American Journal of Animal and Veterinary Sciences

Original Research Paper

Evaluation of Different Oral Rehydration Solutions for Diarrheic Dairy Calves

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Article history
Received: 09-08-2018
Revised: 02-11-2018
Accepted: 22-11-2018

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Abstract: Regardless of the causative agent, diarrhea results in large loss of water and electrolytes, which leads to reduced performance and mortality. The aim of this study was to evaluate oral rehydration solutions in order to determine the most effective in restoring electrolytes and water while maintaining animal performance. Forty two calves (14 per treatment) were distributed in one of three treatments (1) Common electrolytes solution (dextrose, sodium bicarbonate and sodium chloride; 507±2 mOsm), (2) Glutellac® (sodium acetate, glucose and sodium and potassium chloride; 665±1 mOsm); and (3) Common electrolytes solution + Aminogut® (glutamate and glutamine; 575±2 mOsm). Treatments were administered when fecal score ≥3, in a scale of 1 to 5. During diarrhea daily water intake was registered and blood sample was taken for metabolites, electrolytes and gases determination. Animals did not differ in days in diarrhea, fecal score or hematocrit. Calves receiving Glutellac® presented higher voluntary water intake. There was no effect of treatment for animal performance and starter feed intake. Concentrations of Na^+ and HCO_3 tended to be higher for animals rehydrated with Glutellac®. While there was an increase in HCO_3 from the first to the second day, K^+ and glucose decreased. The increased voluntary water intake in animals rehydrated with Glutellac®, which shows greater simplicity of use, are the main advantages of this solution.

Keywords: Calf Health, Dehydration, Electrolytes, Neonatal Diarrhea

Introduction

The main cause of death in dairy calves from birth until weaning is diarrhea, followed by respiratory diseases. This metabolic disorder may be the result of faults in colostrum ingestion, leading to failure of passive immunity transfer; poor nutrition during liquid-feeding phase; or even a consequence of management failures. Diarrhea causes great economic losses, related to lower performance, mortality, costs with veterinary treatments and labor to treat animals affected (Torsein *et al*., 2011).

Diarrhea may present an infectious origin, when caused by microorganisms such as bacteria, viruses and protozoa; or may occur as a response of alimentary factors that might change osmotic pressure in the intestine (Blanchard, 2012). Despite of causal agent, in general diarrhea results in a great loss of water and electrolytes, due to morphological damage in the intestinal mucosa, which leads to an increase in the susceptibility to bacterial attack (Constable, 2004). In most cases of diarrhea, calf's death occurs because of dehydration and loss of electrolytes and not directly by the action of the pathogens (Smith and Berchtold, 2014). Thus, the maintenance of water balance and electrolytes is a critical factor for animal survival (Davis and Drackley, 1998). In this way, oral electrolytes solutions are used in an attempt to restore the electrolytes level in animals with dehydration less than 8%, however with suckle reflex, which suffer from reduction in blood volume (hypovolemia), metabolic acidosis or hyponatremia (low blood sodium), as a result of the diarrhea (Lorenz *et al*., 2011). These solutions should provide sufficient sodium concentration to normalize the extracellular fluid, in addition to agents as glucose, acetate, propionate or glycine, which facilitate the absorption of sodium and water in the intestine. Solutions must also provide alkalizing agents, as acetate, propionate or sodium bicarbonate to treat metabolic acidosis and provide enough energy (Smith and Berchtold, 2014).

Some amino acids can also be included in the formulation of these rehydration solutions, like glutamine that facilitates Na absorption in the intestine and hepatic absorption of glucose, but are also responsible for maintaining the shape and function of intestinal villi (Van der Hulst *et al*., 1993; Rao and Samak, 2012). This function became interesting in diarrheic calves because many times liquid feeding intake is diminish affecting the enteric function (Brooks *et al*., 1997). However, responses in animal models, to glutamine supplementation have been inconsistent (Naylor *et al*., 1997; Brooks *et al*., 1998; Drackley *et al*., 2006).

There is no certainty about ideal electrolyte concentrations, type of buffer and type, amount and source of energy, as well as the pH and osmotic pressure of the rehydration solution (Sayers *et al*., 2016). However, it is clear that an important aspect to consider about oral rehydration solutions is the osmolality, which is related mainly with glucose content of the solution. In that sense, hypertonic solutions (>312 mOsm/L) provide greater nutritional support as compared with isotonic solutions (280-300 mOsm/L) and reduce the body weight loss that normally occurs when diarrheic calves are deprived of milk (Constable *et al*., 2001). Although milk is more efficient to maintain blood glucose concentration, oral rehydration solutions are more effective in rehydrating and replacing electrolytes because the higher sodium concentration (Constable *et al*., 2001). However, milk feeding should continue in diarrheic calves that are being rehydrated with oral solutions, since it allows the calf to maintain growth (Goodell *et al*., 2012). While hyperosmotic rehydration solutions are recommended for treatment of diarrheic calves, extremely high osmolalities (>700-750 mOsm/L) could exacerbate hypersecretion of electrolytes and water into the small intestinal lumen (Sen *et al*., 2009). This can increase the risk of bloat and/or abomasitis because the lower empty rate of the abomasum causing anorexia, abdominal distension and often death in 6 to 48 hours (Sen *et al*., 2009).

During the occurrence of diarrhea, calves tend to increase water intake because of the great losses of fluid in feces (Wenge *et al*., 2014). There are evidences that animals receiving oral electrolytic solutions mixed with the liquid diet, consumes greater volumes of water compared with those that receive the therapy of oral hydration separated (Wenge *et al*., 2014). This is because these solutions cause thirst sensation, incrementing the voluntary water intake. However, this type of treatment implies that water supply is available ad libitum in order to avoid situations of hypernatremia (Wenge *et al*., 2014).

Therefore, the hypothesis of this study was that water and milk-based oral rehydration solutions, provide different effects on acid-base status affecting the recovery time from diarrhea and development of the animal. Therefore, the aim of the present study was to compare three solutions for oral rehydration, in regard to restore electrolytes and water, maintaining the performance of calves affected by diarrhea acquired under natural challenge conditions.

Materials and Methods

All experimental procedures were approved by the Animal Care and Use committee of University of São Paulo and were performed in accordance with their guidelines. Forty-two newborn crossbred Holstein-Jersey calves (23 male, 36±9 kg; and 19 female, 30.8±9 kg) from the herd of the University of São Paulo - College of Agriculture 'Luiz de Queiroz', Department of Animal Science - (USP), were blocked by sex, birth weight and level of total serum protein at 48 h. Only calves with total serum protein higher than 5.5 g/dL were enrolled in the study. After birth, calves received 2 L of high quality colostrum for the first meal and 12 h later and were housed in individual hutches with free access to water and starter concentrate (AgroceresMultimix Nutrition Animal Ltda, Rio Claro, SP, Brazil) during the entire experimental period. Calves were weighed and randomly distributed in one of the following rehydration solution, resulting in 14 calves per treatment: (1) Common handmade electrolytes solution, composed of 25 g Dextrose, 10 g of sodium bicarbonate and 5 g of sodium chloride, diluted in 1 L of water (osmolality 507±2 mOsm), administered at 10:00 h and at 14:00h; (2) Glutellac: a commercial electrolyte solution, based on sodium acetate 18.325 g, glucose 19.5 g, sodium and potassium chloride 3.2 g, flavoring 0.015 g and sodium diacetate 1.6 g in a blister of 50 mL (Bayer SA, São Paulo, Brazil) added to the milk replacer (osmolality 665±1 mOsm); and (3) Common handmade electrolytes solution (25 g of dextrose, 10 g of sodium bicarbonate, 5 g of sodium chloride) and 22 g of Aminogut (glutamine and glutamate mixture, Ajinomoto of Brazil Industria e Comercio de Alimentos Ltda Limeira São Paulo), diluted in 1L water and supplied at 10:00 h and at 14:00 h (osmolality 575±2 mOsm). Animals from all the three treatments continued receiving milk replacer while the course of naturally acquired diarrhea.

Fecal score was monitored daily as described by Larson *et al*. (1977) regarding the fluidity of feces, being: (1) Normal and firm, (2) loose but with general healthy aspect, (3) very loose no watery separation, (4) watery and (5) very watery. The solutions were provided when the fecal score was ≥3, until score was lower than or equal to 2 for two consecutive days. Because common oral solutions was bottle fed calves received 2 L of water a day, which did not occurred with calves fed Glutellac. Because of that, during occurrence of naturally acquired diarrhea, voluntary water intake from the buckets was daily measured. Rectal temperature, respiratory frequency and heartbeat were also measured during diarrhea occurrence.

Table 1: Milk replacer and starter concentrate chemical composition

	Milk replacer[1]	Starter concentrate[2]
Dry matter, %	96.94	91.06
Ash, % DM	7.24	7.81
Crude protein, % DM	19.50	21.19
Crude fat, % DM	14.37	5.04
NFD, % DM	0.65	17.91
Digestivel total nutrients (DTN), % DM	-	83.58
Gross energy, Mcal/kg	4,557.92	-

[1]Feedtech, De Laval Ltda., Campinas, São Paulo, Brazil (12.5% solids)
[2]Ag Milk Agroceres Multimix Nutrição Animal Ltda

After birth, calves received 4 L/d of colostrum in two daily meals, until the second day of life, being fed 4 L/d of milk replacer (19.5% crude protein, 14.4% fat, 12.5% solids; Feedtech, DeLaval Ltda., Campinas, São Paulo, Brazil) at 7 am and 5 pm. Calves were housed in individual shelters and had free access to water and starter concentrate (Ag Milk AgroceresMultimix Nutrition Animal Ltda, Rio Claro, São Paulo, Brazil) during the entire experimental period, with orts being weighted to calculate daily intake. Samples of starter concentrate and milk replacer were collected during the experimental period for subsequent analysis (Table 1). Animals were weighed weekly in a mechanical scale (ICS-300, Coimma Ltda., Dracena, SP, Brazil), before the morning feeding, until the 8th week of life.

Blood samples were collected weekly, regardless of diarrhea occurrence, always two hours after the morning feeding, through jugular venipuncture by vacuum tubes containing sodium fluoride and potassium EDTA (Vacuette of Brazil, Campinas, SP, Brazil). Samples were centrifuged (Universal 320R, Hettich, Tuttlinger, German) at 2.000 x *g*, for 20 min at 4°C and plasma or serum were stored in a freezer (-26°C) until subsequent analysis. Specific commercial enzymatic kits (LabtestDiagnóstica S.A., Lagoa Santa, MG, Brazil) were used to analyzed plasma glucose (Ref.:85), total serum protein (Ref.:99), serum creatinine (Ref.:35), plasma urea (Ref.:104) and serum albumin (Ref.:19), using the Automatic System for Biochemistry - SBA Model 200 (CELM, Barueri, SP, Brazil). Hematocrit was determined with an aliquot of blood collected from tube containing anticoagulant, using a microcentrifuge hematocrit SPIN model 1000 (MICROSPIN).

During the first two days after the detection of diarrhea, a blood sample was taken two hours after morning feeding, with vacuolated tubes containing sodium heparin as an anticoagulant for blood analyzes of gases, electrolytes, using a portable i-Stat® (Abbot Point of Care Inc., Princeton, New Jersey, USA) and the $EC8^+$ cartridge (Abbot Point of Care Inc., Princeton, New Jersey, USA).

The data of total feed intake, live weight, average daily gain and blood parameters were analyzed as repeated measures (week of age) through the PROC MIXED SAS (1991), with the model: Yijk = μ + Ti + Ij + TIij + Bk + Eijk, where: μ = average general, Ti = effect of treatment, ij = effect of age, TIij = interaction treatment x Age, Bk = effect of block, eijk = experimental error. The data collected while calves were affected by diarrhea such as water intake, blood electrolytes and clinical signs were analyzed through the following model: Yijk = μ + Ti + Bk + Eijk, where: μ = average general, Ti = effect of treatment, Bk = effect of block, eijk = experimental error.

Gasometric analyzes were analyzed through the following model: Yijk = μ + Ti + Ij + TIij + Bk + Dj + Eijk, where: μ=average general, TI = effect of treatment, ij = effect of age, TIij = interaction treatment x Age, Bk = effect of block, DJ = effect of the day on which it was held the blood collection, eijk = experimental error. The averages were compared by the test of least squares (LSMEANS), with a significance level of 5%.

Results

All calves had at least one episode of infectious diarrhea throughout the treatment phase, mainly during the second and third week of life and all of them recovered from diarrhea. The number of days with diarrhea, as well as fecal score, were not affected by rehydration solutions (p>0.05; Table 2; Fig. 1).

Calves that were rehydrated with Common electrolytes solution had the highest total water intake (voluntary water intake + water bottle fed with the rehydration solution) as compared to calves that consumed Glutellac® (p<0.05; Fig. 2; Table 2), while animals that were rehydrated with Common electrolytes solution + Aminogut® had an intermediate intake, with no difference from the other two treatments (p>0.05). On the other hand, calves rehydrated with Glutellac® presented the greatest voluntary water intake (p<0.05), suggesting the thirst effect of this solution.

Rehydration solutions had no significant effect on body weight or average daily gain (p>0.05; Table 3). However, there was significant effect of the animals' age for those variables (p<0.05). Milk replacer, starter concentrate, as well as the total dry matter intake, were also not affected by type of rehydration solution (p>0.05), but there was an intake increase as animal aged (p<0.05). During the second week of life, there was a decrease in milk replacer intake observed for all calves, being restored at the third week of age, from when it remained constant until the eighth week of age, when calves were abruptly weaned. During the occurrence of diarrhea, clinical parameters, such as rectal temperature, also did not show differences among rehydration therapies ($p>0.05$).

Biochemical parameters during the pre-weaning phase were also not affected by the type of oral rehydration solution ($p>0.05$), with average values of 98.2 mg/dL for glucose; 5.88 g/dL for serum protein; 2.34 g/dL for serum albumin; 18.45 mg/dL for plasma urea nitrogen and 0.78 mg/dL for creatinine, being all of them within normal values. These results show that animals were on normal metabolism. However, there was an age effect for all this metabolites with increasing concentrations of glucose, urea and albumin and decreasing concentrations of total serum protein as animals' aged ($p<0.0001$; Table 3). Even though calves were affected by diarrhea episodes, normal growth and metabolism were observed, regardless of the oral solution given.

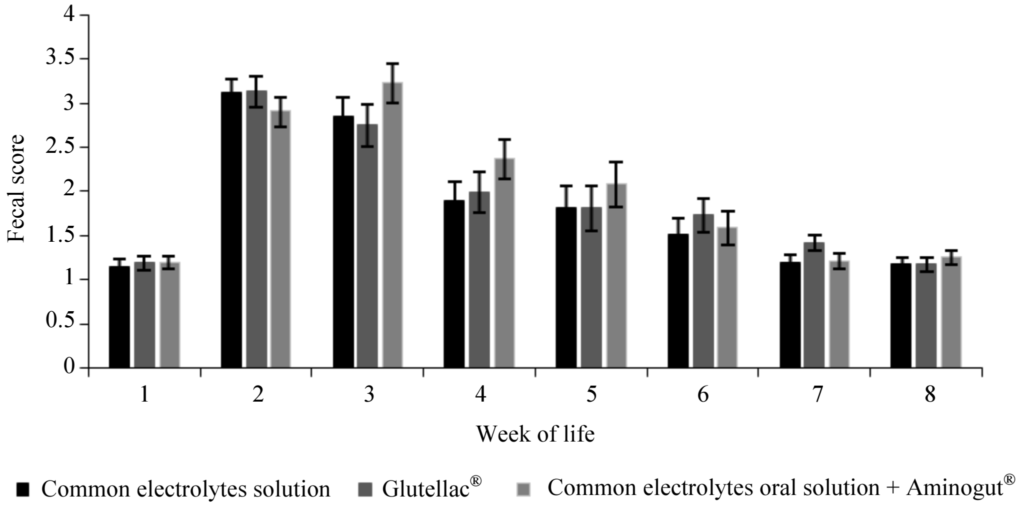

Fig. 1: Fecal score in Crossbred calves during the period of diarrhea, receiving Common electrolytes oral solution, Glutellac® or Common electrolytes oral solution + Aminogut®, as rehydration therapy

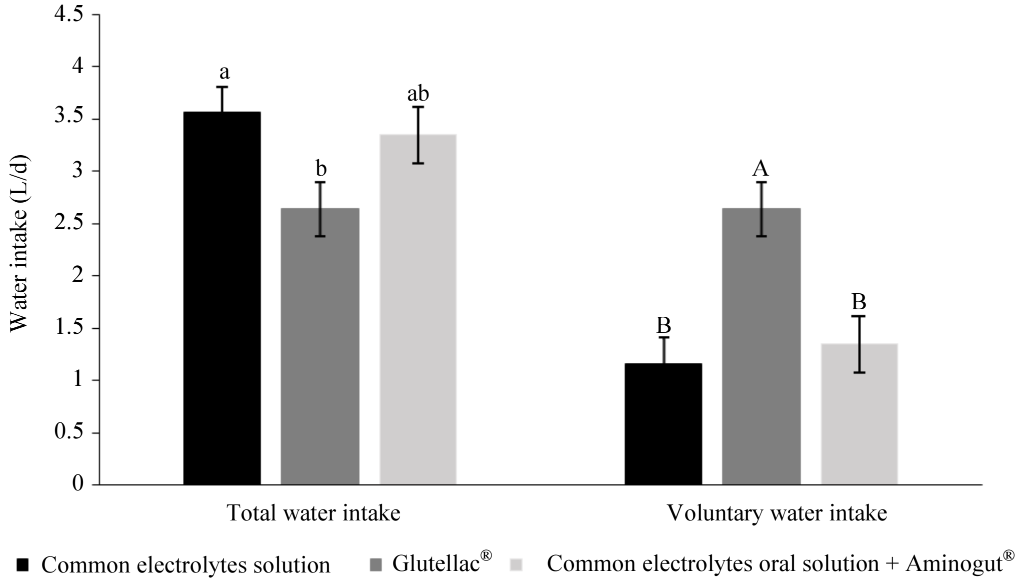

Fig. 2: Total water intake (L/d) and voluntary water intake (L/d) in Crossbred calves during the occurrence of diarrhea, receiving Common electrolytes oral solution, Glutellac® or Common electrolytes oral solution + Aminogut®, as rehydration therapy. a, b; A,B Different letters indicate statistical difference ($p<0.05$)

Table 2: Number of days with diarrhea, fecal score, hematocrit, voluntary water intake and total water intake in Crossbred calves during the first and second day of diarrhea occurrence, receiving different rehydration solutions

	Treatment				*P value*[1]		
	Common electrolytes solution	Glutellac ®	Common electrolytes solution + Amonigut®	SEM	T	A	TxA
Number of days with diarrhea	14.29	14.00	18.36	2.50	0.39	.	.
Fecal score	1.84	1.90	1.98	0.11	0.66	<0.01	0.46
Hematocrit (%)	22.81	24.57	22.83	0.97	0.15	<0.01	0.03
Water intake							
Voluntary water intake	1.16^b	2.64^a	1.35^b	0.26	0.01	.	.
Total water intake	3.56^a	2.64^b	3.35^{ab}	0.26	0.02	.	.

[1]T: treatment effect, A: age effect, TxA: interaction Treatment x age, SEM: Standard error of the mean.
[a,b] Different letters indicate statistical difference ($p<0.05$) within the row.

Table 3: Performance, feed intake and blood metabolites of Holstein-Jersey calves during the pre-weaning phase, receiving different rehydrating solutions when diarrhea occurred

	Treatment				*P value*[1]		
	Common electrolytes solution	Glutellac ®	Common electrolytes solution Amonigut®	SEM	T	A	TxA
Body weight, Kg							
Initial	34.67	35.37	35.77	2.629	0.72	-	-
Weaning	51.22	51.75	50.09	4.067	0.84	-	-
Average total period	39.36	39.31	37.93	2.449	0.63	<0.0100	0.13
Weight gain, g/d							
Average daily gain	287.47	294.84	261.52	28.854	0.65	<0.0100	0.71
Intake, g MS/d							
Starter intake	269.89	293.31	230.36	33.402	0.41	<0.0100	0.37
Milk replacer intake	480.36	476.23	477.85	3.891	0.71	<0.0100	0.87
Total intake	749.89	773.61	707.59	33.941	0.39	<0.0100	0.30
Glucose (mg/dL)	95.26	99.45	99.85	2.149	0.25	<0.0100	0.89
Total Proteín (g/dL)	5.91	5.86	5.87	0.214	0.98	<0.0100	0.92
Albumin (g/dL)	2.34	2.37	2.32	0.054	0.73	<0.0100	0.52
Urea (mg/dL)	18.30	17.76	19.29	0.695	0.30	<0.0001	0.15
Creatinine (mg/dL)	0.80	0.79	0.77	0.040	0.71	<0.0001	0.87

[1]T: treatment effect, A: age effect, TxA: interaction Treatment x age, SEM: Standard error of the mean

Table 4: Blood gas and electrolytes in Crossbred calves, receiving Common electrolytes oral solution, Glutellac® or Common electrolytes oral solution +Aminogut®, as rehydration therapy (Average of first and second day of diarrhea)

	Treatment				*Pvalue*[1]			
	Common Electrolytes solution	Glutellac®	Common electrolytes solution + Amonigut®	SEM	T	D	TxD	A
pH	7.35	7.39	7.37	0.019	0.16	0.07	0.85	0.001
HCO_3 (mmol/L)	26.98	30.18	28.78	1.073	0.00	0.04	0.98	0.06
PCO_2 (mmHg)	48.31	49.29	49.22	1.163	0.80	0.92	0.69	0.42
TCO_2 (mmol/L)	28.43	31.64	30.19	1.093	0.11	0.06	0.93	0.06
Na (mEq/L)	136.03	137.74	135.35	1.058	0.07	0.11	0.79	0.01
K (mEq/L)	5.00	4.80	4.87	0.113	0.45	<0.01	0.77	<0.01
Anion Gap (mmol/L)	13.28	13.70	13.29	0.534	0.90	0.17	0.99	0.47
BE(ecf) (mmol/L)	4.08	4.31	3.20	1.069	0.63	0.05	0.18	0.02
Glucose (mg/dL)	81.82^b	92.46^a	79.02^b	2.538	0.01	0.04	0.05	00.12
Hematocrit (%)	28.78	27.99	25.03	1.594	0.23	0.20	0.49	<0.01
Hemoglobin (g/dL)	9.77	9.52	8.61	0.534	0.29	0.42	0.69	<0.01

[1]T: Treatment effect, D: blood collecting day, TxD: interaction Treatment x blood collecting day, A: animal's age at the evaluation moment, SEM: Standard error of the mean.
[a,b]Different letters indicate statistical difference ($p<0.05$) within the row.

During the occurrence of diarrhea, clinical parameters, such as rectal temperature, also did not show differences among rehydration therapies ($p>0.05$). The concentrations of glucose during the first two days of treatment with rehydration therapy was higher for calves rehydrated with Glutellac® (p<0.05), while the concentrations of HCO_3 and Na^+ tended to be different among treatments (P = 0.088 and P = 0.073, respectively; Table 4). The HCO_3 increased from the first to the second day of blood gas evaluation, while K^+ and glucose decreased and the BE (ecf) varied according to rehydration therapy used (p<0.05). There was a significant effect of interaction treatment x day of evaluation only for the concentration of glucose (p<0.05). Because of that, Table 4 shows average values for the first and second day of diarrhea.

The pH, Na^+ concentration and BE (ecf), were higher, while K^+, hematocrit and hemoglobin, where lower for older calves (p<0.05; Table 4). For calves rehydrated with Common electrolytic solution, HCO_3 had a slight increase from the first to the second day; while PCO_2 decreased, resulting in an increase in pH since the relationship $HCO_3/(PCO_2*0.003)$ increases. An increment in HCO_3 was also observed for the other two rehydration therapies; however, there was also an increment in PCO_2, despite the relationship between these two parameters increases causing a pH increase. The hematocrit and hemoglobin concentration, presented only effect of age at the time of occurrence of diarrhea (p<0.05), without effect of treatments neither the day of diarrhea occurrence (p>0.05).

Discussion

The rehydration solutions evaluated in this trial had in their composition sodium bicarbonate (Common electrolytes solution and Common electrolytes solution + Aminogut®) or sodium acetate (Glutellac®). These alkalinizing components are the most commonly used in this kind of rehydration solutions and although both of them are effective alkalinizing agents, they have some important differences (Sen *et al*., 2009). Sodium bicarbonate does not require metabolism to exert its effect unlike acetate, which have a faster metabolism rate in calves from the second week of life (Goodell *et al*., 2012). However, sodium acetate has the advantage of providing energy without alkalinizing the abomasal fluid or affect milk clotting, facilitating the absorption of water and Na in the small intestine being metabolized readily by peripheral tissues (Sen *et al*., 2009). The lack of differences on acid-base status, recovery time from diarrhea and calves' performance may be because the all rehydration solutions were hyperosmotic, with very similar values of osmolarity.

The occurrence of at least one episode of diarrhea in all calves of the present study may be a consequence of the decrease of the immunity acquired through the ingestion of colostrum in the first hours of life and the incipient production of antibodies by the calf, making the animal more vulnerable to this kind of metabolic disorders (Davis and Drackley, 1998). Fecal score was higher during second and third weeks (Fig. 1), coinciding with an increase in hematocrit of calves as a response to the water loss in feces (Ravary-Plumioën, 2009). Hematocrit values obtained are within the range of values published by Mohri and Eidi (2007) and never exceeded the value of 30% reported by Guzelbektes *et al*. (2007) as an indicator of moderate to severe dehydration.

Rehydration hypertonic solutions increase the plasma osmolarity, also increasing the content of sodium, taking water from intracellular space, which stimulate the brain receptors and stimulate the consumption of water and the release of antidiuretic hormone that reduces the urine volume (Thornton, 2010). According to Wenge *et al*. (2014), when a calf has free access to water, its consumption is approximately one liter per day. This value is similar to the voluntary consumption of water presented by animals rehydrated with Common electrolytes solution and Common electrolytes solution + Aminogut®; however, is lower when compared with the water volume ingested by calves of treatment Glutellac®, which voluntarily consume around 2.5 L/d. However, because calves on Common electrolytes solutions were hand fed 1L of solution, twice a day when diarrhea occurred, total water intake was higher for these calves.

The lower calves' performance in this research in relation to that obtained by Hill *et al*. (2010) may be related to the volume fed and composition of the milk replacer used and to animal's breed, because crossbreed animals have different performance as compared to Holstein. Analysis of milk replacer composition (Table 1) reported values of crude protein and crude fat according the values suggested by NRC (2001). The value of NDF was higher than 0.5%, suggested by Davis and Drackley (1998) as indicative of the presence of vegetable protein in the formula used. This could indicate that it contained large proportion of vegetable protein and due to the lack of development of enzymatic apparatus in calves in this first stage of life, there is a poor digestion of non-milk source protein. Constable *et al*. (2001) obtained similar results to those of this study in relation to body weight. Body weight was similar among rehydration solutions, with a decrease between the first and the second week of life, which coincides with the period of greater fecal scores, which means higher occurrence of diarrhea (Fig. 1).

The results regarding feed intake (Table 3) were expected since the volume of liquid diet provided remained constant throughout the experimental period, resulting in an increment of consumption of solid diet, as the requirements of the growing animal increase. However, these values are still below those reported for calves in the eighth week of life by Quigley (1996). During the second week of life, there was a decrease in the ingestion of milk replacer for all treatments as a response of the higher occurrence of diarrhea, which can lead to rejection of liquid diet by the animals.

During the occurrence of diarrhea and the administration of different oral solutions, rectal temperature, respiratory frequency per minute (rpm) and heart beat per minute (bpm) were found within the range of values considered as normal (38.0-39.5°C, 15 to 40 rpm and 60 to 120 bpm, respectively) according to El-Sheikh *et al*. (2012).

During the pre-weaning period, blood metabolites were within normal values range for calves fed milk replacer and concentrate ad libitum and suggest a metabolism shift from the pre-ruminant to functional ruminant condition. Total serum protein, albumin and glucose concentration agrees with that reported by Mohri and Eidi (2007). Plasma urea, that may be affected by the diet and liver and renal function and showed a differential behavior in relation to age ($p<0.05$) agreeing with Benesi *et al*. (2005). Creatinine is also affected by animals' age and by the increase of renal function, similar to that obtained in the present study. Because there was no differences on days with diarrhea or animals performance, blood metabolites were also not different among rehydration solutions, with effects observed as function of age and increased starter intake leading to rumen development.

Blood gas analysis allows assessment of the severity of diarrhea and monitoring response to treatment through the values of blood pH, concentration of electrolytes and other substances associated to the acid-base balance of the organism (Sayers *et al*., 2016). According with these authors it is necessary to establish a cut-off pH value below which it would be necessary to treat with rehydration therapies the animals (Sayers *et al*., 2016). In this sense some authors propose for newborn calves a pH cut-off of 7.20 and 7.31 (Bleul *et al*., 2007) or 7.36 (Sayers *et al*., 2016) for older animals. Calves in this study had the first episode of diarrhea mainly at the second and third week of life and pH values for the three treatments were above those suggested by Bleul *et al*. (2007); however, those animals rehydrated with Common electrolytes solution had pH slightly below the cut-off suggested by Sayers *et al*. (2016) that may indicate mild acidosis. On the other hand, the pH in the present study had a tendency to increase from de first to the second day, but always within the range of normality reported by Bellino *et al*. (2012) and above the value proposed by Sayers *et al*. (2016).

Another tool to evaluate the acid-base status is the content of total carbon dioxide ($TCO_2 = HCO_3 + CO_2$) of the animal, being considered normal values between 25.6 to 33.4 mEq/L, lower values indicating acidosis and higher values alkalosis (Constable, 2014). All animals had concentrations within 25.6 and 33.4 mEq/L, in normal acid-base status (Table 4). There were no differences between treatments on in plasma HCO_3 content, as expected, since Sen *et al*. (2009) explain that when comparing solutions of similar osmolality, those that contains sodium bicarbonate in their composition are more effective in expanding plasma volume than solutions based in sodium acetate, in the short term. This may have been because solutions containing bicarbonate increases rapidly CO_2 plasma concentration, while solutions containing acetate, because it is slowly oxidized within the mitochondria, reach the levels of CO_2 produced with bicarbonate solutions after 60 to 90 min since rehydration solution has been administrated (Sen *et al*., 2009). So because the blood samples in this experiment were taken two hours after morning feeding, this allowed the metabolization of acetate in the mitochondria, reaching CO_2 levels similar to those produced with bicarbonate solutions.

The concentrations of sodium, chloride, potassium and sodium bicarbonate and excess of base (BE ecf), electrolytes responsible to maintain osmolarity (Freitas *et al*., 2010), have remained in normal values according to the literature regardless of the rehydration solution (Freitas *et al*., 2010). In the present study, BE (ecf) shows values within the range proposed by Bellino *et al*. (2012) as indicators of healthy status (3 to 5 mmol/L), increasing from the first to the second day of treatment without difference among treatments, agreeing with the results obtained by Sayers *et al*. (2016). Values of anion gap within the range of healthy calves (8.9-15.0 mmol/L) corroborates the BE (ecf) data, indicating that calves were not in metabolic acidosis (Guzelbektes *et al*., 2007). During occurrence of diarrhea blood urea concentrations were lower than those reported in the literature (Gregory *et al*., 2004), which may indicate hyperhydration for all calves due to the high water intake (Cunningham, 2004). However, plasma glucose concentration was within the normal range (Demigné *et al*., 1980), but was higher for the Glutellac® rehydrated calves ($p<0.05$; Table 4), probably because of its higher glucose content and because it was fed together with the milk replacer, two hours before blood sampling. Bellino *et al*. (2012) reported that animals with diarrhea and dehydration had higher hematocrit and hemoglobin levels than animals with diarrhea, but hydrated properly, being these values greater than those observed in the present study and without differences between treatments.

Conclusion

The rehydration therapies evaluated did not affect performance or blood parameters that indicate metabolic acidosis in animals affected by diarrhea, perhaps due to the similarity in osmolarity. However, the possibility that offers preparing the oral rehydration solution together with the liquid feeding, with free water access, makes this rehydration therapy an interesting alternative in the treatment of diarrheic calves, reducing the labor costs.

Acknowledgement

The authors would like to acknowledge the continued support received from Escola Superior de Agricultura "Luiz de Queiroz". The authors also appreciate the technical help provided by the co-workers.

Funding Information

The authors wish to express their appreciation for the financial support provided by the Sao Paulo Research Foundation (Fapesp) and by the Coordination for the Improvement of Higher Education Personnel (CAPES) as a scholarship.

Author's Contributions

Evangelina Miqueo: Study conception and design, conduction of the experiment, contribution to data interpretation and writing of the manuscript.

Jackeline Thais da Silva, Fernanda Lavínia Moura Silva, Nathalia Britto Rocha, Thais Manzoni Torrezan and Giovana S. Slanzon: Study conception and design and conduction of the experiment.

Carla Maris Machado Bittar: Contributed to the conception and design of the study, data interpretation and writing of the manuscript, critical revision of the manuscript and final approval.

Ethics

This article is original and contains unpublished material. The corresponding author confirms that all of the other authors have read and approved the manuscript and no ethical issues involved.

Disclosure Statement

The authors warrant that there are no conflicts of interests among authors and between authors and other people, institutions or organizations.

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