. The heart rate response during a race was similar to that found in an interval training program consisting of 6×1000 m sessions with a speed close to maximal speed. The same type of training was also enough to raise blood and muscle lactate to about the same values measured during the race; shorter distances produced a lower lactate level and 3 2000-m sessions produced muscle and blood lactate values equally high (23.8 mmol \times kg⁻¹ and 20.6 mmol \times l⁻¹, respectively).

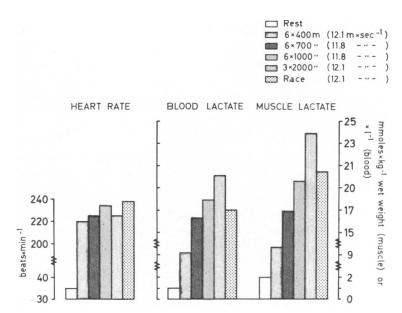


Figure 9. Heart rate, blood and muscle lactate values from 1 standardbred horse at rest and after trotting 6×400 , 6×700 , 6×1000 , 3×2000 m, as well as after a real race. Speed close to maximal (11.8 m \times sec.⁻¹) or maximal (12.1 m \times sec.⁻¹).

Since the horse must exercise with maximal work loads in order to achieve efficient training, high working temperatures are obtained in both muscle and rectal measurements. A muscle temperature of 43°C and a rectal temperature of 41° have been measured. This means that at maximal speed a horse exercises at a critical temperature level, which might have a negative effect on the performance, especially in the races.

Horse musculature is rich in glycogen, compared to other species, including man (Lindholm & Piehl 1974). Glycogen values never fell to critically low levels (low enough to impair the horse's performance capability owing to e.g. carbohydrate deficiency) in any of the results from training models nor in the race presented here.

The horse has 3 major fibre types (Lindholm & Piehl). In addition to the slow twitch (ST) and fast twitch (FT) fibres found in man, the horse, like most other animal species, also has fast twitch, high oxidative (FTH) fibres. All contain glycogen, even if ST fibres appear to have less than the other types. At low speeds mainly ST and FTH fibres, both of which with a large oxidative capacity, are put to use (Lindholm et al. 1974). Fibres with a low oxidative capacity (FT) are only utilized from the start of work when the horse trots at very close to its maximal speed. This explains why lactate in greater amounts only accumulates in this situation. It also means that if such a speed is not used during training, this fibre type which constitutes 43 % of the cross-sectional area of horse muscle, remains inactivated except in races.

REFERENCES

- Asheim, A., O. Knudsen, A. Lindholm, C. Rülcker & B. Saltin: Heart rates and blood lactate concentrations of standardbred horses during training and racing. J. Amer. vet. med. Ass. 1970, 157, 304—312.
- Bayer, A.: Das Verhalten der Herzschlagfrequenz von Trabrennpferden im Training und beim Rennen. (Heart rate in standardbred trotters during training and racing). Berl. Münch. tierärztl. Wschr. 1968, 81, 8—11.
- Bergström, J.: Muscle electrolytes in man. Scand. J. clin. Lab. Invest. 1962, Suppl. 68.
- Ehrlein, H.-J., H. Hörnicke, W. v. Engelhardt & G. Tolkmitt: Die Herzschlagfrequenz während standardisierter Belastung als Mass für die Leistungsfähigkeit von Pferden. (Heart frequency during standardized exercise as a measure of the working capacity of horses). Zbl. Vet.-Med. A. 1973, 20, 188—208.

- Engelhardt, W. v., H. Hörnicke, H.-J. Ehrlein & E. Schmidt: Lactat, Pyruvat, Glucose und Wasserstoffionen im venösen Blut bei Reitpferden in unterschiedlichem Trainingszustand. (Lactate, pyruvate, glucose and hydrogen ions in venous blood of riding horses at different stages of training). Zbl. Vet.-Med. A. 1973, 20, 173—187.
- Hall, M. C.: Cardiac monitoring during exercise tests in the horse. Thesis. Melbourne 1972.
- Hjelm, M. & C. H. de Verdier: A methodological study of the enzymatic determination of glucose in blood. Scand. J. clin. Lab. Invest. 1963, 15, 415—428.
- Karlsson, J., B. Diamant & B. Saltin: Muscle metabolites during submaximal and maximal exercise in man. Scand. J. clin. Lab. Invest. 1970, 26, 385—394.
- Kryzwanek, A., A. Schulze & G. Wittke: Das Verhalten einiger Blutparameter bei Trabrennpferden nach definierter Belastung. (The behaviour of some blood parameters in trotters after defined work). Berl. Münch. tierärztl. Wschr. 1972, 85, 325—329.
- Lindholm, A., H. Bjerneld & B. Saltin: Glycogen depletion pattern in muscle fibres of trotting horses. Acta physiol. scand. 1974, 90, 475—484.
- Lindholm, A. & K. Piehl: Fibre composition, enzyme activity and concentrations of metabolites and electrolytes in muscles of standardbred horses. Acta vet. scand. 1974, 15, 287—309.
- Scholz, R., H. Schmitz, T. Bücher & J. O. Lampen: tber die Wirkung von Nystatin auf Bäckerhefe. (The effect of nystatin of yeast). Biochem. Z. 1959, 331, 71—86.

SAMMANFATTNING

Det fysiologiska och biokemiska svaret på träning av varierande intensitet och utsträckning hos travhästar.

Vältränade travhästar studerades för att undersöka effekten av olika typer av travarbeten på vissa metaboliska parametrar. Intervallträning (400, 700, 1000 och 2000 m), distansträning (1 och 2 tim) samt tävling har jämförts.

Muskel- och rektaltemperatur mättes före och omedelbart efter varje travpass. Hjärtfrekvensen befanns vara direkt relaterad till travhastigheten. Den maximala hjärtfrekvensen (240 slag×min⁻¹) registrerades vid travarbete nära maximal fart (12,0—12,5 m×sek⁻¹) i minst 700 m.

Muskelbiopsier från gluteus medius samt blod från vena jugularis togs före och efter varje travpass. Koncentrationen av muskel- och blodlaktat steg kraftigt först vid farter nära maximal fart (11,4—12,5 m \times sek⁻¹). Vid 6 st 700 m travarbeten (12,5 m \times sek⁻¹) med 5 min intervall erhölls så höga muskel- och blodlaktatvärden som 23,7 och 19,0 mmoler \times kg⁻¹ våt muskel respektive l⁻¹ blod. Motsvarande tävlingsvärden uppgick till 15 mmoler \times kg⁻¹ våt muskel respektive l⁻¹ blod.

Glykogen förbrukningen var relaterad till arbetsintensiteten och var störst under de första passen vid intervallträningen. Vid en hastighet av 12 m \times sek $^{-1}$ och 2000 m distans var glykogenförbrukningen nära 12 mmoler glukosenheter \times kg $^{-1}$ våt muskel \times min $^{-1}$.

Sammanfattningsvis konstateras att intervallträning 700—1000 m upprepade 4—6 ggr i nära maximal fart (11,4—12,5 m \times sek⁻¹) synes ge optimal träningseffekt.

(Received January 7, 1974).

Reprints may be requested from: Arne Lindholm, Department of Clinical Biochemistry, Royal Veterinary College, Fack, S-104 05 Stockholm 50, Sweden.