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Research article

BEHAVIORAL AND PHYSIOLOGICAL REACTIVITY OF MARES AND STALLIONS EVALUATED IN PERFORMANCE TESTS

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In many countries completing the performance test requirements is obligatory only for stallions, but some breeders also decide for the assessment of their mares' performance under standardized conditions. This study is aimed the evaluation of sex related effects on behavioral and physiological reactivity in fearfulness test and performance test scores in Polish warmblood horses of Małopolska breed (22 mares and 34 stallions) assessed at the training station. Equine reactivity to potentially frightening stimuli was assessed in the fearfulness test by behavior scoring and heart rate monitoring. Horses of both sexes were assessed in standardized performance tests accordingly to the rules of performance tests given in the Breeding Program for the Małopolski Horse Breed by the Polish Horse Breeders Association. The mares showed a significantly more quiet response in the fearfulness test in comparison to stallions. A relation among some reactivity and performance traits in both sexes was found. The behavior scores of the fearfulness test were positively correlated with character and temperament only in mares. However, the better the scores for temperament and character the stallions received, the lower the heart rate before testing. The present study demonstrates that temperament and character assessment, as a part of the standardized performance test, should involve not only a subjective trainer's evaluation, but also a horse reactivity assessment based on objective behavioral tests and heart rate monitoring. The results showed that sex has an effect on behavioral reactivity of horses. Thus, it is important to consider the horses' sex during selection for a particular type of riding, and performance assessment should be obligatory for both sexes. The assessment of the same performance traits in both, stallions and mares, significantly improves good breeding practice.

Key words: behavior, fearfulness test, heart rate, horse, performance test, sex

INTRODUCTION

The present breeding of warmblood horses and their selection for usage in leisure and top sport disciplines is based on performance evaluation, including the scores of standardized performance tests, as well as results in breeding shows and sport competitions [1]. Selection of stallions involves the estimation of genetic parameters and evaluation of performance traits accordingly to the selection criteria [2].

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Completing the performance test requirements is obligatory only for stallions, but some breeders also decide for the assessment of their mares' performance under standardized conditions. The majority of evaluated traits in performance tests involve conformation and gait quality characteristics, as well as behavior-related traits such as character, temperament, trainability and rideability. Increasing requirements of top sport performance horses concern not only their jumping and dressage predispositions but also the horse's appropriate interactions with the handler, rider and trainer. Some researchers involved in equine behavior studies underline the great importance of behavior-related traits as a selection criteria in riding horse breeding programs [3-7]. The assessment of animal reactivity provides a more comprehensive measurement using a combination of both behavioural and physiological indicators [8,9].

The aim of the study was to evaluate the effect of sex on behavioral and physiological reactivity in the fearfulness test and scores of performance test in horses assessed at the training station.

MATERIAL AND METHODS

Horses

The study included three and four year old Polish warmblood horses of Małopolska breed: 22 mares and 34 stallions. The studied mares and stallions participated in performance test programs at one of the Polish training stations. The schedule of training period and principles of final performance tests for mares and stallions were in agreement with the breeding program for the Małopolski horse breed administered by the Polish Horse Breeders Association (PHBA) [10].

Ethics

In our study non-invasive methods were used as approved by the Ethics Cmmittee and according to the National Law on the Care and Use of Animals.

Assessment of behavioral reactivity

Behavioral reactivity of mares and stallions was assessed with the fearfulness test [11] at the end of their training period (before the final performance test). The fearfulness test was conducted at the station's riding arena. The test consists of three sessions: (I) testing the horse's response to moving visual stimulus i.e. 2 rotating (40 rotations/min) black-white squares (1m x 1m), (II) testing the horse's response to auditory stimulus (metronome sound of 80 tones/min generated with a frequency of 440 Hz and intensity of 90 dB) and not moving visual stimulus (previously described squares but not rotating), and (III) testing the horse's response to moving visual stimulus (previously described rotating squares) and auditory stimulus (previously described metronome sound). Detailed characteristics of the fearfulness test have been described

previously [9,11,12]. Locomotor horse behavior during every session of the test was assessed by one observer using a 10-point decreasing reactivity scale as follows:

1-2 points = fearful horse, jumping and/or retreating, willing to escape and does not continue passing by the rotating squares despite encouragement;

3-4 points = skittish horse, jumping and/or retreating, continues passing after several trials of encouragement;

5-6 points = shy horse, stopping and/or retreating, continues passing with hesitation;

7-8 points = watchful horse, slowing and/or walking sideways and/or looking at rotating squares, continues passing with interest towards the objects;

9-10 points = fearless horse, no change in pace, no interest towards the objects, passing without hesitation.

Every horse was scored twice at each of the three test sessions (p1 – first point scoring the response to the stimuli, p2 – second point scoring the response to the stimuli). First time the horse was led next to the devices generating the stimuli hidden by the boards and after reversing by a stableman, the horse again was led next to the devices generating the stimuli, this time not hidden by the boards. As a total behavior score of each session (behavior score – session I, behavior score – session II, behavior score – session III) the sum of two scorings (points p1+p2) was considered.

Heart rate estimation

Prior to the fearfulness test the heart rate (HR) monitor and receiver (Polar 810i, OY ELECTRO, Finland) were attached to the horse while still in the stall. For each horse, HR was telemetrically recorded in 5 sec intervals during the testing period. The HR values at the riding arena before testing (HR before testing), in the first test session (HR I), in the second test session (HR II) and in the third test session (HR III) were estimated.

Performance data

The final performance test data compared between mares and stallions included the traits assessed by the chair of the training station such as: character, temperament, trainability, the traits assessed by the PHBA (Polish Horse Breeders Association) jury: free-jumping, walk, trot, canter and final index, as well as rideability assessed by a test rider. The performance traits were rated on a 10-point scale, from 1 point = very bad to 10 points = excellent. The final index was estimated by a 1 to 5 rating scale (from insufficient to perfect) based on the total point number calculated for all evaluated performance traits accordingly to the rules of performance test assessment given in the Breeding Program for Małopolski Horse Breed by the Polish Horse Breeders Association (PHBA) [10].

Statistical analysis

Results were presented as arithmetic means (X) and standard deviations (SD). U Mann-Whitney and Wilcoxon tests were applied and Spearman correlations were calculated (STATISTICA 6.0). In the present study, the differences between means and correlations were considered as statistically significant when $P<0.01$ or $P<0.05$ and tending to be significant when $P<0.1$. This is in agreement with previously published studies [9, 12, 13, 14, 15] where the correlations tending to significance ($P<0.1$) were also considered to reveal trends (tendencies) between some behavioral and physiological traits.

RESULTS

The results of behavioral and physiological reactivity of warmblood stallions and mares assessed in the fearfulness test are shown in Table 1. The behavior scores in response to moving visual stimulus (session I) and in response to auditory stimulus and not moving visual stimulus (session II) were significantly higher in mares than in stallions. Thus, it can be stated that sex influences the level of fear reactivity estimated upon horse locomotor behavior in the fearfulness test. The mares showed a more quiet response compared to stallions. However, there was no significant difference in behavior scores between mares and stallions in fearfulness test session III that could be due to the possibility of habituation to potentially frightening stimuli. The behavior scores in horses of both sexes revealed significant differences among particular test sessions (I, II, III) at $P<0.01$ in stallions and at $P<0.05$ in mares. The lowest behavior score was established in horses of both sexes taking the first test session and their reactivity was decreasing during the 2nd and the 3rd session. Heart rate monitoring in horses of both sexes revealed significant differences among HR before testing and HR during particular test sessions (I, II, III) at $P<0.01$ or $P<0.05$ in stallions and at $P<0.01$ in mares (Table 1). We found no significant differences in HR values between stallions and mares. However, significantly negative correlations were observed for HR among particular fearfulness test sessions (I, II, III) and behavior scores in relevant sessions for stallions: $-0.449 \leq R_S \leq -0.590$ at $P<0.001$ or $P<0.01$ and in mares: $-0.514 \leq R_S \leq -0.590$ at $P<0.01$.

Mean values of performance traits of warmblood stallions and mares assessed in the performance test are shown in Table 2. The quality scores for all gaits (walk, trot, and canter) were significantly higher in stallions than in mares. For temperament score we found a trend to significance (at $P=0.09$) showing that stallions' temperament tended to be better evaluated in comparison to the mares. However, the final index revealed a tendency for better evaluation (a trend to significance at $P=0.08$) in mares compared to stallions and it was influenced by slightly more desirable scores of character, free-jumping and rideability.

Table 1. Behavior scores (points) and HR values (beats/min) in mares and stallions assessed in the fearfulness test
Within columns means bearing the same letters differ significantly: A – I at $P \leq 0.01$, a – c at $P \leq 0.05$.
Within rows means bearing the asterisk differ significantly ** at $p = 0.01$, * at $p = 0.05$.

Fearfulness test indicators and HR values	Mares n = 22		Stallions n = 34	
	X	SD	X	SD
Behavior score - session I	17.50 ^{ab} **	2.70	15.24 ^{AB} **	3.24
Behavior score - session II	18.09 ^{a} *	2.09	17.12 ^{A} *	1.79
Behavior score - session III	18.10 ^{b}	2.09	17.24 ^{B}	2.28
HR before testing	54.50 ^{CDE}	19.27	46.85 ^{FGH}	14.28
HR I	84.50 ^{C}	21.28	81.35 ^{Fc}	22.52
HR II	82.45 ^{D}	21.32	76.74 ^{GIc}	18.21
HR III	86.18 ^{E}	21.97	85.44 ^{HI}	26.76

Table 2. Performance test scores in mares and stallions
Within rows means bearing the same letters differ significantly: C at $P \leq 0.01$, a – b at $P \leq 0.05$.
Within rows means bearing the asterisk show trend to significance: ** $p = 0.08$, * $p = 0.09$.

Performance traits	Mares n = 22		Stallions n = 34	
	X	SD	X	SD
Character	7.82	1.05	7.62	1.54
Temperament	7.07*	0.99	7.44*	0.99
Trainability	6.20	0.85	6.47	1.05
Free-jumping	6.88	0.73	6.85	0.66
Walk	6.46 ^{a}	1.61	7.11 ^{a}	0.55
Trot	6.26 ^{b}	1.58	6.99 ^{b}	0.66
Canter	6.08 ^{C}	1.55	7.06 ^{C}	0.60
Rideability	5.39	1.95	5.29	1.35
Final index	3.77**	0.43	3.53**	0.96

The results showed a relation among reactivity and performance traits in both sexes (Table 3 and 4). The behavior scores in sessions I and II positively correlated with character and temperament only in mares (Table 3). The more quiet response to fear stimuli was revealed, the higher scores for behavior-related traits in the performance test were obtained. This relation was not found in stallions. However, positive correlations between HR during the fearfulness test session III and free-jumping score as well as between HR during fearfulness test session II and final index were found only in mares (Table 3). There were no significant relations among behavior scores in fearfulness test and behavior-related traits evaluated in performance test in stallions. However, we found some significant (at $P<0.01$ or $P<0.05$) negative correlations between HR values before fearfulness testing and some performance traits in stallions

Table 3. Correlation coefficients among fearfulness test behavior scores (points) and HR values (beats/min) and performance tests scores in mares

Performance traits	Fearfulness test behavior scores			HR values			
	Behavior score – session I	Behavior score – session II	Behavior score – session III	HR before testing	HR I	HR II	HR III
Character	0.473 x	0.468 x	0.296	0.151	0.055	0.313	-0.002
Temperament	0.529 x	0.568 x	0.359	0.010	-0.112	0.095	-0.166
Trainability	0.208	0.293	0.007	0.056	-0.237	0.009	-0.133
Free-jumping	-0.260	-0.269	-0.304	0.015	0.229	0.329	0.488 x
Walk	0.076	0.108	-0.145	0.129	0.077	0.222	0.146
Trot	-0.021	-0.025	-0.184	0.189	0.249	0.303	0.166
Canter	0.038	0.031	-0.249	0.142	0.143	0.216	0.018
Rideability	0.048	0.116	-0.037	0.161	0.112	0.101	-0.170
Final index	-0.018	0.091	-0.099	0.334	0.326	0.496 x	0.265

x significant at $P \leq 0.05$

Table 4. Correlation coefficients among fearfulness test behavior scores (points) and HR values (beats/min) and performance tests scores in stallions

Performance traits	Fearfulness test behavior scores			HR values			
	Behavior score – session I	Behavior score – session II	Behavior score – session III	HR before testing	HR I	HR II	HR III
Character	-0.071	0.031	-0.058	-0.379 x	-0.100	-0.277	-0.263
Temperament	0.171	0.139	0.172	-0.412 xx	-0.081	0.041	-0.083
Trainability	0.112	0.151	0.124	-0.222	-0.149	-0.099	-0.022
Free-jumping	0.214	0.215	0.141	-0.125	-0.041	-0.030	-0.050
Walk	0.120	0.201	0.236	0.020	-0.026	-0.176	-0.216
Trot	0.232	0.294 *	0.340 x	-0.020	-0.196	-0.198	-0.356 x
Canter	0.265	0.333 x	0.393 x	-0.061	-0.170	-0.151	-0.254
Rideability	-0.137	-0.149	-0.102	-0.132	0.125	0.237	0.184
Final index	0.127	0.177	0.178	-0.338 x	-0.239	-0.099	-0.201

x significant at $P \leq 0.05$, xx significant at $P \leq 0.01$
* trend to significance at $p = 0.09$

(Table 4). The better scores for temperament and character stallions received, the lower HR before testing was recorded. HR during session III also negatively correlated with the score for trot quality. There was a significantly negative correlation between HR before testing in stallions and their final index. The behavior scores of session II and III were positively correlated with some gait quality characteristics (trot, canter) only in stallions (Table 4).

DISCUSSION

The majority of warmblood breeds horses are used for riding, from leisure to top sport disciplines. Physical and mental predispositions of riding horses are important not only to select an adequate individual for a particular sport discipline or type of riding but also to select an animal that is easy and safe to handle and use. Performance tests are used as a tool of early selection of potentially most suitable horses for breeding and/or elite sport competition. The study on Swedish warmblood riding horses demonstrated significant relations between performance test scores and later competition results in dressage and show jumping [16].

In the majority of European countries a standardized performance testing at the training station is obligatory for stallions and it plays a significant role in the selection for breeding. The performance test is obligatory for stallions predominantly due to the higher number of offspring in comparison to the mares. However, breeders and scientists consider that testing of mares could improve genetic practice and ensure breeding success.

Although the scores in performance tests can be modified by several factors, Olsson et al. [17] emphasize that the level of heritability of some performance traits is high. This is another reason for testing horses of both sexes that could result in a higher probability of great value progeny. Sex-related differences are interesting also in terms of predisposition for a particular type of use. These predispositions should involve not only typical 'utility' traits but also behavior-related ones. Adaptability to environmental stressors and physical exercise can be measured by both, behavioral and physiological parameters [9,18]. Leary and Knapp [19] suggest that male sexual hormones significantly influence the endocrine system and this kind of relation can modify behavioral patterns. In the present study, Małopolska breed mares showed a significantly more quiet response in the fearfulness test in comparison to stallions. An earlier study on Holstein horses also demonstrated a more quiet reaction to potentially stressful stimuli in mares and geldings compared to stallions [20]. However, studies of Sapula et al. [20] and Duberstein and Gilkeson [21] showed that mares were characterized by less desirable behavior than geldings. These findings also confirm that behavioral responses are controlled by the endocrine system and sexual hormones, as well as melatonin and leptin which in both sexes may play an important regulating function [20-22].

A combination of behavioral and physiological measures ensured that behavior scores were validated by heart rate assessment. In the present study behavior scores and physiological measures were related during testing of fear reactivity in both sexes. We found no significant differences in HR between stallions and mares; however significant correlating behavioral measures with physiological measures confirm that these measures can be used as objective tools in equine reactivity assessment. Noble et al. [23] established that heart rate was higher in mares compared to geldings during measuring their reactivity to sudden stimuli. However, Seaman et al. [24] suggest that the reactivity level in horses can be more influenced by age than by sex.

In our study we found that in mares behavior scores in the fearfulness test were positively related to temperament and character evaluated in the performance test. Le Scolan et al. [25] and Momozawa et al. [26,27] suggest that temperament assessment can be carried out by both methods, questionnaire survey of persons familiar with the horse and behavioral tests. In stallions behavior scores were positively related with gait quality characteristics and this could be due to easy horse adaptation to rapidly changing environmental stimuli. The easier the adaptation of stallions to frightening stimuli, the better quality of trot and canter they perform. Our results showed also decreasing behavioral reactivity within fearfulness test sessions II and III that could be due to habituation. This finding could be the result of habituation to potentially frightening stimuli which is one of the adaptation mechanisms developed in herbivorous animals classified as potential preys. Horses tend to react with avoidance or flight to potential danger, however, with repeated exposure, they can become accustomed to their surroundings and cease to avoid nonthreatening stimuli [28].

CONCLUSIONS

The present study demonstrates that temperament and character assessment as a part of standardized performance test should involve not only the subjective trainer's evaluation but also the horse reactivity assessment based on objective behavioral tests and heart rate monitoring. Our results showed that sex affects behavioral reactivity of horses. Thus, it is important to consider the sex during selection for a particular type of riding and performance assessment should be obligatory for both sexes. The assessment of the same performance traits in both, stallions and mares, significantly improves the breeding progress.

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PROCENA FIZIOLOŠKIH PARAMETARA I REAKTIVNOSTI KOBILA I PASTUVA NA OSNOVU PERFORMANS TESTA

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U mnogim zemljama obavezan je performans test samo za pastuve ali se neki uzgajivači odlučuju da obave i procenu performansi kobila u standardizovanim uslovima. Ispitivanje je vršeno radi procene uticaja pola na ponašanje i reakciju na strah kao i na rezultate performans testa kod poljskih punokrvnjaka (22 kobile i 34 pastuva). Ispitivanja su obavljena na lokaciji gde konji treniraju. Procenjivane su reakcije konja na potencijalnu opasnost, a rezultati testa ponašanja i praćenja srčanog rada prikazani su putem skora. Životinje oba pola su procenjivane na osnovu performans testa, standarizovanog prema kriterijumima Asocijacije odgajivača poljskih konja. Kobile su značajno mirnije reagovale u testu reakcije na opasnost (strah) u poređenju sa pastuvima. Uočena je povezanost pojedinih reakcija kod oba pola. Pozitivna korelacija skora ponašanja i temperamenta u testu opasnosti (strah), uočena je samo kod kobila. Međutim, bolji skorovi za temperament i karakter kod pastuva su uočeni kod životinja sa nižim vrednostima pulsa. Ova studija prikazuje da procena temperamenta i karaktera, kao deo standardizovanog performans testa, treba da obuhvata ne samo subjektivnu procenu trenera nego i objektivnu, testom procenjenu reaktivnost konja, zasnovanu na monitoringu ponašanja i srčanog rada. Na osnovu dobijenih rezultata može se zaključiti da pol ima uticaja na reaktivnost konja. Otuda je važno da se u obzir

uzme i pol životinje prilikom odabira za određene tipove jahanja i da performans test bude obavezan za oba pola. Procena istih performans karakteristika za oba pola - pastuve i kobile, značajno poboljšava dobru praksu u uzgoju konja.