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Multivariate Analysis of Exercise Parameters Measured during an Incremental Treadmill Test

J.-P. VALETTE, E. BARREY and R. WOLTER

Laboratoire de physiologie sportive INRA. Ecole Nationale Vétérinaire d'Alfort, 94704 Maisons Alfort Cedex, France

ABSTRACT. The exercise fitness or poor performance of horses was investigated by measuring parameters of the cardiovascular, muscular, metabolic and locomotor systems. Fifteen Standardbred racehorses were tested on a high-speed treadmill. Heart rate, haematocrit, venous blood lactate, stride length and frequency were measured at each step of an incremental test consisting of 10 min warm up, then four 3 min steps of 1.5, 4, 7 and 9 m s^{-1}, with a 3% slope. Current exercise-related parameters which describe aerobic and anaerobic capacities as well as the adaptations of haematologic, cardiovascular and locomotor systems were defined using the relationships between these measurements and velocity. The results were analysed by a multivariate procedure, i.e. the principal component analysis. This statistical tool describes the relationships among parameters and compares the exercise adaptation of each horse by considering all the information obtained. It was possible to discriminate horses with a better aptitude to sprint from those with a poorer aptitude depending on one of the studied body systems. With increasing number of horses tested, this method becomes a useful aid in the diagnosis of poor performance.

Key words: Horses; principal component analysis; fitness; poor performance; exercise test.

INTRODUCTION

Exercise fitness depends on the integration of several body systems. Cardiovascular, respiratory, haematological, biochemical and muscular factors have been investigated independently of each other.[3,8,9,12,16] There have been studies of the relationships between physiological adaptation, energy metabolism and stride parameters.[10,11,13,15,17,18,21] It was demonstrated[7] that at each speed the horse adopts the most economical gait. Some studies[1,6] have examined the time relationship between the limb cycle and the respiratory cycle.

In forming a comprehensive picture of the interrelationships between the different body systems, classical statistical procedures were insufficient to synthesize all the information obtained during a standard exercise test. Therefore multivariate procedures such as principal component analysis (PCA) were used in this study to explain the complex behaviour of a system which is described by several parameters.

MATERIALS AND METHODS

Horses

Fifteen Standardbred horses, aged from 2 to 9 years, were tested. Eleven of them were normally trained for racing. All horses were sound and were tested on the treadmill for the first time without previous acclimatization.

Exercise test

The standardized test consisted of a 10 min warm up on the treadmill (Sato, Sweden) followed by four 3 min steps of 1.5 (walk), 4, 7 and 9 m s^{-1}, with a 3% slope. Animals were allowed to rest for 2 min between each step before the measurements.

Measurements

Heart rate. During the incremental test, the heart rate (HR) was recorded (Horse Tester PEH 200, Polar Electro) every 15 seconds and the data averaged at each of the 4 steps.

Blood analysis. Blood samples were taken by jugular venepuncture at rest, during the rest periods and after the recovery phase. A sample (0.5 ml) was immediately deproteinized by adding 1 ml of ice cold 0.6 N perchloric acid and lactate concentration was measured by an LM2 Analyser (Analox Instrument). The haematocrit (PCV) was measured by centrifugation in a vacuum tube (Monovette, Sarstedt).

Gait analysis. The stride frequency (SF) was determined at each step from the time interval required for 40 strides. Velocity (V) was measured by the treadmill electronic equipment. The stride length (SL) was then deduced from the equation: SL = V/SF.

Definition of the derived exercise parameters

For each horse, the synthetic parameters were calculated by means of regression equations. The relationships of HR, PCV and SF vs velocity were studied by linear regression. These relationships were entirely described by two characteristics, i.e. the slope and intercept.

The relationship between blood lactate (LA) and V was analysed using the exponential model[5] which was closely correlated to the experimental data:

$$LA = \exp(A \cdot V + B) + C$$

where A is a coefficient of curvilinearity and B and C the constants; LA is expressed in mmol l^{-1} and V in m s^{-1}.

For each horse, the values of A, B and C were found using a solver software (Eureka, Borland International) by considering the LA data at the first, third and fourth steps.

For each horse, 12 derived parameters were calculated and constituted quantitative variables (Table 1). The relationship between LA and V was described by the following five criteria: the intercept (LA_0), the coefficient of curvilinearity (A), the velocity which increased blood lactate to 2 mmol l^{-1} (V_{LA2}), the velocity at which the mean variation of blood lactate was 0.2 mmol l^{-1} for an increasing velocity of 1 m s^{-1}($V_{deltaLA}$) and the corresponding blood lactate threshold (LAT). The V_{LA2}, $V_{deltaLA}$ and LAT parameters were estimates of the onset of blood lactate accumulation (OBLA).[4,20] The relationship between HR and V was described by the slope (HR_s), velocity at 150 beats m^{-1} (V_{150}) and heart rate at V_{LA2}(HR_{LA2}). The relationship between SL and V was described by the slope (SL_s) and stride length at V_{LA2}(SL_2). The relationship between PCV and V was described by the intercept (PCV_0) and the slope (PCV_s).

Multivariate analysis

The data were studied by means of the (PCA) (Factor procedure, SAS STAT software). This statistical method describes the relationships between parameters and compares the exercise adaptation of each horse by considering all the quantitative exercise data obtained. This method was previously applied to equine biomechanical studies[14] and in exercise fitness evaluation.[2,19]

The PCA procedure consists of creating new variables called principal components, which result from a linear combination of the studied variables. The principal components (1,2,3, ..., N) were calculated by the software to maximize the variance of the observations in the multidimensional space. The first component explains the highest percentage of the variance and the next component numbers indicate their rank order. Analysis of the correlation between the studied characters and the individuals can be shown in a two-dimensional approximation (first two component axes) of the multidimensional plot. For further details on this statistical procedure, see the SAS STATE guide.

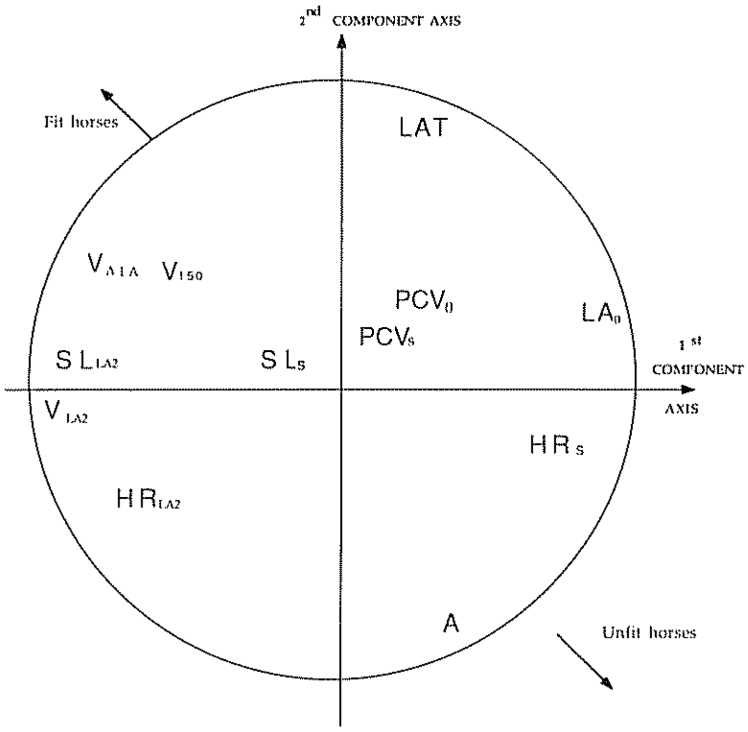

Fig 1 Diagram of the correlations between parameters on the first two component axes.

Table 1. *Derived exercise parameters obtained in horses*

	Variable												
Horse	A	LA_0	$V_{\Delta LA}$	V_{LA2}	LAT	HR_{LA2}	V_{150}	HR_s	SL_s	SL_{LA2}	PCV_s	PCV_0	AGE
1	34.1	0.25	7.10	8.00	0.85	171	6.70	15.8	50.4	4.72	2.60	22.2	3
2	16.3	0.40	7.40	7.85	1.65	205	5.00	19.3	35.9	3.98	1.90	31.2	3
3	17.1	0.40	8.10	8.65	1.55	189	6.20	16.0	40.9	4.50	1.90	28.1	5
4	16.1	1.10	7.15	6.55	2.40	162	5.80	16.1	40.1	3.71	2.30	23.7	3
5	8.9	0.30	10.55	9.80	2.45	202	6.55	15.8	41.1	4.89	1.25	44.1	5
6	21.5	0.70	6.15	6.60	1.60	188	4.80	21.1	42.6	3.90	1.95	39.4	5
7	62.4	1.35	4.05	4.35	1.65	140	4.80	22.2	35.9	2.80	0.95	33.0	2
8	16.1	1.00	8.30	7.95	2.20	171	6.60	15.6	36.0	4.21	1.95	38.0	4
9	17.8	1.00	6.70	6.55	2.10	181	5.05	20.3	42.7	3.81	2.25	41.3	6
10	15.4	0.95	7.05	6.70	2.20	157	6.30	18.5	43.2	3.85	2.25	37.5	6
11	12.1	1.30	5.35	3.40	2.95	106	5.80	18.1	43.8	2.32	1.95	40.4	9
12	16.3	1.50	5.35	3.85	2.75	114	5.75	19.1	38.7	2.71	3.00	34.7	8
13	27.3	0.50	6.85	7.55	1.25	170	6.40	17.5	35.4	3.94	1.85	26.8	7
14	41.7	0.40	4.05	4.85	0.90	145	5.15	15.9	39.8	2.83	1.95	30.3	5
15	23.8	0.30	6.60	7.15	1.45	173	5.80	17.1	44.1	4.23	1.15	31.5	5
Means	23.1	0.80	6.70	6.65	1.85	165	5.80	17.9	40.7	3.76	1.95	33.5	–
SD	13.3	0.40	1.60	1.75	0.60	28	0.65	2.0	3.9	0.74	0.50	6.4	–
(Units)		$mmol\ l^{-1}$	$m\ s^{-1}$	$m\ s^{-1}$	$mmol\ l^{-1}$	$beats\ m^{-1}$	$m\ s^{-1}$			m		p. 100	

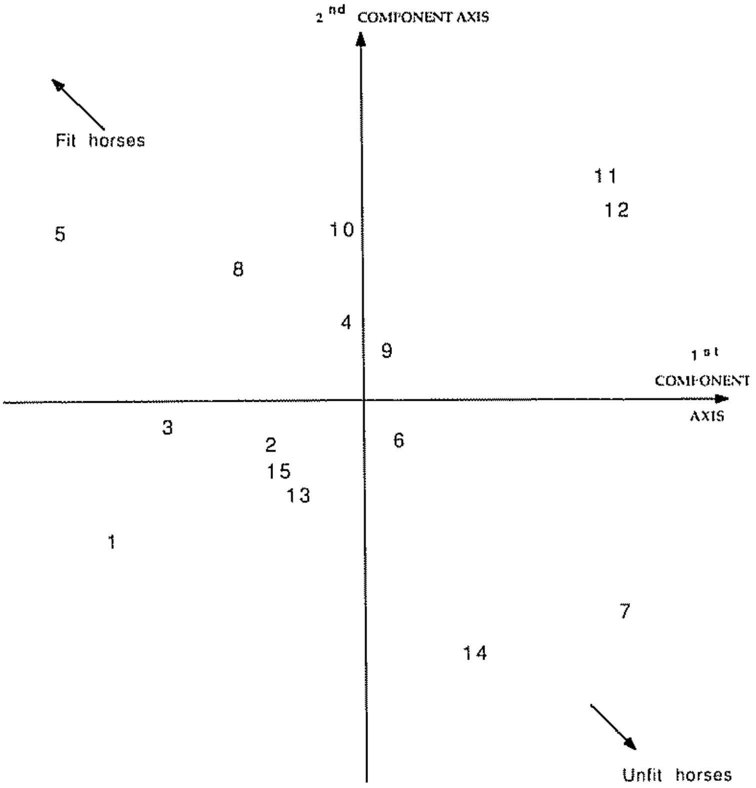

Fig 2 Comparison between the exercise ability of the horses on the first two component axes.

RESULTS

The PCA shows (Fig. 1) the relationships between the exercise parameters. The first and second component axes explained 41.8 and 21.0%, respectively, of the total variation and hence, the first two component axes represented 62.8% of the total variance.

The first component axis included the most significant parameters, V_{LA2}, $V_{deltaLA}$, LA_0, SL_{LA2} describing the energy metabolism and V_{150}, HR_s, HR_{LA2} the cardiac response. The second component axis distinguished the horses according to the lactate threshold parameters, LAT and A. The third component axis explained 15.4% of the variance and included the haematocrit parameters, PCV_0 and PCV_s. The fourth component axis explained 8.2% of the variance (SL_s).

There was a good correlation between V_{LA2} and $V_{deltaLA}$ ($r=0.89$), which gave the same type of information. V_{LA2} was correlated with HR_{LA2} ($r=0.90$) and SL_{LA2} ($r=0.97$). $V_{deltaLA}$ was correlated with HR_{LA2} ($r=0.70$) and SL_{LA2} ($r=0.84$). There was a poor correlation between PCV_0, PCV_s plus SL_s, and the other parameters. The lactate threshold parameters, LAT and A, were negatively correlated ($r=-0.59$).

Plotting the parameters relative to the first two component axes shows the relationships between the variables and principal components (Fig. 1). Part of the diagram corresponds to the good exercise ability, with higher values for LAT, V_{LA2}, $V_{deltaLA}$, V_{150}, SL_{LA2} and HR_2 and the other part to the poor exercise ability, with higher values for HR_s and A. Near the circumference, the closer the parameters the more correlated they are. For example, V_{LA2} and SL_{LA2} are positively correlated, while A and LAT are negatively correlated. The parameters situated close to the centre of Fig. 1, such as SL_s, PCV_0, PCV_s, are not clearly explained by the 1st and 2nd components.

Table 2. *Coordinates of horses on the principal component axes*

Horses	Axis 1	Axis 2	Axis 3	Axis 4	Axis 5
1	−2.59	−1.59	−3.09	1.32	0.44
2	−0.97	−0.86	1.59	0.10	−1.69
3	−2.56	−0.38	−0.37	−0.34	−0.31
4	−0.03	0.78	−1.20	−0.74	−0.95
5	−3.89	1.74	1.86	−0.37	1.23
6	0.49	−0.59	1.56	1.90	−0.44
7	4.12	−2.60	1.74	−0.92	0.80
8	−1.36	1.52	0.34	−1.58	−0.06
9	0.67	0.73	1.21	1.67	−0.51
10	−0.06	1.37	−0.33	0.55	0.26
11	3.29	2.40	−0.73	0.16	1.35
12	3.35	2.18	−1.30	−0.14	−1.06
13	−1.01	−1.19	−0.31	−1.47	−0.63
14	1.58	−2.48	−1.18	−0.32	0.31
15	−1.01	−1.03	0.21	0.18	1.26

Plotting the horses relative to the first two components (Fig. 2, Table 2) gave an overall view of their exercise ability. Away from the origin, the nearer the horses to one another the more similar their traits. Horses situated close to the centre of this diagram are not clearly explained by the 1st and 2nd components. The fit horses are situated in the top left-hand corner of the diagram, while the unfit horses are located in the bottom right-hand corner. Horses 3, 5, 8, and 10 were better than the others. Horses 7 and 14 exhibited poor exercise abilities. The two inactive older horses (11 and 12) showed the same response to the exercising test and were limited by their aerobic capacity. The other horses were located between these two groups.

DISCUSSION

Multivariate analysis takes into account all information available in a table of quantitative data and aims at synthesizing the results in a diagram. The procedure reveals the most efficient factors to be used to distinguish between individuals.

The coefficient of curvilinearity A, negatively correlated with LAT, indicated whether the shape of the lactate-velocity curve was regular and moderate (low value of A and high value of LAT) or dramatically near the threshold (high value of A and low value of LAT).

The fit horse (i.e. Horses 3, 5, 8, 10), should have low A, HR_s and LA_0 values and high values for the other parameters. On the other hand, Horses 7 and 14 had poor exercise abilities. Horse 7 was young and in its pre-training phase. Horse 14 was untrained and its aerobic capacity was not developed. This horse also had a short stride length and a high stride frequency. Horses 1, 4 and 15 were not sufficiently trained. The exercise ability of horses 2, 6 and 9 was not clearly explained by the body systems examined. Horses 11 and 12 were old Standardbred trotters which had raced in the past. They were inactive and limited by the cardiovascular response to exercise. Their lactate threshold was high even if the V_{deltaLA} was low. Horse 13 exhibited a good physiological adaptation to exercise, but did not increase its stride length.

According to the present results and previous studies,[2,4] the principal component is a valuable tool for comparing the exercise ability of horses in a standardized test. The introduction of additional reference values obtained in top level horses might increase the accuracy of this analysis. With increasing number of horses tested, this method could become a useful aid in the detection of exercise abilities and poor performance.

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