Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.
The central role of chloride in the metabolic acid–base changes in canine parvoviral enteritis

Richard K. Burchell *, Johan P. Schoeman, Andrew L. Leisewitz

Department of Companion Animal Clinical Studies, Faculty of Veterinary Science, University of Pretoria, Onderstepoort 0110, South Africa

**ABSTRACT**

The acid–base disturbances in canine parvoviral (CPV) enteritis are not well described. In addition, the mechanisms causing these perturbations have not been fully elucidated. The purpose of the present study was to assess acid–base changes in puppies suffering from CPV enteritis, using a modified strong ion model (SIM). The hypothesis of the study was that severe acid–base disturbances would be present and that the SIM would provide insights into pathological mechanisms, which have not been fully appreciated by the Henderson–Hasselbalch model.

The study analysed retrospective data, obtained from 42 puppies with confirmed CPV enteritis and 10 healthy control dogs. The CPV-enteritis group had been allocated a clinical score, to allow classification of the data according to clinical severity. The effects of changes in free water, chloride, L-lactate, albumin and phosphate were calculated, using a modification of the base excess algorithm. When the data were summated for each patient, and correlated to each individual component, the most important contributor to the metabolic acid–base changes, according to the SIM, was chloride ($P < 0.001$). Severely-affected animals tended to demonstrate hypochloraemic alkalosis, whereas mildly-affected puppies had a hyperchlo- raemic acidosis ($P = 0.007$). In conclusion, the acid–base disturbances in CPV enteritis are multifactorial and complex, with the SIM providing information in terms of the origin of these changes.

© 2014 Elsevier Ltd. All rights reserved.

Introduction

Assessment of acid–base status is frequently used in veterinary critical care cases, because it can be used to detect early physiological derangements, alerting clinicians to the possibility of decompensation, as well as providing treatment directives (Hopper, 2012). Traditionally, the Henderson–Hasselbalch (HH) technique has been used, when assessing plasma acid–base disturbances, which involves measuring two variables, namely carbon dioxide tension ($pCO_2$) and bicarbonate ($HCO_3^-$) concentration (Constable, 2000). A change in either of these will invoke a compensatory response in the other, to maintain a constant plasma pH (Constable, 2000). The reciprocity of this relationship may obfuscate interpretation of a mixed or non-compensated disorder, where it is unclear to what extent $pCO_2$ and $HCO_3^-$ have changed due to the primary disorder, or due to compensatory mechanisms (Constable, 2000; Sirker et al., 2002). Calculations based on the standard base excess (titratable acidity or alkalinity) and the anion gap have been used in conjunction with the HH model, in an attempt to abrogate this problem (Siggaard-Andersen et al., 1960; Siggaard-Andersen and Fogh-Andersen, 1995).

The strong ion model (SIM), also known as Stewart's strong ion model, is an alternative method, used in assessment of acid–base disturbances (Fencl and Leith, 1993; Gilfix et al., 1993; Whitehair et al., 1995; Constable, 2002; Wooten, 2004; Greenbaum and Nirmalan, 2005; Story and Kellum, 2005; de Morais and Constable, 2006; Morgan, 2009). The fundamental premise of the SIM is that the strong ion difference (SID) of plasma (difference between the sum of all strong cations and strong anions) is the most important determinant of the hydrogen ion activity in a system. In addition to $pCO_2$ and SID, the SIM considers a third variable, namely the sum of weak non-volatile organic acids, such as albumin and phosphate (denoted $A_{weak}$) (Fencl and Leith, 1993; Kellum, 2007). The SIM therefore provides rational explanations for acid–base disturbances that are not well understood in terms of the HH model, such as the effects of free water and plasma proteins (Kellum, 2008).

Notwithstanding the apparent advantage of the SIM, its application in medicine has met with some degree of opposition, as it violates the traditional dogma of the Arrhenius principle of acid–base chemistry (Kurtz et al., 2008). In spite of such criticism, the SIM has gained momentum in critical care medicine, mainly...
because it provides more information in terms of disturbances within the metabolic compartment (Constable, 2000; Sirker et al., 2002; Kellum, 2007).

Canine parvoviral (CPV) enteritis is characterised by severe vomiting and diarrhoea, often associated with a high mortality rate (Prittie, 2004; Goddard and Leisewitz, 2010; Schoeman et al., 2013). Medical therapy is implemented according to the severity of disease and includes fluid therapy, nutritional management and use of anti-emetic and/or antimicrobial drugs (Prittie, 2004).

There is, however, a paucity of information available regarding the acid–base disturbances in CPV enteritis, on which to practise evidence-based medicine.

In one study, arterial blood gas and venous blood electrolyte data were collected for 17 puppies affected with CPV enteritis (Heald et al., 1986). Blood pH was within normal limits in 59% of cases and, of the remaining dogs, six were alkalaeemic and one was acidaemic. In contrast, in the study by Rai and Nauriyal (1992) a significant acidaemia was demonstrated in 21 cases of CPV enteritis. Furthermore, a decrease in actual and standard HC0₃⁻ and base excess was observed. In a later study, plasma pH was consistently increased in dogs affected with CPV enteritis, compared to the healthy dogs, but HC0₃⁻ was consistently decreased and the pH increase was deemed to be due to compensation, in the presence of decreased HC0₃⁻ concentration (Nappert et al., 2002).

Of particular interest in the study by Nappert et al. (2002) is the fact that specific criteria were present that would allow a classification of metabolic acidosis (decreased HC0₃⁻ and increased l-lactate production), but the blood pH was in fact higher than normal (i.e. alkalaeaemia). Within this deviation of HC0₃⁻, we predict that the SIM might unmask mixed acid–base disturbances, which would be obscured according to the HH model. Thus, the purpose of the present study was to utilize the SIM to dissect metabolic homeostasis in dogs affected with CPV enteritis, to provide further insights into the pathogenesis of the acid–base disturbances present.

Materials and methods

Sample population

Clinical and laboratory data, collected from 42 unvaccinated puppies affected with CPV enteritis and 10 age-matched control dogs were analysed using a modified strong ion approach, based on the base excess algorithm (Fencl and Leith, 1993; Hopper and Haskins, 2008). CPV had been confirmed in the clinical cases by electron microscopy, and confounding infection with rotavirus, coronavirus, Angulostoma spp. and Giardia spp. had been excluded.

Affected dogs were recruited as part of a project to assess the utility of biomarkers in assessment of severity and survival in CPV enteritis (Schoeman et al., 2007, 2013; Schoeman and Herrtage, 2008). All data were collected on the day of admission, before any therapy had been initiated. Diagnostic testing was undertaken by the Department of Clinical Pathology, Faculty of Veterinary Science, University of Pretoria reference laboratory. The control population consisted of healthy vaccinated dogs under 6 months of age, which were presented for routine clinical examination. The study was approved by the Animal Care and Ethics committee (V076/05, 20/12/2005).

A clinical score was assigned to each CPV-affected dog on admission, which stratified the population according to the severity of clinical signs (mild, moderate or severe). The clinical score was based on appetite, habitus, vomiting, diarrhoea and mucous membrane colour (see Appendix A: Supplementary Table 1). This clinical scoring system was designed for a PhD thesis (Schoeman, 2008) and, although not formally validated, has been used extensively in the Onderstepoort Veterinary Academic Hospital. All clinical scoring was performed by a single observer (JS). Patients with scores <9 were classified as severely affected, scores between 9 and 16 were considered moderately affected and scores >16 were classified as mildly affected.

Data analysis

Comprehensive records were obtained for each animal, containing a complete serum biochemistry profile, with the exception of chloride and serum inorganic phosphate. Serum stored at –70 °C was analysed for these latter electrolytes, using a Cobas Integra 400 Plus (Roche) analyser. Since no blood gas measurements had been taken, it was not possible to assess pH, base excess or bicarbonate. The calculation of the contribution of each of the components to the base excess (according to the SImp) is shown in Appendix A: Supplementary Table 2. The data for each category were compared to the control group using a commercial statistics package (Medcalc, version 12.7.2). D’Agostino–Pearson test for normality was performed for all data sets. The Mann–Whitney U test was used to compare medians, and Spearman’s rank correlation was used to assess the relationship between different variables.

To assess the relative contribution of each component to the overall metabolic acid–base changes, each of the components was summed and the sum obtained compared to each individual component by means of a Spearman’s rank correlation. Each of the variables used in the quantification of the metabolic acid–base compartment were compared, according to clinical disease severity. Using the principle of the strong ion approach, the free water, chloride and l-lactate effect were summed and, if a negative value was obtained (within a –2.0 to 2.0 mEq/L tolerance range), a strong ion acidosis was diagnosed; whereas a strong ion alkalosis was diagnosed if the value was positive.

The albumin and phosphate effects were summated to yield the Atot and the values interpreted as for the strong ion compartment. This calculation was performed for each CPV-affected dog and a diagnosis for the metabolic compartment was assigned as follows: strong ion acidosis, A_tox acidosis (designated A); strong ion acidosis, A_tox alkalosis (designated B); strong ion alkalosis, A_tox acidosis (designated C) and strong ion alkalosis, A_tox alkalosis (designated D). The sum of all the effects was taken to represent the base excess, and significantly negative values interpreted as a metabolic acidosis and positive values as a metabolic alkalosis. When the value of the sum was within the tolerance range, but significant changes were present in the constituents, a mixed neutralising disorder was diagnosed. A final classification for the metabolic compartment could then be assigned as follows: metabolic acidosis/alkalosis (or neutralising), characterised by strong ion acidosis/alkalosis, and A_tox acidosis/alkalosis. The metabolic acid–base status of the dogs was displayed visually, using Venn diagrams as previously described by Vu et al. (2010).

The base excess algorithm suggested by Hopper and Haskins (2008), based on the original work of Fencl and Leith (1993), has not been validated in dogs, and is based on the assumption that albumin is the most important contributor to A_tox. A simplified technique (referred to subsequently as the simplified model, SM), using experimentally-determined values, validated for dogs was used (Constable and Stamphilli, 2005) and compared to the traditional base excess algorithm. Briefly, the SID₄ was calculated from four major strong ions (Na⁺ – K⁺ – Cl⁻ – l-lactate) for both the CPV-affected and control groups. The A_tox was estimated from albumin, based on the determination of a net protein charge of 0.42 mEq/g in albumin dogs, yielding a value of 15.8 mEq/L for normal dogs (Constable and Stamphilli, 2005). The net protein charge was also determined using total protein (0.25 mEq/g of total protein). This was determined for both groups and compared to the experimentally-determined normal value of 15.8 mEq/L (Constable and Stamphilli, 2005). SID₄ and the A_tox derived from albumin and total protein (TP) in the CPV-affected and control groups were compared using the Student’s t test.

Finally, using the simplified model, a metabolic acid–base classification was assigned to each case, by comparing the CPV-affected group values for SID₄ and A_tox (calculated from albumin and TP) to the experimentally-validated values for these variables (values taken from Constable and Stamphilli, 2005). In addition, the SID₄ and A_tox obtained from the SM were compared to those obtained for the control dog samples. This enabled assessment of the validity of comparing samples from CPV-affected dogs to experimentally-determined values. Diagnosis were then assigned to the categories A–D as described previously, with the outcomes of this simplified method and the base excess algorithm compared using an inter-rater agreement plot.

Results

According to the SIM, 20 of 42 patients in the CPV-affected group were considered to have a metabolic acidosis, 10/42 had a metabolic alkalosis and in 12/42 patients the overall effect was neutralizing. Of the 20 patients affected with metabolic acidosis, all had a SID₄ acidosis and within this group, 19 had a concurrent A_tox alkalosis and one had a mild A_tox acidosis, due to mild hyperphosphataemia (Fig. 1a). Of the individuals with metabolic alkalosis, 9/10 had a SID₄ alkalosis and 1/10 had a SID₄ acidosis. All 10 patients had a concurrent A_tox alkalosis (Fig. 1b). Within the neutralizing group, 8/12 had a SID₄ acidosis, with all eight of these having an A_tox alkalosis. The remaining four dogs had a SID₄ alkalosis and, within this group, two had an A_tox alkalosis, with the remaining two having an A_tox acidosis (Fig. 1c).
None of the other variables correlated significantly with the sum of all the effects. Furthermore, when each of the SIM variables were statistically compared, according to clinical disease severity, a significant difference was noted within the chloride effect (Fig. 3), where mildly-affected puppies tended to have a hyperchloremic acidosis and severely-affected puppies had a hypochloremic alkalosis. According to these findings, and those of the Spearman’s rank correlation, knowledge of the chloride effect most consistently predicted the outcome of the metabolic compartment in CPV-affected dogs according to the SIM.

According to the SM, the SiΔ of the CPV-affected group was 35 ± 3.0 mEq/L, compared to 39.2 ± 6 mEq/L for the control group and 39 mEq/L, experimentally determined by Constable and Stampfli (2005). The difference was significant for the CPV-affected group compared to the control group (P = 0.01). Arot of the CPV-affected group, using albumin alone, was 8.9 ± 2.9 mEq/L, compared to 10.5 ± 1.5 mEq/L for the control dogs and 15.8 mEq/L (Constable and Stampfli, 2005). This difference between CPV-affected dogs and the controls was significant (P < 0.005).

When the diagnoses were categorised (A–D), using the simplified method (based on comparison to the experimental values), and the outcomes using this method compared to the diagnosis using the base excess algorithm, there was good agreement between the two models (kappa = 0.72). When Arot was calculated from total protein (Arot-tp), the value was 12.58 ± 2.3 mEq/L and 14.6 ± 1.8 mEq/L for the CPV-affected and control dogs, respectively (P = 0.006). When the diagnostic outcomes were performed using the Arot-tp instead of Arot-alb (compared to experimental values) and compared to the base excess algorithm, the diagnosis only changed in two cases (Arot normal), and there was still good agreement between the two models (kappa = 0.71).

**Discussion**

Both vomitus and diarrhoea are electrolyte-rich fluids and therefore the presence of plasma electrolyte disturbances was not surprising in dogs affected with CPV enteritis. Sodium was consistently low in the CPV group, indicating a relative free water excess. According to the SIM principles, a free water excess will invoke acidosis, partly due to a decrease in the strong ion difference (SID) (Kellum, 2007). In addition, an Arot alkalosis may occur with a free water excess, due to decreased plasma protein concentration, which may offset the magnitude of SID acidosis, as is the case in dilutional acidosis (Constable, 2003). In the case of CPV enteritis, this mechanism could be even more complex, due to dehydration and concurrent protein loss through the gut, resulting in diverse outcomes in the plasma protein concentration and therefore the contribution of Arot. Conversely, a free water deficit would result in an increased SID alkalosis with a possible Arot acidosis, due to increased concentration of plasma proteins (Constable, 2003). In addition, an Arot alkalosis was also common due to significant albumin losses.

The findings of the present study gave insight into the complexity of the acid–base changes in CPV enteritis and potentially explained why minor changes in bicarbonate concentrations have been observed in previous studies, in the face of significant metabolic disturbances. When the Arot was estimated from TP (according to the SM), the value was consistently higher; however, the value was still statistically lower than the Arot (estimated from the TP) in the control group. This finding confirms the assertion that plasma globulin proteins play a more significant role in the determination of Arot in dogs compared to humans (Constable and Stampfli, 2005). Therefore, the base excess algorithm using albumin, overestimated the contribution of Arot in CPV enteritis. Many of the control dogs had significant Arot alkalosis, according

![Fig. 1. Venn diagrams characterising the CPV-affected dogs with (a) metabolic acidosis, (b) metabolic alkalosis or (c) neutralising effects within the acid–base compartment. The circles indicate the metabolic changes (SID or Arot) and the numbers indicate the number of animals within each group.](image)
Table 1
Median and interquartile range (IQR) of serum electrolytes and the effects of free water, chloride, i-lactate, albumin and phosphate in puppies with CPV enteritis compared to healthy controls.

| Parameter      | CPV group (n = 42) | Control group (n = 10) | P-value |
|----------------|-------------------|------------------------|---------|
| Sodium (mMol/L) | 137               | 143                    | <0.001  |
| Potassium (mMol/L) | 4.29          | 4.73                   | 0.001   |
| Chloride (mMol/L) | 106            | 111                    | 0.001   |
| Corrected chloride (mMol/L) | 113       | 112                    | 0.58    |
| Albumin (g/L)   | 21                | 25                     | 0.01    |
| i-Lactate (mMol/L) | 2.43          | 2.7                    | 0.17    |
| Sum of effects (mEq/L) | -2.44      | -1.80                  | 0.21    |
| Free water effect (mEq/L) | -2.0        | -0.51                  | <0.0001 |
| Chloride effect (mEq/L) | -2.51      | -1.61                  | 0.5     |
| i-Lactate effect (mEq/L) | -2.5        | -2.10                  | 0.22    |
| Albumin effect (mEq/L) | 3.64        | 2.0                    | 0.01    |
| Phosphate effect (mEq/L) | 0.07        | -0.69                  | 0.39    |

One limitation of this study was the absence of blood gas analysis, which precluded a direct comparison between the HH and the SIM model. Notwithstanding this constraint, valuable information can still be obtained from this study, regarding the pathophysiology of the acid–base disturbances present in dogs affected with CPV enteritis. A previous study showed a marked increase in i-lactate production, a significantly decreased base excess, with only a mild decrease in bicarbonate and significantly reduced carbon dioxide tension (Nappert et al., 2002). Bicarbonate values roughly approximated to those expected during compensation with a primary respiratory alkalosis, in the face of significantly increased i-lactate and β-hydroxybutyrate, which would be expected to result in a metabolic acidosis. These findings, compared with those of the present study, suggest the HH paradigm is too simplistic to explain the complex underlying metabolic acid–base changes in CPV enteritis.

When the sum (summated components of the base excess algorithm) was correlated with its constituents, the most significant relationship was with chloride (corrected). This was an interesting finding, since it would have been expected that a significant relationship would have been observed between the sum and free water changes, although this was not the case. Interestingly, the two most consistently deranged variables, namely sodium and albumin, showed the least significant relationship with the sum. This finding appears to emphasise the significance of chloride disturbances in the pathogenesis of acid–base changes in CPV enteritis, which is not appreciated by the HH model. Therefore, regardless of a consistent hyponatraemic acidosis and hypoalbuminaemic alkalosis, the direction and magnitude of the changes in chloride will likely determine the outcome of the sum, in most cases. The importance of chloride changes was further highlighted, since it was the only variable in which differences were observed, according to clinical severity. The more severely-affected puppies, according to clinical score, tended to have a hyperchloraemic alkalosis, whereas mildly-affected individuals tended to have a hyperchloremic acidosis. These changes might reflect differences in the severity of vomiting or the period of illness that had elapsed, before sampling. Further studies are needed to determine if chloride changes correlate with outcome and whether therapeutic chloride correction is warranted.

Finally, this study was able to demonstrate the utility of a simplified SIM technique, making use of standard plasma electrolyte and albumin analysis. When SID and A\textsubscript{tot} were calculated and compared to experimentally-established values, there was good agreement between the two models. This method, therefore, compared well with the base excess algorithm approach in reaching a diagnosis of strong ion and A\textsubscript{tot} changes. The base excess algorithm was to the base excess algorithm, although when the A\textsubscript{tot} was estimated using TP, the value was close to the normal value previously calculated (15.8 mEq/L) in the control dogs.

Fig. 2. Spearman’s rank correlation of the sum of effects and the chloride effect. The horizontal line on the graph indicates the neutral point of the sum, and the vertical line represents a neutral chloride effect. The change in the chloride effect is strongly correlated with the change of the sum, both of which consistently change in the same direction, either positive or negative (P < 0.001).

Fig. 3. Box plot comparing the chloride effect in mildly-affected (n = 10), moderately-affected (n = 21) and severely-affected (n = 11) dogs with CPV enteritis. Boxes indicate interquartile range (IQR), the solid horizontal lines represent the median, the whiskers 1.5 ± IQR and the open circles outliers. *P = 0.007 comparing severely-affected and mildly-affected dogs.
principally employed, due to its common use in determining acid-base status. Our results showed that the model was robust and that it compared well with data obtained using a model validated in dogs. Clinicians should recognise that in dogs the base excess algorithm using albumin rather than TP might overestimate the magnitude of $A_{tot}$ changes.

Conclusions

Application of the SIM for clinical assessment of the acid–base status in puppies affected with CPV enteritis, indicated that significant electrolyte and albumin disturbances are present, but that chloride is the most important variable in the pathogenesis of the acid–base disturbances.

Conflict of interest statement

None of the authors has any financial or personal relationship that could inappropriately influence or bias the content of the paper.

Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.tvjl.2014.01.017.

References

Constable, P.D., Stampfli, H., 2005. Experimental determination of net protein charge $A_{tot}$ and $K_a$ of non-volatile buffers in canine plasma. Journal of Veterinary Internal Medicine 19, 507–514.

Constable, P.D., 2002. Calculation of variables describing plasma non-volatile weak acids for use in the strong ion approach to acid–base balance in cattle. American Journal of Veterinary Research 63, 482–490.

Constable, P.D., 2003. Hyperchloric acidosis: The classic example of strong ion acidosis. Anaesthesia and Analgesia 96, 919–922.

Constable, P.D., 2000. Clinical assessment of acid–base status: Comparison of the Henderson–Hasselbalch and strong ion approaches. Veterinary Clinical Pathology 29, 115–128.

de Morais, H.A., Constable, P.D., 2006. Strong ion approach to acid–base disorders. In: Fluid, Electrolyte, and Acid–Base Disorders in Small Animal Practice, Third Ed. W.B. Saunders, Saint Louis, MO, USA, pp. 310–321.

Fencl, V., Leith, D.E., 1993. Stewart’s quantitative acid–base chemistry: Applications in biology and medicine. Respiration Physiology 91, 1–16.

Gilfix, B.M., Bique, M., Magder, S., 1993. A physical chemical approach to the analysis of acid–base balance in the clinical setting. Journal of Critical Care 8, 187–197.

Goddard, A., Leisewitz, A., 2010. Canine parvovirus. Veterinary Clinics of North America: Small Animal Practice 40, 1041–1053.

Greenbaum, J., Nirmalan, M., 2005. Acid–base balance: Stewart’s physicochemical approach. Current Anaesthesia and Critical Care 16, 133–135.

Heald, R.D., Jones, B., Schmidt, D., 1986. Blood gas and electrolyte concentrations in canine parvoviral enteritis. Journal of the American Animal Hospital Association 22, 745–748.

Hopper, K., 2012. Incidence, nature and etiology of metabolic acidosis in dogs and cats. Journal of Veterinary Internal Medicine 26, 1107–1114.

Hopper, K., Haskins, S.C., 2008. A case-based review of a simplified quantitative approach to acid–base analysis. Journal of Veterinary Emergency and Critical Care 18, 467–476.

Kellum, J.A., 2007. Disorders of acid–base balance. Critical Care Medicine 35, 2630–2636.

Kellum, J.A., 2008. Acid–base balance: Albumin and strong ions. In: Anaesthesia Science. Wiley-Blackwell, Chichester, UK, pp. 188–197.

Kurtz, I., Kraus, J.A., Orzechik, V., Nguyen, M., 2008. Acid base analysis: Critique of the Stewart and bicarbonate centred approaches. American Journal of Renal Physiology 9, 1009–1031.

Morgan, T.J., 2009. The Stewart approach – One clinician’s perspective. The Clinical Biochemist 40, 41–54.

Nappert, G., Dunphy, E., Ruben, D., Mann, F.A., 2002. Determination of serum organic acids in puppies with naturally acquired parvoviral enteritis. Canadian Journal of Veterinary Research 66, 15–18.

Prittie, J., 2004. Canine parvovirus: A review of diagnosis management and prevention. Journal of Veterinary Emergency and Critical Care 14, 167–176.

Rai, A., Nauriyal, D.A., 1992. Note on acid–base and blood gas dynamics in canine parvoviral enteritis. Indian Journal of Veterinary Medicine 12, 87–88.

Schoeman, J.P., Goddard, A., Leisewitz, A.J., 2013. Biomarkers in canine parvovirus enteritis. New Zealand Veterinary Journal 61, 217–222.

Schoeman, J.P., 2008. Endocrine changes in canine critical illness. Thesis, Doctor of Philosophy, University of Cambridge.

Schoeman, J.P., Hertridge, M.E., 2008. Serum thyrotropin, thyroxine and free thyroxine concentrations as predictors of mortality in critically ill puppies with parvovirus infection: A model for human paediatric critical illness? Microbes and Infection 10, 203–207.

Schoeman, J.P., Goddard, A., Hertridge, M.E., 2007. Serum cortisol and thyroxine concentrations as predictors of death in critically ill puppies with parvoviral diarrhea. Journal of the American Veterinary Medical Association 231, 1534–1539.

Siggaard-Andersen, O., Engel, K., Jorgensen, K., Astrup, P., 1960. Micro method determination of pH, carbon dioxide tension, base excess and standard bicarbonate. Scandinavian Journal of Clinical Laboratory Investigation 12, 172–176.

Siggaard-Andersen, O., Fogh-Andersen, N., 1995. Base excess or buffer base (strong ion difference) as measure of a non-respiratory acid–base disturbance. Acta Anaesthesiologica Scandinavica 39, 123–128.

Sinka, A., Rhodes, A., Grounds, R., Bennet, E., 2002. Acid–base physiology: the “traditional” and “modern” approaches. Anaesthesia 57, 348–356.

Story, D.A., Kellum, J.A., 2005. Acid–base balance revisited: Stewart and strong ions. Seminars in Anaesthesia, Perioperative Medicine and Pain 24, 9–16.

Vivio, J., Jose-Cunilleras, E., Armengou, I., Cesarini, C., Tarancón, I., Rios, J., Monreal, L., 2010. Acid–base imbalances during a 120 km endurance race compared by traditional and simplified strong ion difference methods. Equine Veterinary Journal 38, 76–82.

Whitehair, K.J., Haskins, S.C., Whitehair, J.G., Pascoe, P.J., 1995. Clinical applications of quantitative acid–base chemistry. Journal of Veterinary Emergency and Critical Care 1, 1–11.

Wooten, W., 2004. Science review: Quantitative acid–base physiology using the Stewart model. Critical Care 8, 448–452.