Haematological and biochemical parameters of clinically dehydrated and euhydrated dogs

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Abstract
Dehydration results in alterations in haematological and biochemical parameters of dogs. The aim of this study was to determine the haematological and biochemical parameters of clinically dehydrated and euhydrated dogs. A total of 109 samples obtained from different breeds of dogs were used for this study. The degrees/grades of dehydration of each dog were recorded and the dogs were grouped as follows based on their degrees of dehydration: A; euhydrated, B; mildly dehydrated, C; moderately dehydrated and D; severely dehydrated. Whole blood was collected via cephalic venipuncture for complete blood counts. The concentrations of urea, creatinine, total protein, albumin, glucose, sodium, calcium, chloride, phosphorus, potassium, bicarbonate, blood urea nitrogen (BUN)/creatinine ratio and anion gap (AG) as well as serum activities of alanine aminotransferase, aspartate aminotransferase and alkaline phosphatase were determined using serum prepared from whole blood. Urinalysis was done using fresh urine sample. Significantly higher packed cell volume (PCV) and haemoglobin concentration (Hb) (P < 0.05) due to dehydration was observed in B, while significantly lower PCV and Hb (P > 0.05) were reported in C and D. The concentrations of urea, creatinine, BUN/creatinine ratio, total protein, albumin and urine specific gravity were significantly higher (P < 0.05) in B, C, D compared to A. However, the concentrations of glucose, calcium, sodium, chloride, potassium, phosphorus, bicarbonate and anion gap and the activities of liver enzymes were comparable (P > 0.05). It was concluded from this study that; the relative increases in PCV and Hb concentration observed in B was due to haemoconcentration (dehydration), the reported anaemia in C and D were in existence with certain degrees of dehydration and that the increases in serum urea, creatinine, total protein and albumin concentrations were consistent biochemical indicators of dehydration in dogs.

Keywords: Biochemical, Dehydrated, Dog, Euhydrated, Haematological

Introduction
Dehydration describes a state of negative fluid balance that may result from pathological alterations in the body (Cheuvront et al., 2010a). The word “dehydration” is derived from the Greek words hydor which means water and de which means removal, deprivation or separation (Bryant, 2007).
The negative fluid balance that causes dehydration may result from decreased water intake, increased output and/or fluid shift (Popkin et al., 2010) and hence to maintain fluid balance, water intake should be equal to fluid lose. However, fluid imbalance could results in rapid and severe performance problems in patients due to the many vital roles water plays in the maintenance of nearly all body functions (Cheuvront et al., 2010b).

Dehydration could be graded as mild, moderate or severe according to the presence or absence of physical signs such as dry mucous membrane, loss of skin turgor, enophthalmos, weak pulses, and/or altered level of consciousness (Davis et al., 2013). In clinical settings, dehydration secondary to vomiting and diarrhoea is the major cause of death worldwide (Thomas et al., 2008; Bhat et al., 2013) Estimating the percent dehydration is an important tool in rehydration therapy.

Haematological and serum biochemical parameter are widely used diagnostic tools of dehydration assessment (Sara et al., 2012) in dogs because clinical signs of dehydration are highly subjective (Kaneko et al., 2008, Ganio et al., 2011; Sara et al., 2012). This study was therefore carried out to determine the haematological and biochemical parameters of clinically dehydrated and euhydrated dogs.

Materials and Methods

Study area

The study was conducted at the Veterinary Teaching Hospital and Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria, Nigeria. Zaria lies in the Guinea Savannah belt, located at 2200 feet above sea level within latitudes 11°7’, 11°12” N and longitude 7°41” E with a mean annual temperature of 27°C. Zaria experiences distinct wet and dry seasons. (Abdulhamid et al., 2016).

Experimental animals and study design

A total of 109 samples obtained from different breeds (caucasian, rottweilers, alsatians, cross-bred, and Nigerian indigenous) of dogs were collected and analyzed. The dogs comprised of clinically healthy (euhydrated) and clinically dehydrated dogs. Degrees of dehydration were estimated as previously described (Davis et al., 2013). The dogs were grouped as follows depending on their degrees of dehydration:

Group A: Euhydrated (control) dogs, n = 30.
Group B: Mildly dehydrated (5 % dehydration) dogs, n = 42.
Group C: Moderately dehydrated (8 % dehydration) dogs, n = 30.
Group D: Severely dehydrated (>10 % dehydration) dogs, n = 7.

Determination of haematological parameters

Packed cell volume (PCV), total and differential leukocyte counts were determined as described (Coles, 1980). Mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) were calculated from values of total red blood cell, PCV and haemoglobin concentration as described by Cole (1980).

Determination of serum biochemical changes

Sera were obtained from whole blood and stored at -20°C until analyzed. Concentrations of creatinine, urea, total protein, albumin, glucose and electrolytes such as sodium (Na⁺), potassium (K⁺), chloride (Cl⁻), calcium (Ca²⁺), phosphate (PO₄³⁻), bicarbonate (HCO₃⁻), aspartate amino transferase (AST), alanine amino transferase (ALT) and alkaline phosphatase (ALP) in the serum were measured using commercial test kits with the aid of an ultraviolet digital spectrophotometer (Aktas et al., 2011). The BUN/Cr ratio was calculated by dividing the value of BUN by that of creatinine. Anion gap was calculated to determine acid-base abnormalities using the following equation as described previously by George (1994).

Urine sample collection and analysis

Urine samples (10 mL) were collected aseptically by cystocentesis or transurethral catheterization into sterile sample bottles and labeled accordingly. The fresh samples were analyzed both macroscopically and chemically for the detection of abnormal findings (Khorami et al., 2010).

Statistical analysis

Data from the study was computed as mean ± SEM, analyzed with Graph pad prism version 5.2 using ANOVA (Analysis of variance). Significance was accepted for values of p < 0.05.

Results

Haematological parameters (mean ± SEM) of euhydrated (A) and dehydrated (B, C and D) dogs investigated were as presented in Table 1. There was no significant (P > 0.05) difference in PCV between A and D. However, there was significant (P < 0.05) difference between A and B, and also between A and C. The MCV of group A comparable (P > 0.05) to
those of B and C. Meanwhile, significantly higher (P < 0.05) MCV values was observed A and D. MCHC of A were comparable (P > 0.05) to those of B, C and D. The mean total leucocytes and differential counts did not differ significantly (P > 0.05) between the groups, except for neutrophils and total leucocyte count of D which differs significantly (P < 0.05) from that of A (Table 1).

Changes in biochemical parameters (mean ± SEM): electrolyte profiles, metabolite concentrations, liver enzyme activities and some urinary indices of euhydrated and dehydrated dogs were as presented in Tables 2. There were significant differences (P < 0.05) in the concentrations of creatinine, urea, U/B/C ratio, total protein, albumin and urine specific gravity in A and those of B, C and D. However, there was no significant difference (P > 0.05) in serum concentrations of glucose, Na⁺, K⁺, Ca²⁺, Cl⁻, P, HCO₃⁻, anion gap, the mean activities of AST, ALT and ALP and the urine pH in A compared to those of B, C and D (Table 2).

### Table 1: Haematological parameters (mean ± SD) of dehydrated and euhydrated dogs examined in Zaria, Nigeria

| Parameters          | Group A (Control) (n = 30) | Group B (n = 42) | Group C (n = 30) | Group D (n = 7) |
|---------------------|-----------------------------|------------------|------------------|-----------------|
| PCV (%)             | 37.80±6.09                  | 43.98±7.71†      | 29.10±10.78*     | 30.86±10.93     |
| Hb(g/dl)            | 12.65±2.04                  | 14.58±2.52†      | 9.69±3.54*       | 10.13±3.50      |
| RBC (×10¹²/L)       | 5.43±0.80                   | 6.21±1.30†       | 4.36±1.49†       | 4.97±2.00       |
| MCV (fL)            | 70.31±4.34                  | 68.51±6.02       | 63.17±6.55†      | 67.11±4.46      |
| MCH (g/dl)          | 33.68±1.36                  | 32.40±0.70       | 32.91±0.68       | 33.23±1.50      |
| WBC (×10³/L)        | 11.35±2.72                  | 13.20±6.23       | 15.37±6.32       | 27.93±12.60*    |
| Neutrophils (×10³/L) | 8.33±3.05                   | 9.13±5.75        | 11.77±5.14       | 22.51±11.37†    |
| Lymphocytes (×10³/L) | 1.83±1.04                   | 1.39±1.52        | 2.07±1.55        | 1.16±0.67       |
| Eosinophils (×10³/L) | 0.39±0.40                   | 0.37±0.67        | 0.35±0.87        | 0.20±0.15       |
| Monocytes (×10³/L)  | 0.51±0.49                   | 1.00±1.24        | 0.49±0.45        | 0.47±0.45       |
| Basophils (×10³/L)  | 0.00±0.00                   | 0.01±0.04        | 0.00±0.00        | 0.00±0.00       |

Key: SD = standard deviation, Means with superscript * differs significantly from their corresponding control values (P < 0.05), PCV= packed cell volume, Hb= haemoglobin, RBC= red blood cell, MCHC= mean cell haemoglobin concentration, MCV= Mean corpuscular volume and WBC= White blood cell

### Table 2: Biochemical parameters (mean ± SD) of dehydrated and euhydrated dogs examined in Zaria, Nigeria

| Parameters             | Group A (Control) (n = 30) | Group B (n = 42) | Group C (n = 30) | Group D (n = 7) |
|------------------------|-----------------------------|------------------|------------------|-----------------|
| Creatinine (mg/dl)     | 0.97±0.33                   | 2.65±0.76*       | 3.27±0.99*       | 5.37±1.77*      |
| Urea (mg/dl)           | 11.13±5.00                  | 46.67±9.69*      | 54.57±13.90*     | 127.90±75.94*   |
| BUN/Cr ratio           | 5.57±2.33                   | 8.30±2.21*       | 7.44±2.35*       | 10.59±4.33*     |
| Total protein (g/l)    | 58.80±11.21                 | 73.12±11.12*     | 75.47±9.70*      | 82.43±13.00*    |
| Albumin (g/l)          | 25.40±8.16                  | 37.24±10.33*     | 38.83±7.38*      | 39.43±6.47*     |
| Glucose (mmol/l)       | 5.46±1.33                   | 5.30±1.44        | 5.09±1.38        | 6.24±1.70       |
| USG                    | 1.015±0.006                 | 1.022±0.007*     | 1.024±0.006*     | 1.024±0.007*    |
| Urine pH               | 6.77±1.36                   | 6.88±1.16        | 7.13±1.16        | 7.13±1.35       |
| Na⁺ (mmol/l)           | 142.50±7.88                 | 144.20±7.46      | 142.50±11.76     | 152.00±6.90     |
| K⁺ (mmol/l)            | 4.48±1.00                   | 4.56±1.66        | 4.30±0.87        | 5.20±1.48       |
| Ca²⁺ (mg/dl)           | 9.75±0.59                   | 10.21±0.43       | 9.88±1.49        | 9.21±1.44       |
| Cl⁻ (mg/dl)            | 116.70±8.08                 | 114.60±9.97      | 111.60±15.85     | 123.00±11.15    |
| P (mg/dl)              | 4.31±1.78                   | 4.61±4.02        | 4.29±1.69        | 5.27±2.54       |
| HCO₃⁻ (mmol/l)         | 21.50±4.15                  | 20.70±6.71       | 21.97±4.94       | 18.29±7.59      |
| AG (mEq/L)             | 11.08±8.43                  | 14.11±7.06       | 16.24±8.82       | 14.63±9.36      |
| AST (U/l)              | 12.57±2.66                  | 12.43±4.32       | 12.87±1.94       | 13.29±3.10      |
| ALT (U/l)              | 23.93±23.28                 | 17.12±8.79       | 12.63±18.46      | 62.14±42.07     |
| ALP (U/l)              | 51.33±36.88                 | 37.69±20.17      | 40.00±17.30      | 70.29±39.23     |

Key: SD = standard deviation, Means with superscript * differs significantly from their corresponding control values (P < 0.05), BUN/Cr ratio= blood urea nitrogen/creatinine ratio, USG = urine specific gravity, AG=anion gap
Discussion

Group B dogs had a relatively increase values PCV and Hb concentration when compared to dogs of groups A, C and D. This finding coupled with increases in the concentrations of haemoglobin, total plasma protein, albumin, urea, creatinine and blood urea nitrogen/creatinine were attributed to dehydration ratio. Similar findings have been reported previously (Useh et al., 2005; Panda, 2006; Capitelli and Crosta, 2013; Armstrong et al., 2016). The abnormally lower values of PCV and Hb (anaemia) of dogs in groups C and D compared to group A dogs could be attributed to the clinical history of blood loss in dogs belonging to these groups. The finding of this work agreed with previous studies (Norman; 2009; Atata et al., 2018) but disagrees with previous reports of unchanged mean PCV and Hb concentration in dehydrated dogs (Panda, 2006; Bhat et al., 2013). The anaemia reported in dogs in groups C and D were in existence with certain degrees of dehydration (5% and 8% respectively) as observed during physical examination findings and these values could be highly deceptive because they have been masked by haemoconcentration as a result of dehydration. 

The reported marked neutrophilic leukocytosis observed in group D dogs was attributed to primary bacterial infection or secondary bacterial complication in this group of dogs. However, normal values of total and differential leucocyte counts of groups B and C dogs were suggestive of a normal leukogram indicating the absence of entities such as stress that could result in leukocytic alterations. Higher levels of urea and creatinine were observed in dehydrated (B, C and D) dogs and these increases were consistent with the increase in percentage dehydration. This finding agreed with a recent study (Fathi & Asiaban, 2016) on water deprivation in camels. Even though, several reports had demonstrated an elevated BUN/creatinine ratio (Fortes et al., 2015; Lin et al., 2016) as a result of dehydration, normal ratio was observed in this present study and no explanation could be offered for this variation.

Increased total protein (C and D) and albumin concentrations (B, C and D) were observed due to haemoconcentration as a result of dehydration. This finding agreed with previous reports (Kirchner et al., 2014; Noleto, 2016). The observed normal concentrations of blood glucose in this study was a prove that glucose concentrations was not adversely affected by dehydration and this agreed with the notion that blood glucose level is tightly regulated because of its role in maintenance of the central nervous system function (Rodriguez et al., 2005).

The finding of this study of higher urine specific gravity was in conformity with the fact that urine becomes concentrated during dehydration with normal renal function (Armstrong, 2016; Cheuvront, 2016). However, urinary pH did not differ in this study because urinary pH is determined by blood pH and blood pH did not also differ in this current study. The different degrees of dehydration had no effect on serum concentrations of sodium, calcium, chloride, potassium, phosphorus, bicarbonate ions and anion gaps. Similar finding was reported (Reineke et al., 2013). In this study, the activities of ALT, AST and ALP were within normal limit suggesting an absence of hepatic pathology (Esievo, 2017). This finding also supported the fact that the higher plasma protein (albumin) levels reported was as a result of haemoconcentration and not liver disease.

In conclusion, the relative increases in PCV and Hb concentration observed in B was due to haemoconcentration as a result of dehydration. Also, the reported anaemia in C and D were in existence with certain degrees of dehydration. However, this study showed that increases in serum urea, creatinine, total protein and albumin concentrations were consistent biochemical indicators of dehydration in dogs.

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