REVISTA ESPAÑOLA DE FISIOLOGIA, 51 (2), 55-64, 1995

Blood Parameter and Heart Rate Response to Training in Andalusian Horses

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(Received on October 14, 1994)

E. I. AGÜERA, M. D. RUBIO, R. VIVO, R. SANTISTEBAN, A. MUÑOZ and F. CASTEJÓN. *Blood Parameter and Heart Rate Response to Training in Andalusian Horses.* Rev. esp. Fisiol. (J. Physiol. Biochem.), 51 (2), 55-64, 1995.

A study was performed on Andalusian horses in order to assess the response of heart rate and various blood parameters to training. Two tests were performed, at two and four months of training respectively. Exercise schedules were of increasing intensity, over a distance of 1000 meters. Speed was progressively increased, from 4 m/s to 8.5 m/s, over four exercise stages. In both tests, a recovery period of 5 min followed each stage. Sample collection (by puncture of the external jugular vein) was performed with the animals at rest, within the first minute after each exercise stage, and at 10, 15, 20 and 30 min of final recovery. Samples for analysis contained plasma for measurement of lactate, glucose, ion and creatinine levels. Heart rate was measured using the Polar Sport tester. The most important parameters in both tests proved to be glucose level, heart rate and lactate concentration. Variations in electrolyte and creatinine levels were transitory, normal resting values being regained after 30 minutes' recovery. Response to daily training was most clearly reflected in altered lactate levels and heart rate; recovery improved with increased training which enhanced aerobic capacity and decreased metabolic acidosis.

Key words: Andalusian horses, Blood biochemistry, Heart rate, Lactate, Response to Training.

The training condition of horses is generally assessed by standardized exercise testing in which heart rate, speed and lactate production values are measured.

Hematology and blood biochemistry are often used in assessing the health of *Trotters and English Thoroughbreds* (45). In a large number of studies reviewed by CARLSON (8), various equine blood parameters were contrasted before, during and after exercise in order to evaluate the adaptations induced by training, to monitor the physiological response provoked by exercise-related stress, to evaluate an abnormal response to exercise and to calculate performance potential.

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Many studies have been performed relating lactic acid production to blood biochemistry in English Thoroughbreds (44), Trotters (25), endurance-ride horses (7), and showjumpers (3). Plasma parameters have been studied in horses undergoing prolonged exercise schedules (10), but have not been related to either lactate levels or heart rate. Variations in plasma parameters as a function of exercise have also been determined in Spanish horses (35), but until now there has been little research into the inter-relationship between lactic acid, hematology and blood biochemistry in Andalusian horses as a response to training.

The present paper describes the effects of training on variations of specific plasma biochemical values, showing that regular training at a given intensity modifies certain plasma and biochemical values (30), as well as affecting heart rate.

Materials and Methods

Sixteen four-year-old male Andalusian Horses from the Military Stud at Jerez de la Frontera (Cadiz, Spain) were used for this study. All available animals were included in the study, and no form of selection was practised.

Animals were housed in individual boxes 3 m wide and 4 m long, and fed three times a day with rolled oats and barley (1.5 kg/100 kg live weight).

Initial lunging sessions were carried out with horses unmounted, during which time they became accustomed to the saddle; at a later stage, the same exercises were repeated mounted. The traditional exercise model was followed for a period of roughly 2-3 months. Subsequently, horses underwent regular exercise (training) from Monday to Friday, in a 40-hectare paddock of Mediterranean scrub; ground was sandy-loamy and uneven, with shallow ditches. The exercise consisted of 15-30 minutes' walking; 25-30 minutes' trotting; 12-18 minutes galloping, in the following sequence: walking → 5-7 minutes trotting in each hand → trotting (transition to galloping) → 5-9 minutes' galloping → trotting → walking.

The experiment was performed after two and four months of training.

Tests were performed on a semi-elliptical sand track 1,000 m long and 6 m wide, marked with a red stripe at 100-meter intervals (21).

The exercise schedule consisted of four stages: Stage 1 at a speed of 4 m/s; stage 2 at 5.5 m/s; stage 3 at 7 m/s and stage 4 at 8.5 m/s. Animals were rested for 5 min after each stage.

Blood samples were taken by puncturing the external jugular vein while the animal was at rest, within one minute of finishing each stage and subsequently at 10, 15, 20 and 30 minutes of final recovery.

Samples were placed in one test-tube containing heparin/lithium: for immediate centrifuging, refrigeration and subsequent laboratory analysis; two aliquots of plasma were obtained, one of which was kept under refrigeration and the other frozen.

Samples for analysis contained plasma for the measurement of lactate (*Champion P-LM5* portable lactacidometer), glucose and creatinine (Ames' Quik-lab 2.0 spectrophotometer) and ions $Na^+/K^+/Cl^-$ (Ciba-Corning 644 Selective Electrode Analyzer).

Heart-rate was measured by using a *Polar Sport* tester (42) during exercise and by thorax auscultation during rest and recovery.

Statistical procedures included calculation of the mean and of standard deviation; discriminant analyses (27) were also performed between the 8 parameters studied in order to compare the effects of 2 months' and 4 months' training, and also to establish which variables show the greatest difference between tests.

Data obtained during the recovery period were subjected to regression analysis by using an exponential model. Correlation coefficients were obtained for parameters studied at rest, during exercise and during recovery. A two-way analysis of variance was used for inter-test and inter-stage comparison.

Results

Inter-test discriminant analysis yielded significant results, indicating differences between tests (Chi-square = 144.13; G.L. = 8; $p < 0.001$; canonical coefficient = 0.87). Standardized coefficients of the discriminant function showed the most important parameters to be: glucose (0.8646); heart rate (0.5833) and lactate (–0.5510), while electrolytes and creatinine (table I) proved to be less important.

Table II shows the exponential-model regression analysis performed during the recovery period, with time as an independent variable. Parameters not indicated in the table failed to fit any model of regression.

Glucose.– Mean plasma glucose values for test 1 (2 months' training) varied between 7.63 mmol/l (± 1.87) at rest, and 8.15 mmol/l at the fourth exercise. After 20 minutes' recovery, glucose regained rest values (fig. 1).

Initial rest values in test 2 (4 months' training) were lower than those of test 1, at 5.10 mmol/l, and mean values reached only 5.59 mmol/l.

The overall mean for the first test was 7 mmol/l, and for the second 5.26 mmol/l; an inter-test difference of 2.14 mmol/l was recorded.

Analyses of correlation yielded highly significant Test 1 values for lactate during rest and exercise ($r = 0.3784$), and for heart rate ($r = 0.8277$) and lactate during recovery ($r = 0.5855$).

Two-way analysis of variance performed for inter-test comparison gave values of $F = 118.00$ and $p<0.001$; inter-stage values were $F = 2.41$ and $p < 0.05$, the effect of training is shown at 20 and 30 minutes' recovery (fig. 1).

Heart rate.– Initial resting values differed between tests, but in both cases increased with exercise: to 191 /min in the first test and 179/min in the second. Maximum values reached were 226/min and 211/min for Test 1 and 2, respectively (fig. 2).

Overall mean values were 112.88/min (Test 1) and 103.49/min (Test 2), with a mean inter-test loss of 9.39/min. Two-way analysis of variance gave values of $F = 5.27$ and $p < 0.05$ for training, with inter-stage values of $F = 93.90$ and $p < 0.001$. The training effect is shown at 20 and 30 minutes' recovery ($p < 0.05$ and $p < 0.01$) (fig. 2).

Correlation analyses performed for Test 1 parameters during rest and during training yielded a highly significant positive correlation for lactate ($r = 0.5187$). During the recovery period, the most significant values obtained were for lactate ($r = 0.7276$), glucose ($r = 0.8277$) and chlorine ($r = -0.6805$).

In the second test, correlations were noted for lactate between rest and exercise ($r = 0.5075$) and for potassium ($r = 0.5168$). During the recovery period, the only important correlation noted was for lactate ($r = 0.7737$).

Lactate.– Mean lactate values in test 1 ranged from 0.92 mmol/l at rest to 9.6 mmol/l at the end of maximal training. During recovery, plasma lactate levels fell to a mean 2.52 mmol/l. In the second test, though rising with exercise, mean lactate concentrations did not reach the peak values recorded in test 1 (fig. 3).

Table I. Parameters analysed (mean ± SD) during Rest, Exercise and Recovery. Tests 1 and 2.

	REST	EXERCISE (m/s)				RECOVERY (min)			
		4	5.5	7	8.5	10	15	20	30
TEST 1									
Na^+	129.66 (±2.91)	128.33 (±3.55)	128.73 (±3.95)	129.8 (±3.40)	129.86 (±4.18)	129.6 (±4.08)	129.2 (±2.88)	128.6 (±3.06)	130.46 (±4.67)
						*	**	**	
K^+	4.10 (±0.44)	4.53 (±0.59)	4.40 (±0.34)	4.42 (±0.46)	4.53 (±1.01)	3.85 (±0.34)	3.76 (±0.39)	3.64 (±0.38)	3.67 (±0.25)
Cl^-	93.46 (±9.89)	92.2 (±10.94)	94 (±7.20)	93.8 (±.25)	91.8 (±7.35)	92.4 (±7.21)	91.6 (±7.79)	91.53 (±8.70)	92.73 (±9.52)
Creatinine	118.89 (±15.36)	112.77 (±15.80)	136.22 (±13.63)	127.44 (±19)	142.33 (±24.35)	130.11 (±25.76)	127.33 (±10.29)	126.77 (±17.76)	126.88 (±14.50)
TEST 2									
Na^+	130 (±3.29)	129.5 (±4.10)	130.62 (±1.50)	129.37 (±1.68)	130.87 (±1.45)	129.87 (±7.21)	130.5 (±1.41)	130 (±2.13)	130.12 (±2.16)
		***	***	***	**				
K^+	4.01 (±0.33)	4.87 (±0.48)	4.75 (±0.34)	4.85 (±0.43)	4.72 (±0.45)	3.95 (±0.35)	3.84 (±0.26)	3.84 (±0.32)	3.70 (±0 .70)
Cl^-	97.37 (±2.92)	97.25 (±3.61)	96.37 (±2.61)	95.62 (±2.38)	95.75 (±1.66)	95.75 (±5.09)	96.26 (±1.06)	96.37 (±2.13)	96.5 (±2.07)
Creatinine	107.62 (±9.67)	114.5 (±9.89)	112.5 (±10.04)	117.37 (±16.79)	130.37 (±22.84)	108.12 (±14.57)	116.25 (±14.19)	109.75 (±16.30)	110.25 (±11.71)

Difference of means between rest, the different exercise schedules and recovery, by Student's *t* test.

Table II. *Exponential-model regression analysis for the recovery period, with time as independent variable.* Y = exp (a-bx); Se = standard error; r = correlation coefficient; p = probability; a = cut-off point; b = slope. I and II = test 1 and test 2, respectively. H. R. = Heart rate.

Dependent Variable	a	b	Se	r	p
$Lactate_I$	2.2473	0.0491	0.5341	−0.6820	0.0000
$Lactate_{II}$	1.8868	0.0434	0.4574	−0.6978	0.0000
$H.R._I$	5.1021	0.0435	0.3040	−0.8433	0.0000
$H.R._{II}$	4.9171	0.0454	0.2894	−0.8493	0.0000
K^+_I	1.4436	6.3248	0.1248	−0.4566	0.0004
K^+_{II}	1.4968	7.6311	0.0953	−0.6346	0.0001
$Creatinine_{II}$	4.8036	4.6679	0.1416	−0.3202	0.4392

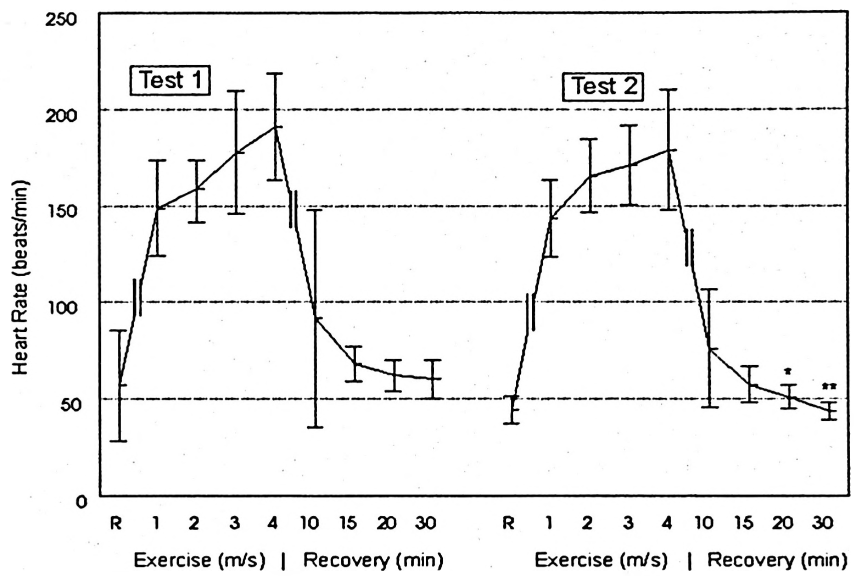

Fig. 1. *Glucose values (mean ± SD) for different exercise stages and for recovery period.* R: rest. Exercise stages: 1, 2, 3 and 4 for 4, 5.5, 7 and 8.5 m/s, respectively. Recovery: 10, 15, 20 and 30 min, respectively. *** $p < 0.001$ test 1 versus test 2.

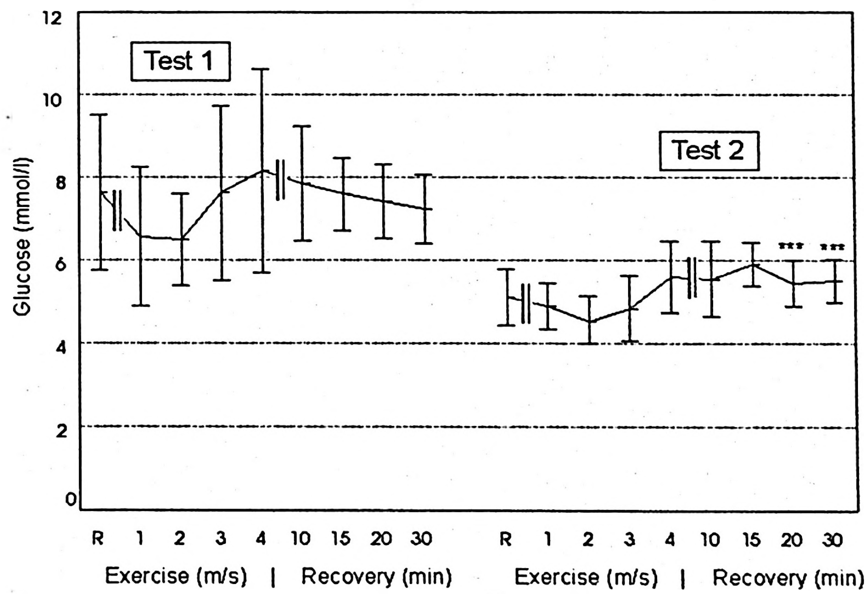

Fig. 2. *Heart Rate values (mean±sd) for different exercise stages and for recovery period.* Legend as in figure 1.

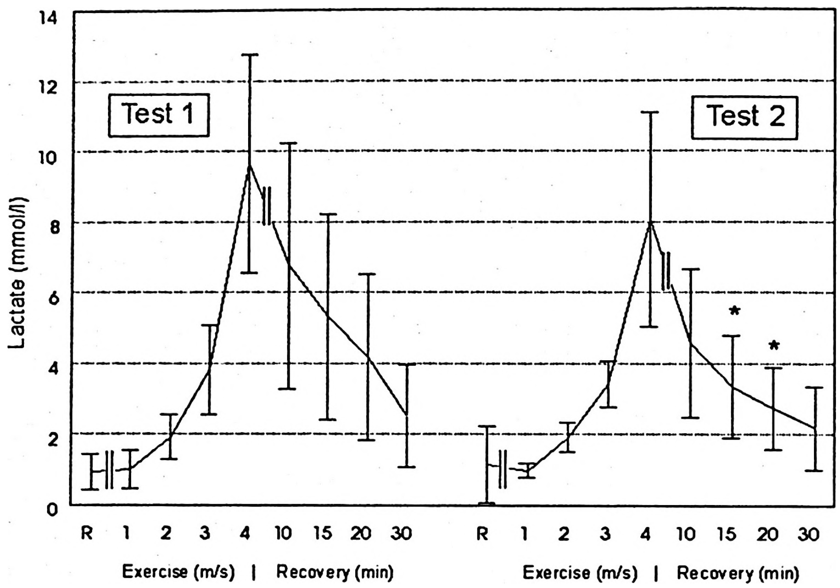

Fig. 3. *Lactate values (mean ± sd) for different exercise stages and for recovery period.* Legend as in figure 1.

The overall mean obtained for the first test was 4.01 mmol/l, compared with 3.16 mmol/l in test 2, a decrease of 0.85 mmol/l lactate after prolonged training.

Inter-test two-way variance analysis showed $F = 9.24$ and $p < 0.01$, while the values for inter-series analysis was $F = 35.38$, $p < 0.001$, the effects of training is shown at 15 and 20 minutes' recovery ($p < 0.05$).

Results obtained for electrolytes (sodium, potassium and chlorine), and creatinine are summarized in tables I and II.

Discussion

Glucose.– Glucose concentrations have been studied in relation to fitness, speed and effects of training (11, 24, 47). Resting values obtained for the Andalusian horses were higher than those reported for endurance-ride horses (28, 37).

With respect to normal glycemia levels in horses, a hyperglycemic effect was noted at rest in comparison to the first two stages of training; this may be attributable to training-related stress (23).

Plasma glucose concentrations rose after intense exercise, a finding reported elsewhere by KEENAN (22) and CASTEJÓN *et al.* (13), and associated with increased levels of plasma catecholamines and glucocorticoids (19), as well as with decreased insulinemia, glucose uptake being inhibited by muscle fiber (33, 34).

Heart Rate.– As ENGELHART's excellent review (15) indicates, the cardiovascular response to exercise in horses has been widely studied, and heart rate appears as a useful parameter in assessing the effects of strenuous speed-related effort.

Resting heart-rate values were perhaps slightly lower in the second test than in the first due to parasympathetic influence; similar results have been reported for comparisons of athletes and non-athletes (18).

KUBO (26) concluded from their research that considerable variations are to be found in maximal equine heart rate, with values ranging from 212/min to 239/min; peak values recorded here fall within these established normal limits.

Comparison of exercise v. recovery heart rate in both tests revealed that the decrease was more gradual in test 2 than in test 1. In test 2 (four months' training), resting values were resumed after 30 minutes' recovery, a finding reported elsewhere for horses (12) and sheep (31).

During all stages of rest, exercise and recovery, heart rate was lower in test 2 than in test 1, where the training schedule was shorter. It is well known that horses in good physical condition show a lower resting heart rate and a faster return to resting values after training (6).

Recovery of normal heart rate was found to take place in two stages: a rapid stage within ten minutes of finishing maximal exercise, and a slower stage from 10-15 minutes onwards. This two-stage recovery has been reported by RUBIO *et al.* (39). Recovery of initial heart rate values has been reported to improve with training, indicating the horse's increasing physical fitness (17). This was reflected in the present study in the observation that baseline heart-rate values were more rapidly recovered in test 2.

Lactate.– The extent of blood lactate accumulation during exercise is generally an indicator of physical fitness and degree of training (4, 32).

Attention has been drawn by some authors (30, 38) to the role of the glucose-alanine cycle in preventing lactate storage in the muscle; these authors report less accumulation during standardised exercise in well-trained than in untrained Trotters. In the present study, accumulation was lower in animals undergoing four months' training.

Lactate concentrations recorded after maximum-intensity exercise in both tests highlight the value of regular training for these animals, as a means of ensuring physical fitness (43).

During the recovery period, values were lower as a result of training. This would indicate greater aerobic capacity (2, 5).

CASTEJÓN *et al.* (12) report differences in heart rate and lactate concentrations between trained and untrained animals. Similar results were recorded here between horses undergoing 2 months' and four months' training. The effects of training are thus clearly reflected in the difference between recorded resting values and those noted following maximal training and during recovery (1).

In a study comparing three breeds (Andalusian, Arab and Anglo-Arab) undergoing the same exercise schedule as that used in the present study, CASTEJÓN *et al.* (14) reported higher lactate values than those observed here, during both exercise and recovery. A considerable decrease in lactate concentrations was noted here during recovery; the decrease was more marked in the second test, suggesting that training improved physical fitness.

Initial lactate levels at rest were slightly, though not significantly, higher in test 2 than in test 1. THORNTON *et al.* (46) report that resting lactate concentrations do not vary significantly as a function of training or physical fitness. Nevertheless, the results obtained following intense exercise were lower than those recorded for horses undergoing endurance exercises, such as saddle horses (41), suggesting that the training schedule employed for these Andalusian horses exerted a greater effect.

Electrolytes.– The changes observed in plasma electrolyte levels before, during and after training varied in significance, but in all cases fell within normal limits (8).

The resting values found for Na^+ and Cl^- in both tests were slightly lower than those indicated by some authors (8, 16) in horses of different breeds or those obtained by RUBIO *et al.* (40) in Andalusian horses. Conversely, the K^+ ion displayed higher values than those reported by the authors in the literature previously cited.

An increase with training in the mean resting values of Na^+ and Cl^- was noted, with a decrease in the K^+ ion.

A transitory rise in the K^+ ion with exercise occurred, coinciding with what was reported by most authors (9, 20, 29). This increase manifested itself more clearly after four months' training, the base levels being regained, in both tests, after finishing the exercise.

The significant rise noted for the K^+ ion concentration in plasma may be partly attributed to the extracellular increase which accompanied the acidemia and liberation of K^+ through the muscular fibres exercised concomitant with losses from sweating.

Creatinine.– Plasma creatinine concentrations increased after maximal exercise, although the increase was not significant; this indicates that phosphocreatine is not greatly involved in the obtaining of energy (36).

E. I. AGÜERA, M. D. RUBIO, R. VIVO, R. SANTISTEBAN, A. MUÑOZ y F. CASTEJÓN. *Respuesta de parámetros sanguíneos y frecuencia cardíaca en Caballos Andaluces sometidos a entrenamiento.* Rev. esp. Fisiol. (J. Physiol. Biochem.), 51 (2), 55-64, 1995.

Se estudia en Caballos Andaluces la respuesta de diversos parámetros sanguíneos y de la frecuencia cardíaca al entrenamiento. Se realizan dos pruebas, una a los dos meses de comenzar el período de entrenamiento y otra a los cuatro meses. Las pruebas realizadas son de intensidad creciente, sobre una distancia de 1.000 metros. Se aumenta paulatinamente la velocidad desde 4 hasta 8,5 m/s en cuatro escalones de ejercicio. En ambas pruebas se da un período de recuperación de 5 minutos entre cada nivel de ejercicio. La toma de muestras (mediante punción en la vena yugular externa) se efectúa en reposo, dentro del primer minuto de finalizado cada nivel de ejercicio y a los 10, 15, 20 y 30 min de recuperación. Para la realización de los análisis de las muestras se centrifugan inmediatamente para obtener el plasma donde se mide lactato, glucosa, iones y creatinina. La frecuencia cardíaca se controla mediante un monitor "Polar Sport Tester". Los parámetros más importantes en ambas pruebas son la glucosa, la frecuencia cardíaca y el lactato. Los electrólitos, la glucosa y la creatinina presentan una alteración pasajera ya que a los 30 minutos de recuperación se normalizan. El entrenamiento diario se refleja en el lactato y la frecuencia cardíaca ya que la recuperación mejora a medida que el entrenamiento aumenta, influyendo en su capacidad aeróbica y disminuyendo la acidosis metabólica.

Palabras clave: Bioquímica sanguínea, Caballos andaluces, Frecuencia cardíaca, Respuesta al entrenamiento, Lactato.

Acknowledgements

The authors wish to thank the authorities at Military Stud at Jerez de la Frontera (Cadiz, Spain), and particularly Lt. Col. Vinuesa, for their unstinting cooperation in this study. This study was financed by the C.I.C.Y.T. Research Project No DEP91-0465.

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