

Plasma ionized calcium and parathyroid hormone concentrations in horses after endurance rides

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Objective—To evaluate changes in plasma ionized calcium (Ca^{2+}) and parathyroid hormone (PTH) concentrations in horses competing in endurance rides.

Design—Longitudinal clinical study.

Animals—28 horses.

Procedure—Venous blood samples were obtained from horses before and after racing 80 km. Plasma pH and concentrations of Ca^{2+} , PTH, inorganic phosphorus, albumin, lactate, and magnesium were measured.

Results—Overall, a significant decrease in mean (\pm SD) plasma Ca^{2+} concentration (from 6.44 ± 0.42 to 5.64 ± 0.42 mg/dl) and a significant increase in plasma PTH concentration (from 49.9 ± 30.1 to 148.1 ± 183.0 pg/ml) were found after exercise. Exercise also resulted in significant increases in plasma inorganic phosphorus, albumin, and lactate concentrations. No changes in plasma magnesium concentration or pH were detected after exercise. Plasma PTH concentration was not increased after exercise in 8 horses; in these horses, plasma PTH concentration decreased from 58.2 ± 26.3 to 27.4 ± 22.4 pg/ml, although plasma Ca^{2+} concentration was also decreased.

Conclusions and Clinical Relevance—Plasma Ca^{2+} concentration was decreased after racing for 80 km, compared with values obtained before racing. In most horses, an increase in plasma PTH concentration that was commensurate with the decrease in plasma Ca^{2+} was detected; however, some horses had decreased plasma PTH concentrations. (*J Am Vet Med Assoc* 2001;219:488–490)

A decrease in plasma ionized calcium (Ca^{2+}) associated with physical exercise has been described in horses competing in the cross-country phase of a 3-day event¹ and in show jumpers.² Moreover, in show jumpers, an increase in plasma parathyroid hormone (PTH) concentration has been detected in association with hypocalcemia, and the increase in PTH was commensurate with the decrease in Ca^{2+} .²

Exercise-associated hypocalcemia has been associated with exertional rhabdomyolysis, synchronous diaphragmatic flutter, and fatigue in horses.³ These abnormalities are commonly found after endurance races. Thus, it may be hypothesized that sustained

hypocalcemia during a lengthy endurance race could be part of the pathogenesis of fatigue and exercise-associated myopathies. This hypothesis is supported by a report⁴ that horses with a significant reduction in blood Ca^{2+} could not finish a 163-km ride.⁴ In addition, horses are more predisposed to develop signs of hypocalcemia (muscle fasciculations, synchronous diaphragmatic flutter, or exhaustion) when they have total body depletion of water, sodium, chloride, and potassium.⁵ Thus, horses performing endurance exercise are likely to have clinically important hypocalcemia.

The cause of exercise-associated hypocalcemia is unclear. Results of previous studies indicate that the decrease in plasma Ca^{2+} concentration is not attributable to changes in the percentage of calcium ionization induced by modifications in blood pH or plasma albumin.² Changes in plasma lactate and phosphorus concentrations are also poorly correlated with changes in plasma Ca^{2+} concentration.^{1,2}

Any decrease in plasma Ca^{2+} concentration should result in a compensatory increase in plasma PTH concentration. In addition, inadequate secretion of PTH could also be responsible for the hypocalcemia observed after exercise in horses. The purpose of the study reported here was to determine the effect of endurance exercise on plasma Ca^{2+} and PTH concentrations in horses.

Materials and Methods

Horses—Twenty-eight Arabian or Arabian-cross horses competing in 2 endurance rides were studied. Sixteen horses participated in ride 1 and 12 horses in ride 2. The route (moderately rugged terrain) and weather conditions (15 to 27 C and 50 to 78% relative humidity) were similar in both rides. All horses were endurance-trained and completed the 80-km course at a mean speed of > 11 km/h.

Experimental design—A venous blood sample was obtained from each horse on the day before the ride (during the preride veterinary examination) and immediately after the horse completed 80 km. Mean \pm SD interval between the end of exercise and collection of the blood samples was 7 ± 5 minutes. All horses successfully passed a veterinary examination after the race.

Sample handling and measurements—Blood samples were collected into vacuum tubes containing lithium heparin and centrifuged immediately, and the plasma was removed and stored in ice. Plasma Ca^{2+} concentration and pH were measured within 1 hour after collection by use of selective electrodes.⁸ Subsequently, plasma was frozen and stored at -20 C. Plasma PTH concentration was measured by use of an immunoradiometric assay⁹ validated for measurement of equine PTH.⁶ Plasma magnesium, inorganic phosphorus,

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albumin, and lactate were measured by use of spectrophotometric techniques. Magnesium was quantified with a procedure based on the reaction of magnesium and calmagite.^c Inorganic phosphorus concentration was measured, using a procedure developed on the basis of reaction of inorganic phosphorus with ammonium molybdate in the presence of sulphuric acid.^d Albumin concentration was measured, using the bromocresol green method.^e Lactate concentration was quantified by use of an enzymatic technique developed on the basis of oxidation of lactate to pyruvate by lactate oxidase.^f

Statistical analysis—To evaluate whether data were normally distributed, a Kolmogorov-Smirnov test was carried out. All variables under study had a normal distribution. Paired *t*-tests were used to compare results obtained before and after exercise. The Spearman test was used to determine correlations between variables. Results are reported as mean \pm SD. Significance was set at $P < 0.05$.

Results

After the 80-km ride, plasma Ca^{2+} concentration (5.64 ± 0.42 mg/dl) was decreased ($P < 0.001$), compared with values obtained before exercise (6.44 ± 0.42 mg/dl). Plasma PTH concentration (148.1 ± 183.0 pg/ml) was increased ($P = 0.007$), compared with values obtained before exercise (49.9 ± 30.1 pg/ml). Individual analysis of the Ca^{2+} and PTH data revealed that although all horses had decreased plasma Ca^{2+} after exercise, PTH concentration only increased in 20 of the 28 horses; in the other 8 horses, a decrease in plasma PTH concentration was detected after the race. To further investigate this finding, data from horses in which PTH was increased after exercise (group 1) and data from horses in which PTH was decreased after exercise (group 2) were analyzed separately. The 8 horses included in group 2 were homogeneously distributed between the 2 rides, representing 5 of 16 horses competing in ride 1 and 3 of 12 horses competing in ride 2. No significant differences between groups 1 and 2 were found in plasma Ca^{2+} concentrations determined before exercise (6.44 ± 0.36 mg/dl and 6.44 ± 0.45 mg/dl, respectively) or plasma PTH concentrations determined before exercise (46.5 ± 31.7 and 58.2 ± 26.5 pg/ml, respectively). Moreover, no differences in any of the other variables were found between groups before or after exercise.

After exercise, in group 1, plasma Ca^{2+} was decreased (5.56 ± 0.36 mg/dl; $P < 0.001$), and PTH was increased (196.5 ± 197.1 pg/ml; $P = 0.002$). In group 2, plasma Ca^{2+} concentration was also decreased after exercise (5.84 ± 0.34 mg/dl; $P < 0.001$), but plasma PTH concentration was decreased (27.4 ± 22.6 pg/ml; $P = 0.009$). The difference between plasma Ca^{2+} concentrations in groups 1 and 2 after exercise was significant ($P = 0.04$).

Exercise resulted in significant increases in plasma inorganic phosphorus (2.98 ± 0.79 vs 5.61 ± 1.63 mg/dl; $P < 0.001$), albumin (3.38 ± 0.26 vs 3.90 ± 0.32 g/dl; $P < 0.001$) and lactate (7.56 ± 3.80 vs 21.98 ± 15.71 mg/dl; $P < 0.001$) concentrations. There was a modest and nonsignificant increase in plasma magnesium concentration after exercise from 2.36 ± 0.37 to 2.53 ± 0.52 mg/dl. The change in blood pH found after exercise was not significant (7.403 ± 0.026 vs 7.412 ± 0.032).

Correlation analysis revealed that Ca^{2+} and PTH concentrations were significantly correlated ($r = -0.384$; $P < 0.001$); however, no significant correlation was found between changes in Ca^{2+} concentration and changes in PTH concentration ($r = -0.278$; $P = 0.152$). Moreover, change in Ca^{2+} concentration was not correlated with changes in any of the other variables studied.

Discussion

A significant decrease in plasma Ca^{2+} concentration, compared with values obtained before exercise, was detected after the 80-km rides. Exercise-associated hypocalcemia has been reported in horses performing short-term exercise.^{1,2} The decrease in plasma Ca^{2+} concentration after the endurance rides was more pronounced than that reported after show-jumping² and similar to the hypocalcemia found after the cross-country phase of a 3-day event.¹ In several reports, changes in plasma calcium concentration after endurance rides are described. In most instances, total calcium has been studied and found to decrease⁷⁻¹¹ or remain unchanged.^{12,13} However, total calcium values should be evaluated with caution, because there is an increase in albumin concentration after endurance rides,^{9,10,12,13} which should result in an artificial increase in plasma total calcium concentration.¹⁴ Lindinger and Ecker⁴ reported a decrease in plasma Ca^{2+} concentration after an 80-km race that was of similar magnitude to the decrease in plasma Ca^{2+} concentration found in our study.⁴

The decrease in plasma Ca^{2+} concentration did not induce neuromuscular disturbances (muscle fasciculations or synchronous diaphragmatic flutter). This finding, which is not surprising because horses with experimentally induced decrease in Ca^{2+} concentrations that were more substantial did not develop clinical signs,² supports the concept that other water and electrolyte abnormalities are usually required, in conjunction with hypocalcemia, to cause neuromuscular irritability.¹⁵

The cause of exercise-associated hypocalcemia in horses is unknown. Several hypotheses have been proposed to explain this phenomenon, including the complexing of Ca^{2+} with organic and inorganic anions that accumulate in blood during exercise (eg, lactate and phosphorus) and changes in the percentage of calcium ionization as a consequence of modifications in blood pH and albumin concentration.¹ Lactate, inorganic phosphorus, pH, and albumin values after the endurance rides were similar to those reported in horses performing long-distance exercise.⁷⁻¹³ In our study, as in previous studies,^{1,2} no significant correlations were found between changes in Ca^{2+} and changes in lactate, inorganic phosphorus, pH, and albumin. Thus, other causes (such as loss of Ca^{2+} by perspiration or intracellular movement of Ca^{2+}) should be investigated as causes of exercise-associated hypocalcemia.

To our knowledge, plasma PTH concentrations had not been studied previously in horses competing in endurance rides. Plasma PTH concentrations have been reported to increase after prolonged exercise in humans.^{16,17} In horses, an increase in plasma PTH concentration has also been described after short-term

exercise.² In our study, an overall increase in plasma PTH concentration was detected after endurance exercise. However, it was noticed that plasma PTH values were decreased after exercise in some horses; therefore, for further analysis, horses were allocated in 2 groups according to their PTH response after exercise. In a previous study,² we compared the PTH response to exercise-associated decreases in plasma Ca²⁺ concentration with the PTH response to EDTA-induced decreases in plasma Ca²⁺ concentration and reported that the increase in PTH detected after show-jumping was commensurate with the decrease in Ca²⁺ concentration. The PTH response to decreased Ca²⁺ concentration (ie, the increase in PTH concentration) may be extrapolated from the PTH-Ca²⁺ curve, which is obtained by inducing low Ca²⁺ concentration (eg, by infusing EDTA) and measuring PTH concentration at different Ca²⁺ concentrations.^{2,6} In the study reported here, analysis of data from all 28 horses revealed a reduction in plasma Ca²⁺ concentration of 0.8 ± 0.4 mg/dl after exercise. By extrapolation of these data to the equine PTH-Ca²⁺ curve,^{2,6} it appears that a decrease in plasma Ca²⁺ of 0.8 mg/dl should result in maximal stimulation of the parathyroid glands with plasma PTH reaching concentrations of approximately 200 pg/ml. However, the PTH concentration measured after exercise was only 148.1 ± 183.0 pg/ml. Thus, the increase in plasma PTH concentration detected after exercise was less than expected.

In the 20 horses in which plasma PTH increased after exercise (group 1), mean decrease in plasma Ca²⁺ concentration was 0.88 ± 1.43 mg/dl. In these horses, postexercise PTH concentration was 196.5 ± 197.1 pg/ml, which was similar to the value predicted from the PTH-Ca²⁺ curve obtained after infusion of EDTA.^{2,6}

In the other 8 horses (group 2), Ca²⁺ and PTH concentrations were decreased after exercise. Although it is tempting to speculate that in group-2 horses there was an inadequate PTH response that was insufficient to counteract the decreased Ca²⁺ concentration, we do not have data to support this claim. Athletic performance was apparently similar in both groups: all group-2 horses completed the rides, and no differences in mean speed, finishing position, or body condition after the race were detected between groups. In addition, failure to secrete PTH would have resulted in lower plasma Ca²⁺ concentrations in group-2 horses. However, it is interesting that despite having decreased PTH values, the decrease in Ca²⁺ concentration was less pronounced in group 2 than in group 1. The PTH response in group-2 horses is contradictory and difficult to explain, because a decrease in plasma Ca²⁺ concentration should result in an increase in plasma PTH concentration. Although speculative, an explanation may be that these horses were recovering from more severely decreased Ca²⁺ concentrations at the time of blood sampling, because postexercise plasma Ca²⁺ concentration was significantly higher in group 2 than in group 1. If so, the lower PTH values detected in group-2 horses could be partially explained by the hysteresis (time lag) of the PTH-Ca²⁺ curve: for the same plasma

Ca²⁺ concentration, the plasma PTH concentration is lower during recovery from hypocalcemia than during the induction of hypocalcemia.¹⁸ Thus, if at the time of sampling group-2 horses were already recovering from decreased plasma Ca²⁺ concentration, the PTH values should be lower than those predicted from the PTH-Ca²⁺ curve.

^aCiba Corning 850, Madrid, Spain.

^bRat PTH, Nichols Diagnostics, San Juan Capistrano, Calif.

^cMagnesium procedure 595, Sigma Diagnostics, St Louis, Mo.

^dPhosphorus Inorganic procedure 360-3, Sigma Diagnostics, St Louis, Mo.

^eAlbumin BCG procedure 631-2, St Louis, Mo.

^fLactato Spinreact, Reactivos Spinreact, Olot, Spain.

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