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EXERCISE-INDUCED CHANGES IN LIPID AND PROTEIN OXIDATION IN THE BLOOD OF MARES AND STALLIONS OF ENGLISH HALF-BREED HORSES LIVING IN THE POMERANIAN REGION (NORTHERN POLAND)

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Physical exertion and exercise-induced stress in horses vary according to the equestrian discipline and breed. The correct training programme aims to adapt the horse's organism to the physiological changes that occur during intense physical exertion. The aim of our research was to investigate the effect of physical training on the levels of markers of oxidative stress [2thiobarbituric acid reactive substances (TBARS), carbonyl groups of oxidatively modified proteins, total antioxidant capacity (TAC)] in the peripheral blood of healthy English half-breed horses. Nine healthy English half-breed horses (7 mares and 8 stallions) living in the village of Karlikowo, in the administrative district of GminaKrokowa, within the Puck district, Pomeranian Voivodeship, in northern Poland were used in this study. Blood was collected from the animals' jugular veins in the morning, 90 min after feeding, while the horses were in the stable and immediately after exercise. The training started at 10:00 a.m., lasted 1 hour and consisted of a cross-country ride at a walk (5 mins), a trot (15 mins), a walk (10 mins), a trot (10 mins), a walk (5 mins), a canter (5 mins) and a walk (10 mins). The results of our study showed that blood levels of TBARS, as a biomarker of lipid peroxidation, showed a non-significant change in both mares and stallions immediately after exercise compared to the resting period. Blood levels of aldehydic and ketonic derivatives of oxidatively modified proteins were reduced in both mares and stallions after exercise compared to pre-exercise levels. These reductions were statistically significant. In the blood of mares and stallions, TAC levels were increased after exercise compared to preexercise levels. This increase was not statistically significant. The results of our study showed that exercise did not induce oxidative stress in the blood of English half-breeds mares and stallions. These results would provide valuable information for understanding the adaptation of horses to exercise.

Keywords: English half-breed horses, exercise, blood, 2-thiobarbituric acid reactive substances (TBARS), carbonyl groups of oxidatively modified proteins, total antioxidant capacity (TAC).

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ЗМІНИ В ОКИСЛЕННІ ЛІПІДІВ ТА БІЛКІВ У КРОВІ КОБИЛ ТА ЖЕРЕБЦІВ АНГЛІЙСЬКИХ НАПІВКРОВНИХ КОНЕЙ СПРИЧИНЕНІ ФІЗИЧНИМ НАВАНТАЖЕННЯМ, ЯКІ ПРОЖИВАЮТЬ У ПОМОРСЬКОМУ ВОЄВОДСТВІ (ПІВНІЧНА ПОЛЬЩА)

Фізичне навантаження та стрес, спричинений фізичними вправами, у коней різняться залежно від дисципліни кінного спорту та породи. Правильна програма тренінгу спрямована на адаптацію організму коня до фізіологічних змін, що відбуваються під час

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інтенсивних фізичних навантажень. Метою нашого дослідження було дослідити вплив фізичного тренування на рівень маркерів оксидативного стресу Греактивні речовини, які взаємодіють з 2-тіобарбітуровою кислотою (TBARS), карбонільні групи оксидативно модифікованих білків, загальна антиоксидантна активність (ТАС)] у периферичній крові здорових англійських напівкровних коней. У цьому дослідженні було використано дев'ять здорових англійських коней-метисів (7 кобил і 8 жеребців) з села Карліково, адміністративного округу гміни Крокова, Пуцького повіту, Поморське воєводство (північ Польші). Кров відбирали з яремних вен тварин враниі, через 90 хв після годування, під час перебування коней у стайні та одразу після фізичного тренінгу. Тренування розпочалося о 10:00, тривало 1 годину і складалося з кросової їзди кроком (5 хв), риссю (15 хв), кроком (10 хв), риссю (10 хв), ходьби (5 хв), галопу (5 хв) і ходьби (10 хв). Результати нашого дослідження показали, що рівні TBARS в крові як біомаркера перекисного окиснення ліпідів як у кобил, так і у жеребців показали незначні зміни відразу після фізичного тренінгу порівняно з періодом відпочинку. Рівні альдегідних і кетонових похідних окиснювально модифікованих білків у крові були знижені як у кобил, так і у жеребців після тренування порівняно з рівнями до тренування. Ці зниження рівня альдегідних і кетонових похідних були статистично значущими. У крові кобил і жеребців рівень ТАС був підвищений після тренування порівняно з рівнями до тренування. Це збільшення не було статистично істотним. Результати нашого дослідження показали, що фізичний тренінг не викликає окиснювального стресу в крові кобил і жеребців англійських метисів. Ці результати нададуть цінну інформацію для розуміння адаптації коней до фізичних вправ.

Ключові слова: англійські метиси, фізичні навантаження, кров, реактивні речовини, які взаємодіють з 2-тіобарбітуровою кислотою (TBARS), карбонільні групи окиснювально модифікованих білків, загальна антиоксидантна активність (TAC).

INTRODUCTION

Equestrian sport is very popular, but the cost of maintaining a horse is high. Maintaining its performance and prolonging its sporting life is a priority for every rider. To achieve this, it is necessary to facilitate the horse's adaptation to the conditions of the training process as much as possible, to correct its physiological state appropriately and in time to avoid overtraining, to minimise psychological and physiological stress and thus provide the animal with the conditions for the full disclosure of its genetic potential [2, 17, 22]. Methods are now available to determine the degree of readiness of a horse's body to display maximum agility, endurance, good jumping ability and precise movement coordination. At the same time, the greatest practical interest is in such an assessment of the functional state of the horse's body as a basis for the fine adjustment and dosage of its training loads [8, 38]. The management of a horse's training cannot be limited to the consideration of individual functional indicators. The trainer's focus should be on the horse's body as a whole, as a natural unit of different physiological functions [25, 52].

The readiness of a sport horse for competition is traditionally determined by clinical and zootechnical indicators. Zootechnical characteristics are very important in the initial selection and early specialisation of a horse, but in the final stages of preparation for competition, indicators reflecting the level of functioning of functional systems and motor qualities become decisive [34]. With physical activity and exposure to other environmental factors, as well as with pathological changes in metabolism or after the use of pharmacological agents, the content of individual

biomarkers of oxidative stress changes significantly [49, 54]. Consequently, the results of a biochemical blood test can be used to characterise the health status of the horse, its level of fitness and the course of adaptation processes [13, 32, 33].

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It is known that all sufficiently strong influences on the body, including physical activity, cause a non-specific adaptive response (stress) [10, 14]. Its obligatory component is lipid peroxidation (LPO). They occur continuously and their intensity usually increases under the influence of stress. LPO products are necessary for many biochemical processes, but are toxic in large amounts [3]. To maintain homeostasis, LPO chain reactions are inhibited by the antioxidant system, which includes vitamin E, catalase, ceruloplasmin, glutathione and other substances [35, 53]. The functional reserves of this system are limited. A deficiency of antioxidants in the body leads to oxidative stress, resulting in increased lipid peroxidation and protein oxidation and the accumulation of their toxic products [47].

It is now recognised that oxidative damage to various macromolecules (nucleic acids, proteins, lipids), which form the structural basis of all living organisms, is the main manifestation of oxidative stress. The latter is understood as a metabolic and energetic disturbance, the accumulation of active damaging agents (free radicals, pro-oxidants, etc.) that cause damage to living organisms at various levels of their organisation (starting, first of all, from the molecular level), leading to the development of various pathological conditions [40, 43]. As proteins are present in all tissues and organs, their modification can be a reliable indicator of pathological processes at both local and systemic levels [7]. Since proteins perform specific functions (often with clearly visible and easily recorded manifestations), assessing the qualitative and quantitative aspects of oxidative modification of proteins has a number of advantages in the diagnosis of pathological conditions. It has been shown that oxidative modification of proteins (rather than lipids or nucleic acids) is one of the earliest and most reliable markers of their presence and even appearance in a wide range of pathologies of different aetiologies [11, 16, 26, 39]. Furthermore, it has been shown that oxidatively modified proteins can persist in living organisms for long periods of time (hours, days and even years (e.g. lipofuscin), whereas the primary intermediates of oxidative stress (free radicals, lipid peroxidation products) exist in the free state for much shorter periods of time (usually a few minutes, at most a few hours) [46, 48, 50]. This circumstance also allows us to consider the phenomenon of oxidatively modified proteins in living organisms as relatively stable diagnostic parameters of their structural and functional state, which is of great importance in clinical practice [31].

The aim of our research was to investigate the effect of physical training on the levels of markers of oxidative stress [2-thiobarbituric acid reactive substances (TBARS), carbonyl groups of oxidatively modified proteins, total antioxidant capacity (TAC)] in the peripheral blood of healthy English half-breed horses living in the Pomeranian Voivodeship, in northern Poland.

MATERIALS AND METHODOLOGY

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Horses. Nine healthy English half-breed horses (7 mares and 8 stallions) living in the village of Karlikowo, in the administrative district of GminaKrokowa, within the Puck district, Pomeranian Voivodeship, in northern Poland (Karlikowo village, Puck district, $54^{\circ}44'12''N 18^{\circ}09'00''E$), aged 7. 4 ± 0.8 years old (for males) and 7.1 ± 0.6 years old (for females) were used in this study. All horses were involved in recreational riding. Horses were housed in individual stalls with feed (hay and oats) provided twice daily, at 08:00 and 18:00, and water available *ad libitum*. All horses underwent a thorough clinical examination and haematological, biochemical and vital indices were within reference ranges. None of the mares were pregnant.

Blood samples. Blood was collected from the animals' jugular veins in the morning, 90 min after feeding, while the horses were in the stable (between 8.30 and 10 am) and immediately after exercise (between 11 am and 2 pm). Blood was stored in tubes containing K_3 -EDTA and sodium citrate (3.8%) and kept on ice until biochemical analysis.

Exercise test. The training started at 10:00 a.m., lasted 1 hour and consisted of a cross-country ride at a walk (5 mins), a trot (15 mins), a walk (10 mins), a trot (10 mins), a walk (5 mins), a canter (5 mins) and a walk (10 mins).

2-Thiobarbituric acid reactive substances (TBARS). Lipid peroxidation was determined in whole blood collected from horses before and after exercise using the method developed by Kamyshnikov (2004) [24]. The absorbance of each aliquot was measured at 540 nm and the level of lipid peroxidation was expressed as nanomoles of TBARS formed per mL using a molar extinction coefficient of $1.56 \cdot 10^5 \text{ M}^{-1} \cdot \text{cm}^{-1}$.

Carbonyl derivative content of the oxidative modification of proteins (OMP) assay. The rate of oxidative modification of proteins was estimated from the reaction of the resulting carbonyl derivatives of the amino acid reaction with 2,4dinitrophenylhydrazine (DNPH) as described by Levine and co-workers (1990) [30] and modified by Dubinina and co-workers (1995) [12]. DNPH was used to determine the carbonyl content of soluble and insoluble proteins. The carbonyl content was calculated from the absorbance at 370 nm and 430 nm and an absorption coefficient of 22,000 $M^{-1} \cdot cm^{-1}$. Carbonyl groups were determined spectrophotometrically from the difference in absorbance at 370 nm (aldehydic derivatives, OMP_{370}) and 430 nm (ketonic derivatives, OMP_{430}) and defined as 1 nmol per mL of blood.

Total antioxidant capacity (TAC) assay. Blood TAC levels were estimated by measuring TBARS levels after oxidation of Tween 80 according to Galaktionova and co-workers (1998) [19]. Plasma inhibits the Fe^{2+} /ascorbate-induced oxidation of Tween 80, resulting in a decrease in the TBARS level. The absorbance of the solution obtained was measured at 532 nm. The absorbance of the blank was defined as 100%. The content of TAC in the sample (%) was calculated from the absorbance of the blank.

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Statistical analysis.Results are expressed as mean \pm S.E.M. Statistical analysis was performed using the STATISTICA 13.3 package (TIBCO Software Inc., USA). All variables were tested for normal distribution using the Kolmogorov-Smirnov and Lilliefors tests (p > 0.05). The significance of differences between levels of haematological parameters (significance level, p < 0.05) was tested using the Mann-Whitney *U* test [55]. All statistical calculations were performed on separate data from each individual using STATISTICA 13.3 software (TIBCO Software Inc., USA).

RESULTS AND DISCUSSION

Levels of TBARS as a biomarker of lipid peroxidation, aldehydic and ketonic derivatives of oxidatively modified proteins and total antioxidant capacity (TAC) in blood collected from horses before and after exercise were assessed and are shown in Figures 1-3.

The most prominent and currently used assay as an index of lipid peroxidation products is the 2-thiobarbituric acid reactive substances (TBARS) assay. It is based on the reactivity of a final product of lipid peroxidation, malonic dialdehyde (MDA), with 2-thiobarbituric acid to form a red adduct [20]. Blood TBARS levels in mares showed a non-significant increase (by 1.7%, p > 0.05) immediately after exercise compared to the resting period. Blood TBARS levels in stallions showed a non-significant decrease of 3.7% (p > 0.05) after exercise. There were no significant differences in erythrocyte TBARS levels between the resting period and after exercise for either mares or stallions. Both at rest and after exercise, the TBARS level in stallion erythrocyte suspensions was significantly higher by 22.3% (p < 0.05) and 15.9% (p < 0.05) compared to mares (Fig. 1).



Fig.1. Levels of lipid peroxidation as determined by quantification of 2-thiobarbituric acid reactive substrate (TBARS) levels (nmol MDA·mL⁻¹) in the blood of mares and stallions at rest and after exercise.

Values are expressed as mean \pm S.E.M.

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The most common non-specific type of oxidative protein modification is the oxidation of amino acid residues with the formation of carbonyl groups. The formation of additional carbonyl groups as a result of oxidative modification is considered a reliable indicator of free radical modification [31]. Aldehydic and ketonic derivatives of oxidatively modified proteins in the blood of mares and stallions at rest and after exercise, are shown in Figure 2.





Values expressed as mean \pm S.E.M.

*– statistically significant differences between rest and post-exercise values (p < 0.05).

The effect of exercise on blood carbonyl levels in mares and stallions is shown in Figure 2. The levels of aldehydic derivatives of oxidatively modified proteins were reduced in the blood of mares after exercise $(19.33 \pm 0.96 \text{ nmol/mL})$ compared with the values obtained before exercise $(22.79 \pm 1.18 \text{ nmol/mL})$. These reductions were statistically significant (15.2%, p < 0.05). In the blood of the stallions, the levels of aldehydic derivatives of oxidatively modified proteins were reduced after exercise (19.45 ± 0.88 nmol/mL) compared with the values obtained before exercise (21.96 ± 1.12 nmol/mL). This reduction was statistically significant (11.4%, p < 0.05) (Figure 2).

The levels of ketonic derivatives of oxidatively modified proteins were reduced in the blood of mares after exercise (23.27 ± 0.84 nmol/mL) compared with the values obtained before exercise (26.84 ± 1.09 nmol/mL). These reductions were statistically significant (13.3%, p < 0.05). In the blood of the stallions, the levels of ketonic derivatives of oxidatively modified proteins were reduced after exercise (22.50 ± 1.13 nmol/mL) compared with the values obtained before exercise ($25.77 \pm$

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1.13nmol/mL). This reduction was statistically significant (12.7%, p < 0.05). The levels of ketonic derivatives of oxidatively modified proteins in the blood of stallions were 4% (p > 0.05) and 3.3% (p > 0.05) lower before and after exercise, respectively, compared with the values obtained for mares (Fig. 2).

It is well known that the total antioxidant capacity (TAC) includes enzymatic antioxidants such as superoxide dismutase, catalase, glutathione peroxidase, as well as some macromolecules (albumin, ceruloplasmin, urea, glutathione, ferritin, etc.), and its assessment may contain more information than a single review of its components [18]. Total antioxidant capacity (TAC) in the blood of mares and stallions at rest and after exercise is shown in Figure 3.



Fig. 3. Total antioxidant capacity (TAC) in the blood of mares and stallions at rest and after exercise. Values expressed as mean ± S.E.M.

Blood TAC levels of the mares were increased after exercise ($89.18 \pm 5.21\%$) compared to pre-exercise levels ($85.49 \pm 6.71\%$). This increase was not statistically significant (4.3%, p > 0.05). In the blood of the stallions, TAC levels were increased after exercise ($84.90 \pm 4.65\%$) compared to the values obtained before exercise ($78.56 \pm 5.25\%$). This increase was not statistically significant (8.1%, p > 0.05). Blood TAC levels in stallions were 8.1% (p > 0.05) and 4.8% (p > 0.05) lower than in mares before and after exercise, respectively (Fig. 3).

Horses are susceptible to oxidative stress, but increased antioxidant capacity may improve the management of exercise-induced oxidative stress. Many studies indicate the time-dependent nature of oxidative stress in relation to prolonged stressors such as exercise. For example, Ott and co-workers (2022) identified oxidative biomarkers in the blood plasma of exercising horses [41]. Stock-type horses were subjected to a standardised moderate-intensity exercise protocol 3 times

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per week for 8 weeks. The exercise protocol followed NRC guidelines and consisted of 30% walking, 55% trotting and 15% galloping, with a target heart rate (HR) of 90 BPM. Blood plasma was collected in weeks 1, 2, 7 and 8 immediately before and 0, 30, 60 and 90 min after exercise and analysed for TAC, TBARS, glutathione peroxidase activity (GPx) and superoxide dismutase activity (SOD). Data were analysed as repeated measures with wk, d, time and their interactions as fixed effects. The TAC on day 2 (0.40 mMTrolox) was 7.5% higher than on day 3. There were wk \times d \times time interactions for SOD, TBARS and GPx. TBARS remained at pre-exercise baseline (d^{-1} wk⁻¹; 2.7 µM malondialdehyde) for most sampling times within weeks 1, 7 and 8; however, TBARS increased by 0.24 to 0.41 µM on day 2 of week 2 post-exercise and remained similarly elevated on day 3 pre- and immediately post-exercise. GPx remained similar to baseline (172.6 µM/min) but increased by 48.18 to 83.4 μ M/min at most sampling times on days 1 and 2 of week 2. SOD remained at baseline (167.2 U/mL) until it increased by 11.28 to 15.61 U/mL at 30 minutes post-exercise on day 1, week 1 and at most sampling times on day 3, week 8. Amino acids with antioxidant properties such as Met, Tyr and Trp decreased dramatically from week 2 to week 8. Met and Tyr also decreased from -60 to 90 min, whereas there was no time effect on Trp concentration [41].

Regular physical activity has been reported to be the most effective nonpharmacological intervention to enhance endogenous antioxidant capacity and reduce oxidative stress-induced tissue damage as a result of adaptive responses [23, 29, 42]. The majority of studies have reported an increase in oxidative stress as evidenced by increased lipid peroxidation, protein oxidation and changes in glutathione redox status, although a few studies have reported null findings for each (lipid, protein, glutathione) [15]. Accumulating evidence suggests an association between oxidative stress and strenuous physical exercise [9, 27, 28] and the beneficial effects of chronic exercise training on physical condition [37].

BrkljačaBottegaroand co-workers (2018) determined the effects of endurance races on the oxidative and antioxidant status of horses by assessing changes in reactive oxygen metabolites (d-ROMs), malondialdehyde (MDA), biological antioxidant potential (BAP) and oxidative stress index (OSI) levels [6]. The study was carried out on 53 starters (28 individual horses) competing in endurance races of different distances (40 and 80 km) and difficulty levels (easy and difficult). Blood samples were collected before and after the races. Changes in all measured OS biomarkers suggest that prolonged aerobic exercise during endurance racing may contribute to the oxidant/antioxidant imbalance in horses, which is mainly characterised by a pronounced antioxidant response. Biological antioxidant potential was found to be the most reliable biomarker of OS in endurance horses in the present study [6].

An acute bout of high-intensity exercise in horses induces systemic inflammation and oxidative stress. MacNicol and co-workers (2018) provided a detailed description of the response of articular and systemic oxidative and inflammatory biomarkers following high-intensity, exhaustive exercise in horses

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[36]. A group of horses (Ex) underwent repeated bouts of high-intensity exercise at a target heart rate of 180 beats/min to voluntary exhaustion. Baseline plasma and synovial fluid (SF) samples were collected 24 h before exercise and then at 0.5, 1, 2, 4, 8 and 24 h after exercise. This time course was repeated in a group of untrained control (Co) horses. Plasma and SF samples were analysed for prostaglandin E_2 (PGE₂), nitric oxide (NO), total antioxidant status (TAS) and glycosaminoglycans (GAG). The Ex group had significantly higher plasma NO at 0.5, 1 and 2 h and higher plasma PGE₂ at 0.5 and 1 h compared to the Co group. SF PGE₂ and GAG were also higher in Ex horses at 8 h compared to Co. It is concluded that high-intensity exercise in horses results in a rapid increase in systemic oxidative and inflammation and cartilage turnover at 8 h post-exercise. Articular inflammation and cartilage turnover at 8 h post-exercise. Articular inflammation and cartilage turnover at 8 h post-exercise.

The marked increase in oxidative stress and antioxidant functions occurred simultaneously in the intensively exercised horses. Tsubone and co-workers (2013) elucidated changes in oxidative stress and antioxidant functions in treadmillexercised Thoroughbred horses (n = 5, 3 to 7 years old) using techniques to measure serum d-ROMs for oxidative stress and BAP for antioxidant markers [51]. The effect of nasogastric administration of hydrogenated water (HW) or placebo water prior to treadmill exercise on these parameters was also investigated. Each horse was subjected to maximum treadmill exercise, which exhausted the horses at an average speed of 13.2 ± 0.84 m/sec. Blood samples were taken 4 times, immediately before the intake of HW or placebo water, 30 min before the treadmill exercise, immediately before the exercise (pre-exercise), immediately after the exercise (postexercise) and 30 min after the exercise. In all horses, both d-ROMs and BAP values increased significantly in the post-exercise period. The increase in d-ROMs tended to be lower in the HW group compared to the placebo group in the pre-exercise period. The increase in BAP was significant at approximately 150% of pre-exercise values in both the HW and placebo groups. The BAP/d-ROM ratio was significantly increased at post-exercise in both treatment trials, while a significant increase was also observed at pre-exercise in the HW trial. BAP, d-ROM and the BAP/d-ROM ratio tended to decrease at 30 min post-exercise, except for BAP and BAP/d-ROMs in the placebo trial [51].

In our previous study, we also investigated the influence of moderate-intensity exercise on haematological and biochemical values and on the acid resistance of erythrocytes in mares and stallions of the Holstein breed [1]. A total of seventeen Holstein horses (seven mares and ten stallions aged 6 years) were used in this study. Blood samples were analysed for haematocrit (HCT), haemoglobin concentration (HGB), red blood cell (RBC) count, white blood cell (WBC) count, platelet count (PLT), leukogram, mean corpuscular haemoglobin concentration (MCHC), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), red cell distribution width (RDW) and platelet distribution width (PDW). Serum

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concentrations of aspartate aminotransferase (AST), alanine aminotransferase (ALT) and lactate dehydrogenase (LDH) and biomarkers of oxidative stress were analysed. Stallions showed a significant increase in leukocytes and granulocytes as well as in erythrocytes, haemoglobin and haematocrit after the exercise test. The mean corpuscular haemoglobin concentration before exercise was higher in stallions. At the same time, mares showed a significant decrease in platelet volume after the exercise test. Exercise induced a significant increase in aspartate aminotransferase activity in stallions. Exercised mares and stallions showed a decrease in lipid peroxidation after exercise. Exercise also caused an increase in oxidative stress. The resistance of erythrocytes in 0.1 M HCl was similar between females and males. No statistically significant differences were observed in the percentage of haemolysed erythrocytes before and after exercise [1].

Exhaustive or intense exercise causes increased production of ROS and increases oxidative stress in muscles and other organs, leading to cellular damage. Long-term, high-intensity exercise is a stimulus that transiently perturbs the pro-oxidant and antioxidant (i.e. redox) environment, often leading to oxidative stress and damage [44]. While the manifestation of exercise-induced oxidative stress may be important for exercise adaptation [5, 21], excessive production of reactive oxygen and nitrogen species (RONS) during prolonged exercise has been associated with skeletal muscle fatigue [4, 44]. Thus, exercise-induced RONS can be described by a bell-shaped (hormesis) curve in which there are two endpoints of physiological function [45].

The results of our study showed that exercise did not induce oxidative stress in the blood of mares and stallions of English half-breeds. Biomarkers of lipid peroxidation were not altered (Fig. 1). A decrease in the levels of aldehydic and ketonic derivatives of oxidatively modified proteins was accompanied by an increase in the levels of total antioxidant capacity in the blood after exercise (Figs 2 and 3).

CONCLUSIONS

The aim of the present study was to investigate the effect of exercise on the levels of markers of oxidative stress [2-thiobarbituric acid reactive substances (TBARS), carbonyl groups of oxidatively modified proteins, total antioxidant capacity (TAC)] in the peripheral blood of healthy English half-breed horses living in the Pomeranian Voivodeship, northern Poland. Blood TBARS levels as a biomarker of lipid peroxidation in both mares and stallions showed a non-significant change immediately after exercise compared to the resting period. Blood levels of aldehydic and ketonic derivatives of oxidatively modified proteins were reduced in both mares and stallions after exercise compared to pre-exercise levels. These reductions were statistically significant. In the blood of mares and stallions, TAC levels were increased after exercise compared to pre-exercise levels. This increase was not statistically significant. The results of our study showed that exercise did

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not induce oxidative stress in the blood of English half-breeds mares and stallions. These results would provide valuable information for understanding the adaptation of horses to exercise.

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