How to Use the Routine Serum Biochemical Profile to Understand and Interpret Acid-Base Disorders in the Horse

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Modern equine serum biochemical profile laboratory reports include all of the important electrolytes, total carbon dioxide (tCO_2), and total protein concentrations. Using case material it is demonstrated that this information is sufficient to provide an accurate understanding of acid-base balance in many circumstances without actually measuring pH and pCO_2 directly. The Gamble gram is simple to construct and assists in proportionally visualizing the electrolyte, anion-gap, and protein abnormalities which result in acid-base imbalances. Author's addresses: Department of Clinical Studies, Ontario Veterinary College, University of Guelph, Guelph, Ontario Canada N1G 2W1 (Stämpfli); Department of Medicine and Epidemiology, School of Veterinary Medicine, University of California—Davis, Davis, California 95616, USA (Carlson).

1. Introduction

Blood gas analysis has proven to be a useful adjunct tool for the assessment of metabolic and acid-base imbalances in the critically ill patient. Access to blood gas machines may not be routinely available to the practitioner and is often reserved for referral clinics and veterinary teaching hospitals. Traditional acid-base interpretation has relied heavily on the Henderson Hasselbalch equation and has largely ignored the importance of relative electrolyte concentration and protein concentration in the determination of acid-base balance and anion gap assessment. Routine serum biochemical profile should include measurements of all major electrolytes and serum protein. For the purposes of this discussion, three electrolytes (Na, K, Cl) are extremely important because they are present in high concentrations, they are completely dissociated in aqueous solution, and their relative concentration is one of the principal determinants of acid-base balance. More recently, biochemical profiles have also included tCO_2, which provides a close approximation of bicarbonate concentrations in blood and serum.

The hypothesis of this article is that the serum biochemical profile can be useful for the detection of acid-base disorders in horses and that the construction of a simple Gamble gram can be used to provide a better understanding of the various factors that contribute to acid-base balance in a clinical patient. The electrolytes Na, Cl, and K, as well as the serum proteins, together with the pCO_2 are the determining factors in acid base disorders.[1] Understanding the interrelationships between electrolytes, proteins, and acid-base balance can be very helpful in patient assessment as well as therapeutic decision making. In this article we would like to introduce the concept of Gamble gram and illustrate

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with selected cases the application of biochemical profiles in the evaluation of nonrespiratory acid base disorders in the horse.

2. Material and Methods

Concept of Gamble gram

Blood and its components are aqueous solutions, and the laws of chemistry apply to such solutions.[1] One of the key laws used to develop the Gamble gram is the law of electroneutrality.[2] Simply stated, the law of electroneutrality shows that the sum of the cations must equal the sum of the anions at all times in aqueous solutions. For serum samples at physiologic conditions this means that $([Na^+] + [K^+]) = [(Cl^-] + [A^-] + [HCO_3^-])$. There are, in addition, unmeasured strong cations (calcium and magnesium) and anions (phosphates, sulfates, and lactate) but they usually balance each other out unless lactate is involved. A^- is the negative charge attributed to the serum proteins and can be easily calculated once the serum protein concentration has been determined (conversion factors used: unit gram protein per decilitre (g/dl) is 1.75 mEq × this number or unit gram protein per liter (g/l) is 0.175 mEq × grams; the final unit concentration is mEq/l).[1,3] Under normal physiologic conditions the negative charge on the serum proteins is actually the major component of the traditional anion gap (unmeasured anions) calculated as $(Na^+ + K^+) - (Cl^- + HCO_3^-)$. The bicarbonate concentration is available if the total carbon dioxide (tCO_2) is reported in the serum biochemical profile, because bicarbonate accounts for 95% of tCO_2. While this estimate of bicarbonate can be useful it is not necessarily needed in the following approach to estimating metabolic acid-base balance. Table 1 depicts the parameters of a normal serum biochemical profile and includes the electrolytes sodium, potassium, chloride, lactate, tCO_2, anion gap, and A^- (the negative charge on serum proteins).

Table 1. Relevant Parameters for Acid-Base Interpretation of a Normal Biochemical Serum Profile

Serum Parameter		Reference Values
Sodium (mmol/l)	138	132–144
Potassium (mmol/l)	4.0	2.9–4.5
Chloride (mmol/l)	100	96–104
SID (mEq/l)	42	38–44
Total Protein (g/l)	71	54–75
A^- (mEq/l)(0.175 × [Total Protein])	12.5	11–16
Anion Gap (mEq/l)	14	12–18
tCO_2 (mmol/l)	28	24–32
Lactate (mmol/l)	1.5	0–2

SID = Strong ion difference $(Na^+ + K^+) - (Cl^- + Lactate^-)$ or $(Na^+ + K^+) - (Cl^-)$ as lactate is not often measured; A^- = Anion load of total protein in serum.

In this graph mEq/l is equal to mmol/l because all ions present are monovalent.

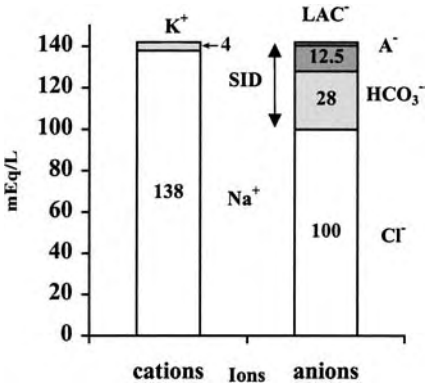

Fig. 1. Normal Gamble gram of the healthy adult STB racehorse. SID = Strong ion difference; LAC^- = Lactate; A^- = the anion gap and represents the negative charge on the serum proteins. (Total Protein concentration = 71 g/l; 71 × 0.175 mEq/l = 12.425 mEq/l; pH = 7.42.[2] All units are in mEq/l (= mmol/L for all substances shown here).

The Gamble gram is a method to visually and quantitatively demonstrate the balance or, in some cases imbalance, between cations and anions in an aqueous sample (e.g., serum, blood). Figure 1 shows the basic normal Gamble gram of an adult Standardbred racehorse for which electrolyte and bicarbonate concentrations have been determined. In this figure the two principal positively charged cations Na^+ and K^+ are placed in the left column with the concentrations indicated proportionally. The negatively charged anions, Cl^-, (HCO_3^-), lactate$^-$, and A^- are represented proportionally in the right column. As can be clearly seen, the sum of the positive ions (Na^+ and K^+) is equal to the sum of the negative ions (Cl^-, (HCO_3^-), A^- and lactate$^-$). Lactate is not frequently measured so the negative ions usually included in the Gamble gram are Cl^-, HCO_3^-, and A^-. It is shown here for completeness.

An additional entity that may be included as an indicator of acid-base balance is the strong ion difference (SID). The SID is defined as the difference of sodium plus potassium minus chloride plus lactate and should be within the range of 38–44 mEq/l (milliequivalent is equal to mmol/l in aqueous solution, which contain univalent ions) (Figure 1). It can be mathematically shown that SID, like the partial pressure of carbon dioxide (pCO_2) and total protein concentration, is a major determinant of acid base balance in blood, plasma, and serum. From this Gamble gram in Figure 1 it is also evident that the major component of the so-called anion gap is the anionic contribution of the largely negatively charged serum proteins (12.5 mEq/l). The minor component of the anion gap is the lactate (1.5 mEq/l). Figure 2 shows a Gamble gram of a horse in which electrolyte concentrations are within normal limits but the protein concentration, which is very low (25 g/l) as might be found in a case of

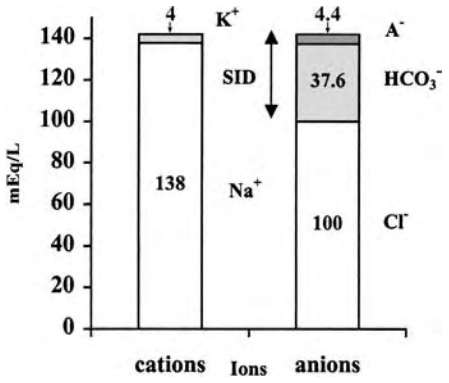

Fig. 2. Gamble gram of a horse with normal electrolytes but severe hypoproteinemia. A^- is the anion gap and represents the negative charge on the serum proteins. (Total protein concentration is 25 g/l; 25 × 0.175 mEq/l = 4.4 mEq/l; pH = 7.55, pCO_2 = 40 mmHg). This hypoproteinemia is causing a metabolic alkalosis. All units are in mEq/l.

protein losing enteropathy or colitis. Because the protein concentration is markedly reduced, the anionic contribution of the proteins is commensurately reduced as well (4.4 mEq/l). Because of the decreased protein concentration, the anion gap is also markedly reduced from a normal value. Blood gas analysis will reveal hypoproteinemic alkalosis in which the bicarbonate concentration is increased. It should be pointed out here that the relationship of decrease of proteins and extend of alkalosis is not a linear relationship but a *complex* polynomial mathematical relationship.

The following are useful relationships to keep in mind.

Electrical Neutrality in Serum or Plasma

Positive Cations = Negative Anions

$$Na^+ + K^+ = (Cl^- + HCO_3^- + \text{Anion Gap})$$

$$\textbf{Anion Gap} = (Na^+ + K^+) - (Cl^- + HCO_3^-)$$

The anion gap is really the difference between the concentrations of the unmeasured cations (Ca and Mg) and the unmeasured anions (protein [A^-], phosphate, sulfate, lactate, etc.).

SID = $(Na^+ + K^+) - (Cl^- + \text{Lactate}^-)$, or when Lactate is not measured **SID = $(Na^+ + K^+) - (Cl^-)$.** **A^-** = (protein g/l) × (0.175 mEq) SI units or **A^-** = (protein g/dl) × 1.75 mEq in present units. Measure unit is mEq/L.

3. Results

To further illustrate the detailed approach using the serum biochemical profile as indicators of acid-base balance we will now discuss 3 cases.

Case 1

An 8-year-old Arabian gelding was pulled after the midpoint of a 100-mile endurance ride for failure to recover. The horse was depressed and dehydrated with an elevated heart rate and respiratory rate. It was an exceptionally hot day and heavy sweat losses were evident. The rider reported that the horse drank fairly well, but they had not administered supplemental electrolytes to the horse.

Table 2. Serum Biochemical Profile of Case 1

Serum Parameter		Reference Values
Sodium (mmol/l)	135	132–144
Potassium (mmol/l)	3.6	2.9–4.5
Chloride (mmol/l)	72	96–104
SID (mEq/l)	66.6	38–44
Total Protein (g/l)	87	54–75
A^- (mEq/l)(0.175 × [Total Protein])	15.23	11–16
Anion Gap (mEq/l)	18.35	12–18

Assessment: Hyponatremia and profound hypochloremia with marked increase in SID. There is also a moderate increase in the protein concentration and therefore a high normal anion gap as represented by A^-.

Table 2 shows portion of the serum biochemical profile of this horse at the time he was eliminated from the ride. Figure 3 shows the Gamble gram based on these data. The observed electrolyte alterations are not uncommon for non-supplemented endurance horses and are mainly due to heavy sweat losses and include a mild hyponatremia and marked hypochloremia. The SID in this horse was markedly increased at 66.6 mEq/l (normal 38–44 mEq/l). Even though the tCO_2 was not reported on this profile, it is obvious that the bicarbonate concentration must be markedly increased and the horse probably has a marked metabolic alkalosis. The protein concentration is mildly increased and this has slightly increased our anion gap to a high normal value. The abnormalities observed on the Gamble gram provide a visual illustration of the relationship between the electrolyte imbalance and the associated acid-base disturbance. Here we

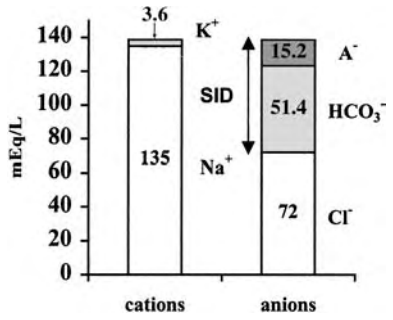

Fig. 3. Case 1 Gamble gram of serum biochemical profile in Table 2. There are massive electrolyte abnormalities with a severe hypochloremia. The bicarbonate concentration is very high. Calculated pH (with assumed pCO_2 = 40 mm Hg) is 7.70, which is exceedingly high. A^- represents the anionic load on the protein fraction of this serum and is equal to the traditional anion gap. In this case the anion gap is high normal.

have a metabolic alkalosis caused by the profound hypochloremia. Viewing information in this way provides a clear indication of the fluid of choice (isotonic to hypertonic saline) for correction of the electrolyte and fluid imbalances. Assuming that this case was clinically about 10% dehydrated, the treatment to be initiated would include parenteral rehydration over a 4–6-h period. The assumed body weight is 410 kg (902 pounds) with calculated total fluid deficit of 40 l. Therefore, theoretically 40 l of fluid would be needed. The electrolyte imbalance recognized to cause the metabolic derangement is a deficit of sodium and chloride and a hidden deficit of lost intracellular potassium. The parenteral fluid of choice therefore would be 40 l of 0.9% NaCl over a 4–6 h period. Another approach would include 1.5 l parenteral hypertonic 7.2% NaCl (a maximum of 3 ml per kg bodyweight) followed by 20 l of isotonic saline or lactated Ringers solution and enteral fluids given via nasogastric tube over a period of 1–2 hours. The potassium deficit will be corrected over a period of days with the consumption of potassium-rich hay. In summary, the acid-base imbalance in this endurance horse is a marked hypochloremic alkalosis with a very mild superimposed hyperproteinemic acidosis.

Case 2

Presented is a 7-year-old Quarter Horse gelding with a history of unthrifty condition, depression, and anorexia of several days duration. The major clinical problems included depression, decreased borborygmi, weight loss, and complete anorexia. Table 3 shows electrolyte and total protein concentrations as well as tCO_2 and anion gap. The major abnormalities observed are hyperchloremia with a decreased tCO_2 and normal anion gap resulting in a severe hyperchloremic metabolic acidosis. Electroneutrality display on the Gamble gram reveals a major imbalance between sodium and chloride concentration and a minor elevation of an unmeasured anion (4.8 mEq/l) (Figure 4). The SID was markedly decreased, indicating a probable metabolic acidosis. Subsequent blood gas analysis confirmed the acidosis: pH = 7.12, pCO_2 = 28.1 mm Hg, bicarbonate 10.5 mEq/l, base balance −14.3 mEq/l. The BUN and creatinine were within normal limits and urine pH was 8.5. This case was diagnosed as having a renal tubular acidosis without major dehydration as discussed in a recent report.[4]

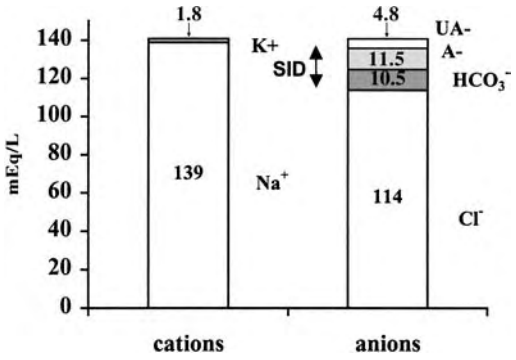

Fig. 4. Gamble gram of Case 2 using values in Table 3. Severe hyperchloremic metabolic acidosis with moderate elevation of unmeasured anions and slight decrease of normal serum protein anion load. (Calculated pH based on above values is 7.19; measured pH was 7.12).

Treatment of this horse involved the administration of large amounts of sodium bicarbonate parenterally and enterally. Initially, 6 l of 5% $NaHCO_3$ was given intravenously to replace the estimated deficit (444 kg × 0.40 l/kg × 14 mEq/l) or 2400 mEq. This partially corrected the deficit and an additional 6 l of 5% $NaHCO_3$ was necessary and it was given over the next 24 h. After the initial intravenous treatment, enteral supplementation with 100 g of $NaHCO_3$ was given twice daily via nasogastric tube. As the acid-base balance returned toward normal a significant hypokalemia developed and potassium supplementation was initiated intravenously and enterally. With the correction of acid-base balance the horse regained a normal attitude and appetite. The horse was maintained on gradually decreasing doses of $NaHCO_3$

Table 3. Serum Biochemical Profile of Case 2

Serum Parameter		Reference Values
Sodium (mmol/l)	139	132–144
Potassium (mmol/l)	1.8	2.9–4.5
Chloride (mmol/l)	114	96–102
SID (mEq/l)	26.8	38–44
Total Protein (g/l)	66	54–75
A^- (mEq/l)(0.175 × [Total Protein])	11.5	11–16
tCO_2 (mmol/l)	10.5	24–32
Anion Gap (mEq/l)	16.3	12–18

Interpretation: Severe hyperchloremic metabolic acidosis with decrease of tCO_2 and decrease of normal serum protein anion load. A^- minus anion gap equals 4.8 mEq/l indicating presence of an unmeasured anion.

Table 4. Serum Biochemical Profile of Case 3

Serum Parameter		Reference Values
Sodium (mmol/l)	139	132–144
Potassium (mmol/l)	3.16	2.9–4.5
Chloride (mmol/l)	109.8	96–104
SID (mEq/l)	32.3	38–44
Total Protein (g/l)	57	54–75
A^- (mEq/l)(0.175 × [Total Protein])	10	11–16
Total-CO_2 mmol/L	12	24–32
Anion Gap (mEq/l)	20.36	12–18

Interpretation: Hyperchloremia with decreased SID. Mild increase in anion gap and severe decrease in tCO_2.

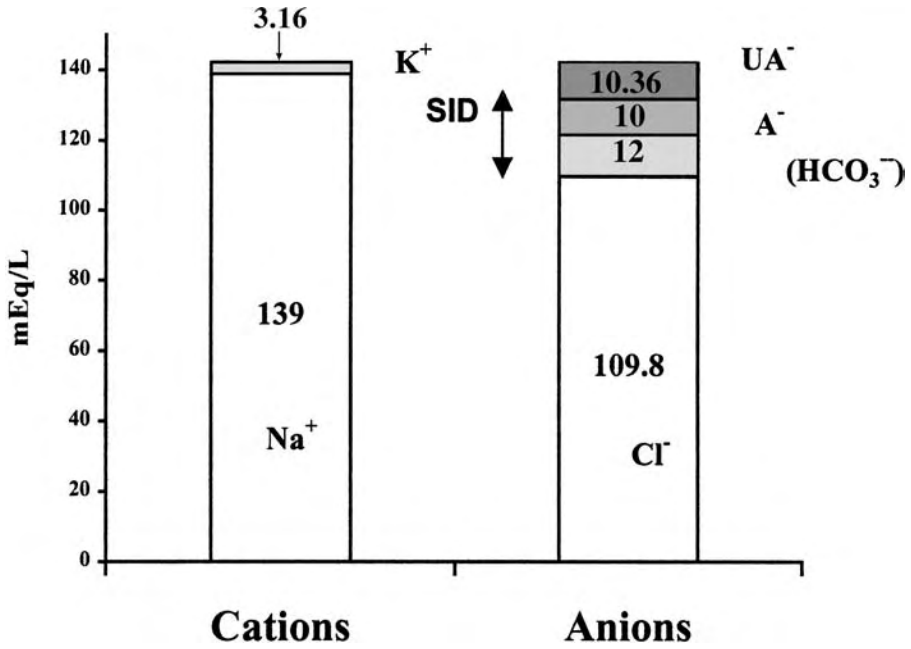

Fig. 5. Gamble gram of Case 3. Significant increase in unmeasured anion and significant decrease in bicarbonate concentrations. Metabolic acidosis with mild increase in anion gap.

given enterally mixed with its feed for several months.

Case 3

A 3-day-old Standardbred colt was delivered normally and suckled well for 12 h, but then broke with profuse watery diarrhea. The diarrhea continued for the next 2 days. He was treated with broad-spectrum antibiotics and NSAIDs. He began to deteriorate and on presentation he showed progressive dehydration, depression, and reluctance to stand. On presentation the foal was severely dehydrated with a profuse yellow watery diarrhea, tachycardia, and depression. The working diagnosis was severe dehydration with possible metabolic acidosis, septicemia, and gastric ulceration. Table 4 and Figure 5 depict the serum biochemical profile and the Gamble gram, respectively. The tCO_2 is very low indicating that the [HCO_3^-] concentration is very low as well signalling profound metabolic acidosis. The Gamble gram shows that there is quite a marked increase in unmeasured anions of 10.36 mEq/l, which is most likely to be lactate. The anion gap was only marginally elevated because the protein concentration was low and therefore its anionic load is low (10 mEq/l) despite marked clinical dehydration. SID is markedly reduced by about one fourth indicating metabolic acidosis. The clinical dehydration was estimated at 10% and the bodyweight was 50 Kg (120 lbs). The body fluid deficit calculated was 5 liters and based on the low HCO_3^- concentration on the Gamble gram and the low SID a metabolic hyperchloremic acidosis was diagnosed. The $NaHCO_3$ deficit in mEq was 0.5 ($NaHCO_3$ space in a foal) $\times$ 12 (base excess as calculated: 24 HCO_3 − 11.9 HCO_3 on the Gamble gram) $\times$ 50 (bodyweight) = 300 mEq of $NaHCO_3$. This may be directly added to 5 l of lactated ringers solution and infused over a 4–6-h period. In summary, we have a marked metabolic hyperchloremic acidosis with a moderately increased unmeasured anion present.

4. Conclusion

In conclusion these case scenarios demonstrate that the serum electrolytes and serum total protein concentrations combined with Gamble gram is a useful method to understand underlying acid-base problems without direct measurements of pH and pCO_2.

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