

## Occurrence of gastric ulcers and serum gastrin levels in jumping horses

[*Ocorrência de úlceras gástricas e níveis séricos de gastrina em cavalos de hipismo*]

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### ABSTRACT

The aim of this study was to determine the occurrence of EGUS and to quantify serum gastrin levels in jumping horses during competition season and interseason period. Forty jumping horses, competing at high level were randomly allocated into two groups, the Training Group: twenty jumping horses undergoing intense training and participating in competitions, and the Rest Group: twenty jumping horses in the interseason (resting period). The gastroscopic examinations and blood samples of the horses in the training group were performed 1-2 days following the competition while in the horses of the rest group, following 4 weeks of rest. The serum gastrin levels were measured at two different times: pre-feeding and two hours after feeding the horses (postprandial) by ELISA kit. Gastric lesion score data were compared by the Mann-Whitney U test ( $\alpha=0.05$ ) and the mean gastrin values were compared between the groups and between the two moments by the paired t tests, respectively ( $\alpha=0.05$ ). Squamous gastric ulcers were detected in 42.5% of all jumping horses examined independent of the period, competition season or interseason. Serum gastrin levels were significantly higher in the Training Group with no difference between pre-feeding and postprandial values.

Keywords: horses, gastric ulcers, gastrin

### RESUMO

O objetivo deste estudo foi determinar a ocorrência de EGUS e quantificar os níveis séricos de gastrina em cavalos de hipismo durante a época de competições e o período de férias. Quarenta cavalos de hipismo de alta performance foram aleatoriamente distribuídos em dois grupos, grupo treinamento: vinte cavalos de hipismo submetidos a treinamento intenso e participando de competições, e grupo descanso: vinte cavalos de hipismo em férias (período de descanso). As avaliações gastroscópicas e as coletas de sangue dos cavalos em treinamento foram realizadas um ou dois dias após as competições, enquanto nos cavalos do grupo descanso foram realizadas após quatro semanas de repouso. Os níveis séricos de gastrina foram mensurados por kit de ELISA, em dois momentos: antes da alimentação e duas horas após. Os dados de escore das lesões gástricas foram comparados pela prova U de Mann-Whitney ( $\alpha=0,05$ ) e os valores médios de gastrina foram comparados entre os grupos e entre os dois momentos pelos testes t e t pareado, respectivamente ( $\alpha=0,05$ ). Foram encontradas úlceras gástricas em 42,5% de todos os cavalos examinados, independentemente do período de competições ou repouso. Os níveis séricos de gastrina foram significativamente maiores no grupo treinamento, sem diferença entre os períodos pré e pós-alimentação.

Palavras-chave: cavalos, úlceras gástricas, gastrina

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## INTRODUCTION

The presence of gastric ulcers in horses has been associated with clinical signs such as decreasing in appetite, rough hair coat, failure to thrive, intermittent mild colic and poor performance (Bruijn *et al.*, 2009). As suitable length endoscopes became available, a gastric ulceration was widely diagnosed, even in asymptomatic horses. The high prevalence of gastric lesions in athlete horses resulted in allowing the use of cimetidine, ranitidine and omeprazole for horses competing under FEI rules (Higgins, 2004). Esomeprazole magnesium has also been used in horses, demonstrating excellent results in the control of acid secretion and gastric pH (Pereira *et al.*, 2009).

Although the prevalence of equine gastric ulcer syndrome (EGUS) has been the focus of numerous studies (Niedźwiedź *et al.*, 2013; Tamzali *et al.*, 2011; Bruijn *et al.*, 2009; Hartmann and Frankeny, 2003); the pathogenesis and treatment (Sykes *et al.*, 2014) of this disease is very poorly studied in jumping horses, especially compared to racehorses. High performance jumping horses are exposed to numerous potential risk factors, including transportation, increased levels of exercise during a competition, changes in diet (Nadeau *et al.*, 2000), altered feeding schedules and housing (Murray and Eichorn, 1996), which makes them highly susceptible to the development of gastric ulcers.

An essential step to control or eliminate EGUS is to be able to identify when and why the lesions develop and progress (Hartmann and Frankeny, 2003). Furthermore, the knowledge of the normal physiology of the athlete horse stomach is particularly relevant to understanding the physiopathology of gastric diseases while little is known about the physiological mechanisms regulating acid secretion in the horse (Sandin *et al.*, 2000). Gastrin is the only hormone known to stimulate secretion of hydrochloric acid (Smyth *et al.*, 1989) and some studies have evaluated the effects of diet and feeding on serum gastrin concentration in adult horses (Smyth *et al.*, 1988), foals (Smyth *et al.*, 1989) and ponies (Baker and Gerring, 1993), but none has been performed on athletic horses.

This study aims at determining the occurrence of gastric ulcers, and serum gastrin levels in jumping horses during competitions and resting periods, hypothesizing that gastric ulceration prevalence and levels of serum gastrin were higher during the competition period.

## MATERIALS AND METHODS

This study has been approved by the Ethics Committee on Animal Use of the College of Veterinary Medicine and Animal Science, Universidade de São Paulo, Brazil (protocol number 3156/2013).

A total of 40 high performance jumping horses of different breeds (Hanoverian, Holsteiner, KWPN, Jumping Brazilian Horses), male and female, aged between 7 and 13 years and weighing between 500 and 600kg were used. The animals were randomly divided into two groups: Training Group and Rest Group. The training group consisted of twenty horses undergoing intense training and participating in a number of competitions while the rest group consisted of twenty jumping horses that had also undergone intense training and participated in competitions throughout the year, but were in the resting season for at least four weeks at the time of evaluation.

The gastroscopic evaluations of the horses of the Training Group were scheduled according to the timetable of the Brazilian Equestrian Confederation and were held during the year always on Mondays after major competitions. On the other hand, the horses of the Rest Group were evaluated at the end of January, when resting period was about to finish.

Before gastroscopic examination, the animals underwent an 18-hour feed fasting period and the last 6 hours of both water and feed fasting. Each animal was then restrained in the horse stock and injected intravenously 0.01mg/kg detomidine (Dormiun V, Agener União, Brazil). Five minutes later, a three-meter-long flexible endoscope was introduced into the nostril, through the nasopharynx, oesophagus, reaching the stomach of the animal. After carefully evaluating the obtained images, the lesions were classified according to Macallister *et al.* (1997) that assigns scores for the number and severity of ulcers.

### Occurrence of gastric ulcers...

The serum gastrin levels were determined for the horses of both Training and Rest Groups. Blood samples were collected at two different times, 6:00AM (immediately before the first feeding of the day, fasting horses) and 8:00AM (two hours after feeding) due to possible changes in gastrin levels as a result of feeding. Blood samples intended for laboratory analyses were collected in tubes without anticoagulant (BD Vacutainer SST II Advance® Gel Tube, Brazil) by jugular venipuncture using disposable needles (25x7mm needles, BD Vacutainer, Brazil) after site antisepsis. The bottles were centrifuged at 3000rpm for 5min in order to separate the serum, of which 700µl aliquots were stored in Eppendorf like tubes (Axygen, Brazil) and frozen at -80°C until processing. Serum gastrin levels were determined using Gastrin I (human) EIA kit (ADI-900-026, Enzo Life Sciences), following the manufacturer's instructions.

The nonparametric Mann Whitney U test ( $\alpha=0.05$ ) was used to compare the Training and Rest groups regarding the score for the number of ulcers found in the horses. The Kolgomorov-Smirnov test was used to check the normality of the data regarding gastrin levels. The Normal distribution ( $P>0.05$ ,  $\alpha=0.05$ ) of the data

allowed to use parametric tests. The t test for two samples was adopted to compare both groups ( $\alpha=0.05$ ), whereas the paired t test was used to compare the two periods of study ( $\alpha=0.05$ ). All analyses were conducted using the statistical software SPSS v. 15.

### RESULTS

The results show that 42.5% of the 40 jumping horses examined had gastric ulcers. Gastric ulcers were identified in eight horses of the Training Group (40%) and nine horses of the Rest Group (45%). The horses that were affected by ulcers received scores one (1 or 2 lesions), two (3 to 5 lesions) and three (6 to 10 lesions). In the Training Group, 50%, 33% and 25% of the animals scored 1, 2 and 3, respectively. In the Rest Group, 45%, 25% and 22% of the animals scored 1, 2 and 3, respectively. These results are shown in Figure 1.

The severity of 100% of the gastric ulcers found in the affected animals had score 1, which means a superficial ulcer with mucosa loss only. There was no significant difference in the gastroscopic evaluations between the horses of the Training and Rest Groups.

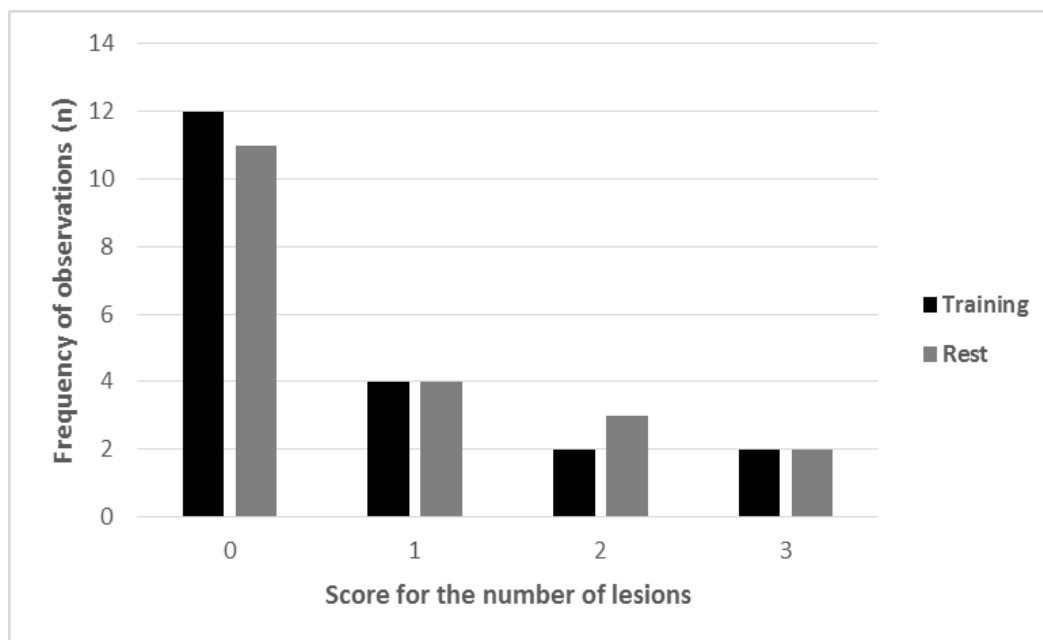


Figure 1. Graph showing the frequency of observations versus the score for the number of lesions according to the gastroscopic evaluation of 40 horses divided into two groups, Training and Rest. No significant difference was observed between groups ( $P=0.739$ ).

Serum gastrin levels were not significantly different between the two times, pre-feeding and postprandial for either of the two groups, Training ( $P= 0.108$ ) and Rest ( $P= 0.102$ ). However, gastrin levels of horses in the Training

Group were significantly higher than the levels found in the horses of the Rest Group for both times, pre-feeding ( $P= 0.032$ ) and postprandial ( $P= 0.001$ ) (Figure 2).

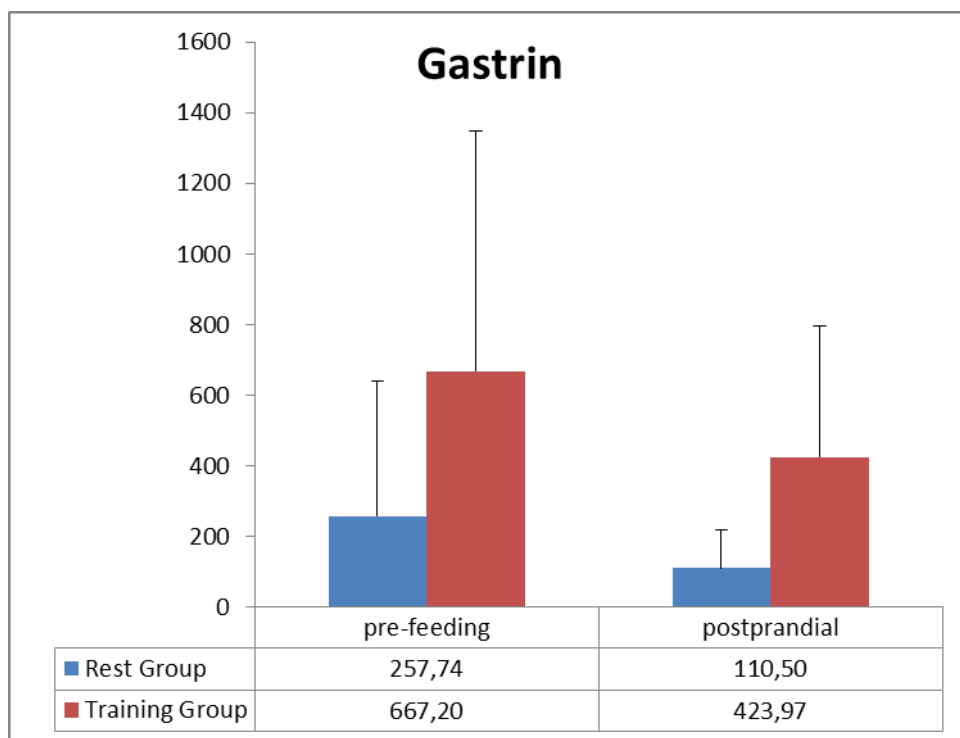


Figure 2. Graph showing the mean values of serum gastrin levels (pg/ml) obtained at two times (before and two hours after feeding) for forty horses divided equally into Training and rest Groups.

## DISCUSSION

Gastric ulcerations were present in a large number of the jumping horses (42.5%) examined. However, these results are relatively low compared to the prevalence reported for racehorses of 74.5% (Marqués *et al.*, 2011), 79.5% (Schaefer *et al.*, 2006), and even higher than 90% (Ferrucci *et al.*, 2003). EGUS prevalence of 67% has been reported for horses examined at the end of either a 50 or 80km ride (Nieto *et al.*, 2004) and 93% for a population of trained horses competing at a higher level (90-160km) (Tamzali *et al.*, 2011).

Researchers seeking to relate the occurrence of gastric lesions to the training practices of racehorses reported ulcers in 38% and 88% of horses at the beginning and in the fourth month of training, respectively. It was also observed that the occurrence of gastric lesions in non-

professional horses (56%) was much lower than that of intensively trained horses (93%). The prevalence of gastric lesions has also been determined in highly athletic endurance horses in two seasons: during the competition and interseason periods, similarly to this study. The authors reported a gastric ulcer incidence of 43% during the rest period, and 93% during the competition period (Tamzali *et al.*, 2011).

The incidence of gastric ulcer in jumping horses, in this study, is lower than that reported for horses of other sports. In addition, there was no difference in the occurrence of gastric ulcers among the horses of training (40%) and the rest (45%) groups. This result may suggest that a four-week-resting period is not enough to compensate for the effects of training and competitions as risk factors for the occurrence of gastric lesions and for the healing of existing injuries since spontaneous healing of stomach

ulcers in athletic horses is not common (Murray *et al.*, 1996).

On the other hand, one can hypothesize that although the stress factors during training and competitions were removed from the rest group, since physical exercise is one of the most physiologically stressful stimuli an animal can undergo (Gondin *et al.*, 2013), there were other stress factors that played a role. Situations like transport to the stables where the horses spent the rest period, or change of environment and management are known risk factors for gastric lesions.

Although the purpose of this work was not to study the effect of diet on gastrin release, serum levels of this hormone was assessed at two different times (before and after feeding) to compare Training and Rest Groups excluding other factors. It has already been reported that a pattern of gastrin secretion is related to how and for how long a particular diet is feed to the animals. Some studies evaluating gastrin release in response to food type showed contradictory results. Smyth *et al.* (1988) fed hay to horses and reported no gastrin release, but observed a significant increase when the horses were fed pellets. Sandin *et al.* (1998) observed greater gastrin release when horses were fed hay compared to grain. In this study, there was no statistical difference pre-feeding and postprandial for serum gastrin levels, although a decrease of 61% was observed in gastrin levels after feeding in the Rest Group and 37% in the Training Group. Probably, there was no statistical difference between the moments because the variability among the samples was very high.

Serum gastrin levels of the horses in the Training Group were higher than the levels found in the Rest Group, at both times pre- and post-feeding, indicating a possible influence of athletic activity on this hormone release. One possible explanation for the higher concentration of serum gastrin in the animals of the Training Group could be a delay in gastric emptying due to exercise. The animals exercise every day during periods of intense competitions, and the increased intra-abdominal pressure during intense exercise causes gastric compression (Lorenzo-Figueiras and Merritt, 2002) that could diminish the speed of gastric emptying. Some researchers have studied the effect of food on

serum gastrin concentration and attributed the delay in gastric emptying as a possible factor in greater gastrin release. The delay in gastric emptying would allow prolonged contact of the ingested meal with the receptors for gastrin release, which results in sustained high levels of serum gastrin (Smyth *et al.*, 1989).

## CONCLUSIONS

The findings of this study support the conclusion that jumping horses have higher gastrin blood levels during competition compared to rest period. Further research needs to be conducted to understand what triggers high gastrin release during intense physical activity and the clinical significance of these findings. Although there was no difference in the occurrence of gastric lesions between the two groups, it can be speculated whether higher blood levels of gastrin in athletic horses could be related to higher gastric acid secretion and consequently greater susceptibility to the development of gastric ulcer disease, but further studies need to be developed.

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