**Effects of total parenteral nutrition associated with glutamine, enteral fluid therapy with or without glutamine, and fluid therapy on the acid–base and electrolyte balance of horses starved after exploratory laparotomy**

Efeitos da nutrição parenteral total associada à glutamina, fluidoterapia enteral com ou sem glutamina, e fluidoterapia no equilíbrio ácido-base e eletrolítico de equinos sob inanição após laparotomia exploratória

**Abstract**

This study aimed to evaluate the effects of the total parenteral nutrition associated with glutamine, enteral fluid therapy with or without glutamine, and fluid therapy on the acid-base and electrolyte balance of horses starved after exploratory laparotomy. Sixteen healthy male and female adult horses of mixed breed, aged between 4 and 14 years, and having a mean body weight of 248.40 ± 2.28 kg and a body score index of 3–4 (scale of 1–5) were divided into four groups with four animals per group. After an adaptation period of 30 days, they were randomly divided into four experimental groups: enteral fluid therapy, enteral fluid therapy associated with glutamine, total parenteral nutrition associated with glutamine, and parenteral fluid therapy. The experiment was further divided into two phases: Phase 1 and Phase 2. In Phase 1, an exploratory laparotomy was performed, treatments were administered to the groups and the horses received no food or water other than those given to their respective groups. In Phase 2, the animals were re-fed. Each phase had a total duration of 144 h. Venous blood samples were collected every 24 h throughout the experimental period for blood gas and electrolyte analyses. The following parameters were evaluated: pH, partial pressure of carbon dioxide, total carbon dioxide, bicarbonate, base shift, anion gap, sodium, potassium, chloride, total calcium and magnesium. Completely randomized designs with a 4 × 7 factorial scheme (groups × harvest time) in Phase 1 and a 4 × 6 factorial scheme (groups × harvest time) in Phase 2 were used with four replications. All values were considered significant when p < 0.05 (95% probability). Blood pH, bicarbonate concentration, and base shift in the PARGL group decreased, indicating metabolic acidosis. Changes in the acid–base and electrolyte balance were more intense in the PARGL group than in the other groups. These results demonstrated the need to monitor blood gas and electrolyte balance in horses with food restriction under nutritional support or prolonged fluid therapy so that such changes are promptly corrected.

**Keywords:** equine, electrolyte imbalances, fluid therapy, metabolic acidosis, parenteral nutrition.
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Introduction

Decreased appetite and/or inability to eat commonly occur under different conditions, resulting in malnutrition, compromised immune function, and impaired tissue repair (Bailey et al., 2016; Carr, 2018; Di Filippo et al., 2021; Hospes & Bleul, 2007; Melo et al., 2008).

Nutritional support, enteral or parenteral, is a therapeutic resource that helps patients recover from and avoid the risk of malnutrition; it has gained importance for reducing hospitalization time, optimizing response to clinical-surgical treatments, promoting tissue repair, and improving immune function (Baek et al., 2020; Lawson et al., 2021).

Nutritional support should be considered for horses with increased metabolic rates, such as growing animals, individual horses with a history of malnutrition or hypophagia, patients with metabolic disorders, and those with diseases that result in an increased metabolic rate (Carr, 2018).

The use of parenteral nutrition in horses has been rarely studied although it has been described mainly in conference proceedings, individual case reports, retrospective studies, and review articles. It can be beneficial in ensuring adequate nutrient supply, particularly during critical illness or recovery from surgical procedures. However, many veterinarians reluctantly implement it because of not only the complexities of formulation, administration, and clinical monitoring but also the potential complications and expenses experienced by clients (McKenzie, 2015).

Therefore, this study aimed to evaluate the effects of total parenteral nutrition (TPN) associated with glutamine, enteral fluid therapy with or without glutamine, and fluid therapy on the acid-base and electrolyte balance of horses starved after exploratory laparotomy.

Materials and methods

Sixteen healthy male and female adult horses of mixed breed, aged between 4 and 14 years, with a mean body weight of 248.40 ± 2.28 kg and a body score index of 3 to 4, scale of 1–5 (Carroll & Huntington, 1988), were divided into four groups, and four horses were included in each group.

The horses that received endo- and ectoparasiticide were housed in paddocks and fed daily with commercial feed (1 kg/100 kg BW), Tifton hay (1 kg/100 kg BW), chopped elephant grass (Pennisetum purpureum), water, and mineral salt ad libitum. After an adaptation period of 30 days, they were randomly divided into four experimental groups (ENTFL, ENTGL, PARGL, and PARFL groups) in accordance with previously described methods (Melo et al., 2021).

For the ENTFL group, horses were administered electrolyte-infused enteral fluid therapy (57 g NaCl, 3.78 g NaHCO₃, 0.37 g KCl, and 10 g glucose/liter of water). For the ENTGL group, the horses were given enteral fluid therapy with electrolytes (5.7 g NaCl, 3.78 g NaHCO₃, 0.37 g KCl, and 10 g glucose/liter of water) associated with glutamine (L-glutamine, Ajinomoto do Brasil Indústria e Comércio de Alimentos Ltda, Laranjal Paulista/SP, Brazil).

A maintenance rate of 60 mL/kg was established to calculate the total volume of fluid to be administered to the ENTFL and ENTGL groups over a 24-hour period (Dias et al., 2019; Melo et al., 2010). The total calculated volume of fluid was divided by 12 and administered by gravity flow every 2 h using an 11 mm × 16 mm nasogastric tube. Glutamine (ENTGL) was administered at a
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dose of 0.5 g/kg body weight. The calculated total amount of glutamine was divided by 12 and administered in a diluted form in enteral fluid therapy every 2 h. The volume of fluid administered (ENTFL and ENTGL groups) and the amount of glutamine (ENTGL group) were adjusted daily according to the weight of the animal.

For the PARGL group, the horses received TPN associated with glutamine. TPN was prepared from equal amounts of solutions of amino acids (10% Aminoven, Fresenius Kabi, Barueri/SP, Brazil), lipids (20% Lipovenos, Fresenius Kabi), and 50% glucose (Fresenius Kabi). The proportions were 33.33% amino acids, 33.33% lipids, and 33.33% of 50% glucose to meet the energy requirements for daily maintenance. Glutamine was intravenously administered as a 1.5% sterile solution (15 g/L) at a dose of 0.5 g/kg of weight.

The total amount of TPN and the amount of glutamine to be administered were adjusted daily according to the animal's body weight. Both solutions were administered as continuous infusions via venous access into the right external jugular vein. The rate of administration (mL/h) was calculated by dividing the total infused volume by 24. The appropriate amount of intravenous fluid therapy with lactated Ringer's solution was administered to complete the daily maintenance requirement.

For the PARFL group, the horses were administered with parenteral fluid therapy. The volume of the fluid administered was 60 mL/kg BW. Half of it was supplied using lactated Ringer’s solution, and the other half was provided via a 0.9% sodium chloride saline solution. Then, 50% glucose was provided in a diluted form in a saline solution at a dose of 1.5 g/kg BW. Both solutions were administered as continuous infusions via venous access into the right external jugular vein.

Venous access for the infusion of TPN (PARGL group) and parenteral fluid therapy (PARFL group) was obtained using a double lumen catheter (Duocath - 12F, Intra Special Catheters, Germany) with a coaxial size of 12 gauge and a length of 20 cm.

This study was divided into two phases: Phase 1 and Phase 2. In Phase 1, treatments were administered depending on the groups, exploratory laparotomy was performed, and the horses were given no food or water other than those described in their groups. In Phase 2, the horses were re-fed. On the first 2 days of Phase 1, TPN administered to the PARGL group provided only 50% and 75% of the maintenance energy needs, respectively. On the third day to the end of Phase 1, TPN provided 100% of these needs. In Phase 2, the horses in the groups were re-fed with Tifton hay, commercial concentrate, and water. Feeding was gradually reintroduced. On the first day, 4 kg of Tifton hay was fed, divided into two 2 kg meals given 12 h apart, and added with 0.5 kg of concentrate. On the second day, the amount of hay was increased to 8 kg and provided in two meals of 4 kg each plus 1 kg of commercial concentrate. From the third day, the amount of food provided was the same as during the adaptation period. Each phase had a total duration of 144 h.

The animals underwent two flank laparotomies to simulate surgical stress and obtain intestinal samples used in a parallel study: one at the beginning and the other at the end of the first phase in accordance with a previously described protocol (Ferreira et al., 2022). Each laparotomy lasted an average of 60 min.

Venous blood samples were collected for blood gas analysis every 24 h during the experimental period (total of 13 samples). For this purpose, 1 mL of whole blood without bubbles and without an anticoagulant was collected from the left external jugular vein through a 3 mL syringe and analyzed immediately after collection.

Blood gas analysis was performed using portable automatic equipment (I-Stat system, Abott Laboratories, Brazil) with specific cartridges (I-Stat cartridge EC8+, Abbott Laboratories, Brazil). The following parameters were evaluated: pH, partial pressure of carbon dioxide (PCO$_2$), total carbon dioxide (TCO$_2$), bicarbonate (HCO$_3$), base shift, anion gap, sodium, potassium, and chloride.

Blood samples were obtained through a venipuncture in the external jugular vein and placed in a vacuum tube without an anticoagulant for measuring the concentration of total calcium (Ca$^{2+}$) and magnesium (Mg$^{2+}$). After clot retraction, blood was centrifuged at 3,000 rpm for 5 min, and serum was separated into 0.5 mL aliquots and frozen at −20 °C until analysis. Measurements were performed in a semi-automatic biochemical device (BA-88A biochemical analyzer, Química Básica Ltda, Belo Horizonte/MG, Brazil) by using specific kits (colorimetric kit for calcium and magnesium dosage, Química Básica Ltda, Belo Horizonte/MG, Brazil).
The experimental design was completely randomized with a $4 \times 7$ factorial scheme (groups $\times$ harvest time) in Phase 1 and a $4 \times 6$ factorial scheme (groups $\times$ harvest time) in Phase 2 with four replications. Data were tabulated in an Excel® spreadsheet, and their averages were compared using Duncan's test. All values were considered significant when $p \leq 0.05$ (95% probability). Data were statistically analyzed using SAS software (1997).

**Results**

The serum sodium concentration did not show significant group $\times$ time interaction ($p > 0.05$) and difference ($p > 0.05$) between the total number of groups in both phases and the total number of times in phase 2. However, a significant difference was observed ($p < 0.05$) in the total number of times in Phase 1 (Table 1). In the PARGL group, hyponatremia was observed between $T_9$ and $T_{10}$ and from $T_9$ until the end of the experimental period.

Chloride ions did not exhibit a significant interaction between the groups $\times$ times ($p > 0.05$) and differences ($p > 0.05$) between the total number of groups and the total number of times in both experimental phases (Table 2). Although the chloride value decreased in the ENTGL, PARFL, and PARGL groups, this decrease did not cause differences between the groups possibly because of the low number of animals used in each group.

Potassium concentration had no significant interaction between groups $\times$ times ($p > 0.05$); however, the total times in both phases and the total number of groups in Phase 1 differed significantly ($p < 0.05$; Table 3). The results demonstrated that serum potassium values had a more pronounced decrease in the PARGL group compared to those of the other groups in Phase 1 although the mean values remained within the reference limits for the species. In Phase 2, the potassium concentration increased.

**Table 1.** Mean ± standard error of serum sodium concentration (mEq/L) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times     | Experimental Groups | Total         |
|-----------|---------------------|---------------|
|           | ENTFL | ENTGL | PARFL | PARGL | T1 | T2 | T3 | T4 | T5 | T6 | T7 | T8 | T9 | T10 | T11 | Total |
| Phase 1 - Starvation |       |       |       |       | 136.25 ± 1.43 | 132.75 ± 1.25 | 133.25 ± 0.81 | 133.25 ± 1.54 | 132.25 ± 0.62 | 133.25 ± 0.75 | 133.39 ± 0.43 |       |       |       |       | 136.81 ± 0.57a |       |
|           | ENTFL | ENTGL | PARFL | PARGL | 135.25 ± 0.94 | 134.50 ± 1.25 | 132.25 ± 2.32 | 133.50 ± 2.12 | 131.00 ± 1.68 | 132.66 ± 1.45 | 133.25 ± 0.63 |       |       |       |       | 134.12 ± 0.49b |       |
|           | ENTFL | ENTGL | PARFL | PARGL | 138.25 ± 0.85 | 134.50 ± 1.75 | 134.50 ± 2.66 | 133.50 ± 2.12 | 134.00 ± 1.68 | 133.50 ± 2.53 | 134.50 ± 0.75 |       |       |       |       | 131.75 ± 1.65b |       |
|           | ENTFL | ENTGL | PARFL | PARGL | 137.50 ± 1.04 | 134.50 ± 1.84 | 131.75 ± 2.68 | 132.75 ± 2.56 | 130.25 ± 1.79 | 133.75 ± 0.47 | 133.17 ± 0.70 |       |       |       |       | 133.12 ± 0.77b |       |
|           | ENTFL | ENTGL | PARFL | PARGL | 138.25 ± 0.85 | 134.50 ± 1.75 | 134.50 ± 2.66 | 133.50 ± 2.12 | 134.00 ± 1.68 | 133.50 ± 2.53 | 134.50 ± 0.75 |       |       |       |       | 132.87 ± 1.04b |       |
|           | ENTFL | ENTGL | PARFL | PARGL | 137.50 ± 1.04 | 134.50 ± 1.84 | 131.75 ± 2.68 | 132.75 ± 2.56 | 130.25 ± 1.79 | 133.75 ± 0.47 | 133.17 ± 0.70 |       |       |       |       | 133.12 ± 0.77b |       |
|           | ENTFL | ENTGL | PARFL | PARGL | 138.25 ± 0.85 | 134.50 ± 1.75 | 134.50 ± 2.66 | 133.50 ± 2.12 | 134.00 ± 1.68 | 133.50 ± 2.53 | 134.50 ± 0.75 |       |       |       |       | 132.43 ± 0.98b |       |
|           | ENTFL | ENTGL | PARFL | PARGL | 137.50 ± 1.04 | 134.50 ± 1.84 | 131.75 ± 2.68 | 132.75 ± 2.56 | 130.25 ± 1.79 | 133.75 ± 0.47 | 133.17 ± 0.70 |       |       |       |       | 132.37 ± 0.81b |       |
|           | ENTFL | ENTGL | PARFL | PARGL | 138.25 ± 0.85 | 134.50 ± 1.75 | 134.50 ± 2.66 | 133.50 ± 2.12 | 134.00 ± 1.68 | 133.50 ± 2.53 | 134.50 ± 0.75 |       |       |       |       | 133.33 ± 0.69b |       |

Reference value: 132.00 – 146.00 mEq/L. Source: Carlson (2006). Averages followed by different lowercase letters in the column differ ($p<0.05$ – Duncan’s test).
**Table 2.** Mean ± standard error of serum chloride concentration (mEq/L) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total |
|-------|---------------------|-------|
|       | ENTFL | ENTGL | PARFL | PARGL |       |
| Phase 1 - Starvation |       |
| T₁ | 100.50 ± 1.44 | 100.50 ± 1.04 | 102.25 ± 0.85 | 100.75 ± 2.05 | 101.00 ± 0.66 |
| T₂ | 99.50 ± 1.44 | 101.00 ± 1.63 | 101.50 ± 0.95 | 100.00 ± 1.77 | 100.50 ± 0.69 |
| T₃ | 100.75 ± 0.85 | 98.75 ± 1.31 | 100.75 ± 2.65 | 99.75 ± 2.01 | 100.00 ± 0.85 |
| T₄ | 100.50 ± 0.28 | 97.00 ± 1.73 | 98.50 ± 2.78 | 96.75 ± 2.28 | 98.18 ± 0.97 |
| T₅ | 99.50 ± 1.55 | 96.50 ± 2.10 | 97.50 ± 3.01 | 99.00 ± 4.14 | 98.12 ± 1.32 |
| T₆ | 99.50 ± 1.84 | 93.00 ± 3.24 | 98.25 ± 2.17 | 99.00 ± 2.48 | 97.43 ± 1.30 |
| T₇ | 99.75 ± 2.09 | 96.33 ± 1.85 | 98.75 ± 2.65 | 100.50 ± 0.95 | 99.00 ± 0.98 |
| Total | 100.00 ± 0.50 | 97.62 ± 0.82 | 99.64 ± 0.83 | 99.39 ± 0.84 |       |

Reference value: 99.00 – 109.00 mEq/L. Source: Carlson (2006).

**Table 3.** Mean ± standard error of serum potassium concentration (mg/dL) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total |
|-------|---------------------|-------|
|       | ENTFL | ENTGL | PARFL | PARGL |       |
| Phase 1 - Starvation |       |
| T₁ | 3.90 ± 0.04 | 3.85 ± 0.21 | 3.95 ± 0.22 | 3.65 ± 0.16 | 3.83 ± 0.08abs |
| T₂ | 3.65 ± 0.15 | 3.70 ± 0.19 | 3.80 ± 0.26 | 3.47 ± 0.12 | 3.65 ± 0.09abc |
| T₃ | 3.52 ± 0.14 | 3.35 ± 0.19 | 3.30 ± 0.17 | 3.22 ± 0.17 | 3.35 ± 0.08abc |
| T₄ | 3.35 ± 0.06 | 3.22 ± 0.16 | 3.17 ± 0.11 | 2.67 ± 0.06 | 3.10 ± 0.08ab |
| T₅ | 3.37 ± 0.14 | 3.00 ± 0.30 | 3.27 ± 0.13 | 2.90 ± 0.17 | 3.13 ± 0.10abc |
| T₆ | 3.57 ± 0.25 | 2.92 ± 0.31 | 3.22 ± 0.11 | 2.85 ± 0.21 | 3.14 ± 0.12ab |
| T₇ | 3.45 ± 0.32 | 3.26 ± 0.12 | 3.27 ± 0.09 | 2.82 ± 0.23 | 3.20 ± 0.11ab |
| Total | 3.54 ± 0.07ab | 3.33 ± 0.09ab | 3.42 ± 0.07ab | 3.08 ± 0.08abc |

Reference value: 2.4 – 4.7 mEq/L. Source: Carlson (2006). Averages followed by different uppercase letters in the row and lowercase letters in the column differ (p < 0.05 – Duncan’s test).
Magnesium concentration did not have significant interactions between groups × times and differences (p > 0.05) between total times and total number of groups in both phases (Table 4). Although this concentration decreased, it did not cause differences between the times throughout the experimental period.

The total calcium concentration (Table 5) showed no significant interaction between groups × times (p > 0.05) and differences (p > 0.05) between the total times in both phases and the total number of groups in Phase 2. However, it had a significant difference (p < 0.05) in the total number of groups in Phase 1.

Blood pH did not have a significant interaction between the time groups × time (p > 0.05) and any difference between the total number of times and the groups in Phase 2. In Phase 1, the total number of groups and the total number of times significantly differed (p < 0.05; Table 6). pH decreased in all groups up to Tₐ, and this decrease was more pronounced in the PARGL group than in the other groups. This decrease was followed by a slight increase in all groups except the PARGL group. In Phase 2, the responses of the groups varied considerably over time.

PCO₂ showed no significant interaction between groups × times (p > 0.05) and no differences between the total number of groups in Phase 2 and the total number of times in both phases. However, there was a significant difference (p<0.05) in the total number of groups in Phase 1 (Table 7). Values close to or above the reference limits for the species were observed in the ENTFL and PARFL groups between T3 and T7, with concentrations returning to the reference values after the end of Phase 1.

The concentration of TCO₂ did not show significant interaction between groups × times (p>0.05) in both phases and difference between the total number of groups and the total number of times in Phase 2. The total number of groups and the total number of times significantly differed in Phase 1 (p < 0.05; Table 8). Despite these differences, the values remained within the reference limits for the species throughout the experimental period.

Table 4. Mean + standard error of serum magnesium concentration (mg/dL) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTFL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Tempos | ENTFL | ENTGL | PARFL | PARGL |
|--------|-------|-------|-------|-------|
|        | T₁    | T₂    | T₃    | T₄    |
|        | 2.10 ± 0.15 | 2.62 ± 0.12 | 2.17 ± 0.17 | 2.37 ± 0.22 |
|        | 2.00 ± 0.15 | 2.30 ± 0.38 | 1.75 ± 0.19 | 2.05 ± 0.24 |
|        | 1.95 ± 0.20 | 2.37 ± 0.47 | 2.05 ± 0.25 | 1.72 ± 0.15 |
|        | 1.92 ± 0.30 | 1.95 ± 0.29 | 2.00 ± 0.27 | 2.00 ± 0.25 |
|        | 5.07 ± 3.4 | 1.92 ± 0.27 | 1.85 ± 0.30 | 1.87 ± 0.31 |
|        | 2.25 ± 0.27 | 2.02 ± 0.28 | 1.85 ± 0.35 | 1.72 ± 0.17 |
|        | 1.75 ± 0.32 | 2.26 ± 0.33 | 2.00 ± 0.07 | 1.65 ± 0.24 |
|        | 2.43 ± 0.48 | 2.20 ± 0.11 | 1.94 ± 0.08 | 1.91 ± 0.09 |

| Tempos | ENTFL | ENTGL | PARFL | PARGL |
|--------|-------|-------|-------|-------|
|        | T₅    | T₆    | T₇    | T₈    |
|        | 2.10 ± 0.24 | 6.36 ± 3.83 | 1.97 ± 0.23 | 2.35 ± 0.37 |
|        | 2.30 ± 0.33 | 2.00 ± 0.20 | 2.00 ± 0.27 | 2.72 ± 0.41 |
|        | 2.35 ± 0.29 | 2.36 ± 0.28 | 1.90 ± 0.27 | 6.90 ± 5.23 |
|        | 2.42 ± 0.26 | 2.13 ± 0.35 | 2.15 ± 0.27 | 2.93 ± 0.69 |
|        | 2.40 ± 0.18 | 2.26 ± 0.41 | 1.85 ± 0.16 | 2.73 ± 0.57 |
|        | 2.26 ± 0.21 | 1.93 ± 0.38 | 2.32 ± 0.18 | 2.30 ± 0.62 |
|        | 2.30 ± 0.09 | 2.84 ± 0.66 | 2.03 ± 0.09 | 3.24 ± 0.76 |

Reference value: 2.2 – 2.8 mg/dL. Source: Carlson (2006).

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Table 5. Mean ± standard error of serum total calcium concentration (mg/dL) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total  |
|-------|---------------------|--------|
|       | ENTFL | ENTGL | PARFL | PARGL |   |
| Phase 1 - Starvation |       |       |       |       |   |
| \(T_1\)  | 7.12 ± 0.25 | 7.37 ± 0.75 | 8.87 ± 1.36 | 8.17 ± 0.51 | 7.88 ± 0.41 |
| \(T_2\)  | 7.70 ± 0.24 | 8.15 ± 0.22 | 9.07 ± 1.07 | 7.92 ± 0.30 | 8.21 ± 0.29 |
| \(T_3\)  | 7.80 ± 0.18 | 8.20 ± 0.14 | 8.55 ± 0.48 | 7.97 ± 0.40 | 8.13 ± 0.16 |
| \(T_4\)  | 7.72 ± 0.12 | 7.65 ± 0.57 | 8.50 ± 0.89 | 7.40 ± 0.30 | 7.81 ± 0.27 |
| \(T_5\)  | 7.80 ± 0.12 | 7.80 ± 0.72 | 9.37 ± 1.44 | 7.77 ± 0.43 | 8.18 ± 0.41 |
| \(T_6\)  | 7.32 ± 0.31 | 7.90 ± 0.42 | 9.00 ± 1.26 | 7.60 ± 0.30 | 7.95 ± 0.35 |
| \(T_7\)  | 7.90 ± 0.12 | 8.10 ± 0.43 | 8.92 ± 1.50 | 6.77 ± 0.08 | 7.91 ± 0.42 |
| Total   | 7.62 ± 0.08\(a\) | 7.87 ± 0.18\(a\) | 8.90 ± 0.40\(a\) | 7.66 ± 0.14\(a\) |   |

Reference value: 7.32 – 7.45. Source: Carlson (2006). Means followed by different uppercase letters on the line differ (p < 0.05 – Duncan’s test).

Table 6. Mean ± standard error of blood pH of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total  |
|-------|---------------------|--------|
|       | ENTFL | ENTGL | PARFL | PARGL |   |
| Phase 1 - Starvation |       |       |       |       |   |
| \(T_1\)  | 7.45 ± 0.00 | 7.45 ± 0.00 | 7.43 ± 0.01 | 7.45 ± 0.00 | 7.45 ± 0.00\(a\) |
| \(T_2\)  | 7.45 ± 0.02 | 7.42 ± 0.02 | 7.41 ± 0.00 | 7.46 ± 0.00 | 7.43 ± 0.00\(b\) |
| \(T_3\)  | 7.41 ± 0.02 | 7.40 ± 0.02 | 7.39 ± 0.00 | 7.38 ± 0.02 | 7.40 ± 0.01\(c\) |
| \(T_4\)  | 7.45 ± 0.00 | 7.41 ± 0.02 | 7.40 ± 0.02 | 7.42 ± 0.03 | 7.42 ± 0.01\(ac\) |
| \(T_5\)  | 7.46 ± 0.00 | 7.42 ± 0.01 | 7.40 ± 0.02 | 7.40 ± 0.00 | 7.42 ± 0.00\(ab\) |
| \(T_6\)  | 7.46 ± 0.01 | 7.45 ± 0.00 | 7.40 ± 0.02 | 7.37 ± 0.01 | 7.41 ± 0.01\(bc\) |
| Total   | 7.44 ± 0.00\(a\) | 7.42 ± 0.00\(ab\) | 7.41 ± 0.00\(b\) | 7.42 ± 0.00\(b\) |   |

Reference value: 7.32 – 7.45. Source: Carlson (2006). The mean followed by different uppercase letters in the row and lowercase letters in the column differ (p < 0.05 – Duncan’s test).
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Table 7. Mean + standard error of the partial pressure of CO\(_2\) (mEq/L) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total |
|-------|---------------------|-------|
|       | ENTFL | ENTGL | PARFL | PARGL |
| **Phase 1 – Starvation** | | | | |
| \(T_1\) | 44.72 ± 1.53 | 44.92 ± 1.10 | 45.75 ± 2.50 | 45.50 ± 1.91 | 45.22 ± 0.82 |
| \(T_2\) | 42.42 ± 2.33 | 44.62 ± 2.61 | 43.82 ± 1.28 | 40.57 ± 1.28 | 42.86 ± 0.96 |
| \(T_3\) | 44.35 ± 2.60 | 46.45 ± 4.37 | 45.57 ± 2.83 | 43.97 ± 1.88 | 45.08 ± 1.39 |
| \(T_4\) | 42.13 ± 1.06 | 45.70 ± 4.38 | 47.50 ± 2.95 | 42.83 ± 1.27 | 44.83 ± 1.51 |
| \(T_5\) | 43.96 ± 1.93 | 47.30 ± 5.06 | 46.95 ± 2.56 | 42.63 ± 0.14 | 45.48 ± 1.59 |
| \(T_6\) | 41.93 ± 0.67 | 47.42 ± 4.23 | 48.30 ± 4.22 | 39.46 ± 1.68 | 44.79 ± 1.86 |
| \(T_7\) | 44.06 ± 1.91 | 45.16 ± 1.24 | 48.12 ± 3.79 | 42.06 ± 2.17 | 45.10 ± 1.38 |
| Total | 43.42 ± 0.68 | 45.97 ± 1.26 | 46.57 ± 1.04 | 42.55 ± 0.67 |

Reference value: 38.00 – 46.00 mEq/L. Source: Carlson (2006). The averages followed by different capital letters on the line differ (p<0.05 – Duncan’s test).

Table 8. Mean + standard error of total CO\(_2\) (mEq/L) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total |
|-------|---------------------|-------|
|       | ENTFL | ENTGL | PARFL | PARGL |
| **Phase 1 – Starvation** | | | | |
| \(T_1\) | 33.00 ± 1.22 | 32.50 ± 1.04 | 32.00 ± 1.22 | 33.50 ± 1.19 | 32.75 ± 0.54 |
| \(T_2\) | 31.00 ± 0.91 | 30.20 ± 0.89 | 29.50 ± 1.19 | 30.50 ± 1.44 | 30.30 ± 0.52 |
| \(T_3\) | 28.87 ± 0.82 | 30.25 ± 0.94 | 29.27 ± 1.02 | 26.90 ± 0.98 | 28.82 ± 0.52 |
| \(T_4\) | 30.33 ± 0.88 | 30.75 ± 1.93 | 31.25 ± 1.93 | 30.00 ± 2.08 | 30.64 ± 0.82 |
| \(T_5\) | 32.66 ± 1.85 | 32.50 ± 2.32 | 30.77 ± 1.47 | 28.00 ± 0.00 | 31.07 ± 0.92 |
| \(T_6\) | 31.00 ± 0.57 | 33.50 ± 2.10 | 32.25 ± 1.25 | 26.00 ± 1.00 | 31.00 ± 1.01 |
| \(T_7\) | 31.33 ± 1.33 | 32.66 ± 1.33 | 31.25 ± 0.85 | 26.00 ± 1.00 | 30.38 ± 0.85 |
| Total | 31.14 ± 0.46 | 31.73 ± 0.59 | 30.90 ± 0.48 | 28.90 ± 0.68 |

Reference value: 24.00 – 32.00 mEq/L. Source: Carlson (2006). Averages followed by different uppercase letters in the row and lowercase letters in the column differ (p<0.05 – Duncan’s test).
The anions gap did not have a significant interaction between time and group (p > 0.05) and no significant difference (p > 0.05) between the total number of groups and the total times of the phases (Table 11). Although the groups and times did not differ, anion gap varied between the groups. Anion gap increased in the PARGL group and suddenly decreased after the horses were re-fed. The other groups also showed a numerical decrease in anion gap values after the start of re-feeding, with lower mean values observed at T\_8 in the ENTFL group and at T\_9 in the other groups. This numerical decrease was followed by an increase in the mean values of all groups until T\_10.

**Table 9.** Mean ± standard error of bicarbonate concentration (mEq/L) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total |
|-------|---------------------|-------|
|       | ENTFL               | ENTGL | PARFL | PARGL    |
| **Phase 1 – Starvation** | | | | |
| T\_1  | 31.50 ± 1.10        | 31.35 ± 1.00 | 30.77 ± 1.24 | 32.30 ± 1.16 | 31.48 ± 0.52\(^a\) |
| T\_2  | 29.47 ± 0.83        | 28.97 ± 0.90 | 27.97 ± 1.05 | 29.47 ± 1.49 | 28.97 ± 0.51\(^b\) |
| T\_3  | 28.35 ± 0.25        | 28.82 ± 0.77 | 28.02 ± 1.04 | 26.30 ± 0.50 | 27.87 ± 0.40\(^b\) |
| T\_4  | 28.93 ± 0.88        | 29.35 ± 1.89 | 29.92 ± 1.71 | 28.70 ± 2.04 | 29.28 ± 0.78\(^b\) |
| T\_5  | 31.33 ± 1.66        | 30.67 ± 2.16 | 29.42 ± 1.31 | 26.93 ± 0.17 | 29.65 ± 0.84\(^b\) |
| T\_6  | 29.33 ± 0.72        | 32.20 ± 1.95 | 30.87 ± 1.24 | 24.76 ± 0.83 | 29.61 ± 0.98\(^b\) |
| T\_7  | 29.93 ± 1.48        | 31.46 ± 1.19 | 29.95 ± 0.90 | 24.90 ± 1.05 | 29.13 ± 0.85\(^b\) |
| **Total** | 29.82 ± 0.40\(^a\) | 30.36 ± 0.56\(^a\) | 29.56 ± 0.46\(^a\) | 27.84 ± 0.66\(^a\) |

| Times | Experimental Groups | Total |
|-------|---------------------|-------|
|       |                     |       |
| **Phase 2 – Refeeding** | | | | |
| T\_8  | 30.13 ± 1.19        | 31.00 ± 1.16 | 31.55 ± 1.82 | 23.83 ± 1.05 | 29.31 ± 1.08 |
| T\_9  | 31.80 ± 0.75        | 29.16 ± 0.43 | 31.22 ± 3.19 | 26.73 ± 0.84 | 29.84 ± 1.07 |
| T\_10 | 30.03 ± 1.55        | 28.23 ± 0.28 | 31.73 ± 2.43 | 28.50 ± 1.96 | 29.62 ± 0.85 |
| T\_11 | 28.13 ± 0.52        | 31.10 ± 2.99 | 31.70 ± 2.95 | 29.13 ± 2.18 | 30.01 ± 1.10 |
| T\_12 | 28.53 ± 0.33        | 27.36 ± 1.95 | 32.20 ± 2.08 | 27.96 ± 2.71 | 29.01 ± 1.01 |
| T\_13 | 28.70 ± 0.30        | 30.13 ± 1.61 | 32.43 ± 2.64 | 28.23 ± 2.21 | 29.98 ± 1.03 |
| **Total** | 29.60 ± 0.45\(^a\) | 29.50 ± 0.66\(^a\) | 31.76 ± 0.92\(^a\) | 27.40 ± 0.79\(^a\) |

Reference value: 20.00 – 28.00 mEq/L. Source: Carlson (2006). The averages followed by different uppercase letters in the row and lowercase letters in the column differ from each other (p < 0.05 – Duncan’s test).

**Discussion**

Blood gas analysis and electrolyte measurement are important laboratory tests for characterizing and evaluating the intensity of hydroelectrolyte and acid-base imbalances in different clinical situations; they also enable veterinarians to institute appropriate therapeutic interventions (Ribeiro Filho et al., 2007) or monitor a patient’s response to established therapies.

This study showed that hyponatremia developed in the PARGL group in both phases. Hyponatremia is usually associated with cases involving intense sodium loss, such as enterogastric...
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Table 10. Mean + standard error of base deviation of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total |
|-------|---------------------|-------|
|       | ENTFL   | ENTGL   | PARFL  | PARGL  |
| Phase 1 – Starvation | |
| T₁    | 7.75 ± 1.03 | 7.50 ± 1.04 | 6.75 ± 1.37 | 8.50 ± 1.19 | 7.62 ± 0.54<sup>a</sup> |
| T₂    | 5.50 ± 0.86 | 4.50 ± 1.19 | 3.50 ± 1.25 | 5.75 ± 1.70 | 4.81 ± 0.62<sup>c</sup> |
| T₃    | 3.97 ± 0.83 | 4.00 ± 0.40 | 3.50 ± 1.25 | 1.20 ± 0.77 | 3.16 ± 0.49<sup>c</sup> |
| T₄    | 4.66 ± 1.20 | 4.75 ± 1.97 | 5.00 ± 1.95 | 5.33 ± 1.85 | 4.92 ± 0.82<sup>c</sup> |
| T₅    | 7.33 ± 1.66 | 6.25 ± 2.09 | 4.75 ± 1.43 | 2.33 ± 0.33 | 5.21 ± 0.87<sup>b</sup> |
| T₆    | 5.66 ± 0.88 | 8.25 ± 1.88 | 7.50 ± 0.86 | 1.66 ± 2.33 | 6.07 ± 0.98<sup>b</sup> |
| T₇    | 5.66 ± 1.76 | 7.00 ± 1.00 | 5.25 ± 0.75 | 0.66 ± 1.20 | 4.38 ± 0.96<sup>c</sup> |
| Total | 5.78 ± 0.46<sup>a</sup> | 6.00 ± 0.58<sup>a</sup> | 5.17 ± 0.51<sup>b</sup> | 3.65 ± 0.78<sup>b</sup> |

Reference value: -2.00 – 4.00 mEq/L. Source: Carlson (2006). The mean followed by different uppercase letters in the row and lowercase letters in the column differ (p < 0.05 – Duncan’s test).

Table 11. Mean + standard error of the anion gap (mEq/L) of horses under total parenteral nutrition associated with glutamine (PARGL), enteral fluid therapy with (ENTGL) or without glutamine (ENTFL), and fluid therapy (PARF) submitted to starvation after exploratory laparotomy.

| Times | Experimental Groups | Total |
|-------|---------------------|-------|
|       | ENTFL   | ENTGL   | PARFL  | PARGL  |
| Phase 1 – Starvation | |
| T₁    | 8.00 ± 0.40 | 7.00 ± 2.00 | 9.00 ± 0.91 | 7.80 ± 0.52 | 7.95 ± 0.54 |
| T₂    | 7.75 ± 0.47 | 8.25 ± 0.75 | 8.00 ± 1.47 | 8.75 ± 1.25 | 8.18 ± 0.48 |
| T₃    | 6.75 ± 0.94 | 9.25 ± 0.47 | 8.50 ± 0.64 | 8.66 ± 0.66 | 8.18 ± 0.38 |
| T₄    | 7.33 ± 0.33 | 9.25 ± 1.37 | 9.00 ± 0.70 | 8.66 ± 0.66 | 8.64 ± 0.46 |
| T₅    | 7.00 ± 0.57 | 9.25 ± 1.03 | 9.00 ± 0.91 | 9.33 ± 0.33 | 8.71 ± 0.50 |
| T₆    | 7.66 ± 0.88 | 9.00 ± 0.91 | 7.97 ± 1.06 | 11.00 ± 1.00 | 8.85 ± 0.55 |
| T₇    | 7.00 ± 1.15 | 7.66 ± 1.20 | 8.25 ± 0.62 | 11.00 ± 1.00 | 8.46 ± 0.59 |
| Total | 7.37 ± 0.25 | 8.55 ± 0.43 | 8.53 ± 0.32 | 9.13 ± 0.39 |  |

Phase 2 – Refeeding | |
| T₈    | 7.00 ± 1.00 | 5.00 ± 2.08 | 7.25 ± 1.10 | 8.33 ± 2.33 | 8.75 ± 0.50 |
| T₉    | 6.00 ± 0.57 | 7.33 ± 0.88 | 6.00 ± 2.08 | 6.66 ± 1.76 | 6.91 ± 1.04 |
| T₁₀   | 7.66 ± 0.66 | 9.00 ± 1.00 | 8.66 ± 0.66 | 9.66 ± 1.66 | 7.66 ± 0.94 |
| T₁₁   | 7.00 ± 1.00 | 8.66 ± 1.20 | 8.33 ± 1.76 | 7.66 ± 1.20 | 8.36 ± 0.80 |
| T₁₂   | 7.33 ± 1.20 | 8.33 ± 0.33 | 5.33 ± 1.76 | 9.33 ± 2.96 | 6.92 ± 0.78 |
| T₁₃   | 7.50 ± 1.50 | 8.25 ± 0.75 | 6.33 ± 0.88 | 11.00 ± 2.08 | 6.92 ± 0.72 |
| Total | 7.05 ± 0.35 | 7.16 ± 0.78 | 7.25 ± 0.59 | 8.77 ± 0.79 |  |

Reference value: 6.00 – 15.00 mEq/L. Source: Carlson (2006).
reflux, diarrhea, profuse sweating, and adrenal insufficiency (Carlson, 2006); however, none of these etiological factors were identified in the present study. Hyponatremia manifested in the PARGL group likely because of the supply of lower amounts of sodium than that in the other groups. In the PARGL group, water was provided mainly by an electrolyte-free glutamine solution and TPN, and the remaining maintenance requirement was provided by a lactated Ringer's solution. Therefore, providing large amounts of free water would cause a decrease in serum electrolyte levels. However, studies in humans have shown that the total amount of sodium and fluids administered during TPN does not influence the onset of hyponatremia because the kidneys in physiological situations can eliminate excess water and thus maintain homeostasis (Gómez-Hoyos et al., 2019).

Na⁺ is also reduced probably because of the occurrence of false hyponatremia resulting from an increase in triglyceride, total protein, and glucose values in the PARGL group (unpublished data). Hypertriglyceridemia and/or intense hyperproteinemia produce a falsely low Na⁺ because lipids or proteins occupy a significantly higher volume in blood samples and because Na⁺ is present only in the aqueous phase. Severe hyperglycemia causes a reduction in the measured serum Na⁺ concentration of approximately 1.6 mEq/L for every 100 mg/dL increase in glucose concentration. When glucose concentration increases, osmotic forces are generated, causing water to move from the intracellular fluid to the extracellular fluid; consequently, plasma Na⁺ concentration becomes diluted (Carlson, 2006). In the other groups, the supply of isotonic polyionic solutions failed to prevent the decrease in Na⁺ concentration although no hyponatremia occurred.

Serum potassium levels decreased in Phase 1 due to decreased intake of this electrolyte during the starvation phase. Although the polyionic solution used in the ENTFL and ENTGL groups contained potassium, the amount supplied was insufficient to prevent this decrease in circulation. This decrease could also be attributed to increased hormone release, such as insulin and catecholamines. Excitement and pain, which are common in horses exposed to stress, surgical or not, trigger the release of catecholamines, leading to hyperglycemia and a consequent decrease in potassium values (Di Fillippo et al., 2008). However, stress-associated hyperglycemia cannot be considered a probable cause, since only the groups that received 50% glucose (PARFL and PARGL) developed hyperglycemia; conversely, glucose concentration remained stable in the ENTFL and ENTGL groups. The potassium concentration in the PARGL group decreased markedly due to the development of a hyperglycemic condition associated with the provision of TPN in Phase 1. Once the food supply was restored, the potassium concentration increased and reached values similar to the initial levels.

The total calcium levels remained relatively constant throughout the study period although they were below the normal values for horses. However, the absence of a significant difference (p>0.05) at the beginning of the study (T1) demonstrated the homogeneity of the experimental groups. The results obtained in the present study were similar to those observed by other researchers who did not observe changes in calcium concentrations during a 10-day starvation period in a group of horses (Baetz & Pearson, 1972).

The magnesium ion response observed in our study partly corroborated the data of Baetz and Pearson (1972), who did not identify a change in magnesium concentration during the first 6 days of a 10-day starvation period in horses. Regardless of the absence of a significant difference, the magnesium concentration most remarkably decreased in the PARGL group although it decreased below the normal range in the other groups in at least one part of Phase I. Magnesium concentration probably decreased due to the absence of dietary intake associated with the provision of magnesium-free fluid therapy in Phase 1. The small number of animals used per group and the variability of the response in each group and individual resulted in data overlap, preventing the identification of differences.

In addition to the absence of dietary intake (Stewart, 2011), hypomagnesemia development is closely associated with systemic inflammation and cytokine release (Sugiura et al., 2000). Thus, in the present study, hypomagnesemia might be initially associated with an inflammatory process triggered by laparotomy and surgical manipulation for obtaining the samples. However, the return of its concentration to values within the normal range or close in the refeeding phase, mainly in the ENTFL, PARFL, and PARGL groups, suggested that the absence of ingestion was the main cause of the decrease in magnesium concentration in Phase 1. This decrease often follows a
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Changes in the acid-base and electrolyte balance were more intense in the PARGL group than in the other groups even though such changes also occurred in the other groups. More studies are needed to fully understand the mechanisms behind these changes and to identify optimal nutritional strategies to prevent metabolic alkalosis in horses under similar conditions.
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are needed to determine the optimal formulation of TPN for horses. Despite the promising results of this study, its use should not be discouraged because it has numerous clinical benefits. These results demonstrate the need to monitor blood gas and electrolyte balance in horses with food restriction under nutritional support or prolonged fluid therapy so that changes are promptly corrected.

Ethics statement
This study was approved by the Animal Experimentation Ethics Committee (CETEA/UFMG) under number 34/2008.

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Conflict of interests
The authors declare no conflicts of interest in the preparation, execution, and dissemination of the results of this study.

Authors’ contributions
UPM, MSP, CF, FOPL, VAG - Development of methodology; preparation and writing the initial draft. UPM - Writing, Review and Editing manuscript. MSP - Statistical analysis.

Availability of complementary results
Non complementary information.

The study was carried out at horse’s stud in Escola de Veterinária da UFMG (EV/UFMG), Belo Horizonte, MG, Brazil.

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