

EFFECT OF DIARRHEA ON SERUM ELECTROLYTES AND TRACE ELEMENTS IN SHEEP AND GOATS

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ARTICLE INFORMATION

Article History:

Received: 23rd September 2022

Accepted: 10th September 2023

Published online: 30th September 2023

Author's contribution

SM & MKI designed and executed the study. SM did sampling. MAR, MK and MN processed the samples. MKI compiled and analyzed the data statistically. SM and MK wrote the manuscript. BM reviewed the manuscript.

Key words:

Electrolytes, trace elements, Sheep, Goat, Pack Cell Volume

ABSTRACT

Diarrhea leads to deprivation of health and decrease in production potential of an animal. The main objective of this study was to evaluate the serum electrolytes and trace elements in diarrheic sheep and goats. For this study, a total of 100 diarrheic animals were included, comprising 50 sheep and 50 goats experiencing clinical diarrhea, regardless of the cause from different livestock farms situated in peri urban areas of Lahore were examined. The results showed that the levels of serum Sodium (Na) and Potassium (K) were increased significantly ($P < 0.05$) while serum Calcium (Ca) level was decreased significantly ($P < 0.05$) in diarrheic group as compared to the control group. There is no significant change in serum Iron (Fe) and Copper (Cu) levels in diarrheic group as compared to the control group. Serum copper and Pack cell volume (PCV) had a significant correlation in case of diarrheic animals. It was concluded that diarrhea have a significant correlation with serum electrolytes and trace elements. This study will help to veterinary practitioners in field regarding diagnosis and treatment of diarrhea.

1. INTRODUCTION

Diarrhea leads to deprivation of health and decrease in production potential of animals (Ijaz *et al.*, 2019). Diarrhea means increase in frequency, fluid quantity and volume of fecal excretion. Animals suffering from diarrhea may have blood, mucous, foul smell and abnormal color of feces unlike healthy animals. Multiple factors causing diarrhea include environmental factor, improper animal hygiene, nutritional, excessive amounts of milk or milk replacer and infectious causes like rotaviruses, corona viruses, entero-toxigenic *E. coli* (Kaba *et al.*, 2006). Overfeeding, overpopulation, cold temperature, bad hygiene, artificial feeding and colostrums deprivation are all predisposing factors which can be important in the complex etiology of diarrhea (Hemashenpagam *et al.*, 2009).

During diarrhea, dehydration occurs due to loss of water and electrolytes from the gastrointestinal tract and in severe form it leads to death of the animal. Dehydration in animals occurs when extracellular and intracellular fluid, blood volume, water or electrolyte concentration decreases. Severe deficiency of electrolytes in diarrheic ruminants might be the reason of death in serious cases. Even though it is well understood that serum concentration of sodium (Na) and potassium (K) are necessary to rise to the normal level with reference to the fluids used for therapy of diarrhea (Groutides & Michell, 1990). Untreated changes in serum concentrations of Ca, Mg and some trace elements may be the cause of some death or post diarrhea complications such as growth retardation (Tajik & Nazifi, 2012).

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Some macro minerals and trace elements are classified as essential mineral elements. This classification is based on the concentration needed in the diet and in animal tissues (Herdt & Hoff, 2011), and does not reflect the importance of the mineral (Rankins & Pugh, 2012). Certain macro minerals are required in large quantities and include calcium, chloride, magnesium, phosphorous, potassium, sodium and sulphur. Essential micro minerals or trace elements include cobalt, copper, iodine, iron, manganese, molybdenum, selenium and zinc. These elements are required in smaller quantities (Fraga, 2005). Trace elements (Copper, Iron and Cobalt) take part in growth, reproduction and productivity of household animals. Loss of minerals suffers major losses mutually in production and economy (Graham 1991; Jones et al., 1990). The major clinical symptoms of trace element deficiencies in animals are diarrhea, anemia, loss of hair, depigmentation, growth disorders in bones, difficulty in walking, scurf in skin, hyperkeratosis, parakeratosis, lack of appetite, fertility disorders, decrease in reproductively, young animal growth disorders, tetany, decrease in protein synthesis, immune deficiencies, abortus related to non-infectious reasons and pica (Saleh, 2019). Elements required in less quantity are essential to enhance resistance against ailment in animals. However, decrease in concentration of trace elements like cobalt, copper, iron, iodine, manganese, selenium, or zinc leads to decrease reproductive rate and lactation (Arshad et al., 2020). While milk fever (hypocalcaemia), grass tetany (hypomagnesaemia) and abomasum displacement are due to electrolyte imbalance and usually occur at early stage of milk production which leads to considerable economic losses (Graham, 1991). Therefore, these minerals like calcium-phosphate have great value in both adult and growing ruminants (Saleh, 2019).

Keeping in view the significance of electrolytes and other trace elements in diarrhea, this study was designed to determine the serum concentration of electrolytes and trace elements (Ca, Cu, Na, Li, K and Fe) in diarrheic sheep and goat.

2. MATERIALS AND METHODS

Experimental design

In this study, a total of 100 diarrheic animals were included, comprising 50 sheep and 50 goats experiencing clinical diarrhea, regardless of the cause. Additionally, a negative control group of 10 animals was established, consisting of 5 sheep and 5 goats.

Collection of Samples

Blood samples

Blood sample (5ml) was collected from the jugular vein of each of the animal using disposable syringes under aseptic conditions. After collection, blood samples were

carefully transferred to EDTA coated vacutainer (purple-topped) for analysis.

Serum samples

The blood samples were allowed to clot and then clotted blood was centrifuged at 3500rpm for 5 minutes. After centrifugation, the supernatant, clear straw-colored fluid (serum) was aspirated into eppendorf with the help of pasture pipette. The serum samples thus collected were stored at -20°C until analysis was performed (Ijaz et al., 2019). All serum samples were prepared by wet digestion following the procedure was 0.5 ml serum sample was digested with 10ml concentrated nitric acid in a 100ml digestion flask at low temperature for about 15-20 minutes until the contents were clear and then with 5ml perchloric acid for 15 minutes. The solution in the flask was heated vigorously till 2-3ml colourless material was left. After cooling the contents were diluted up to 20ml with redistilled water in a volumetric flask and preserved for the analysis of minerals (Akhtar et al., 2007).

Serum electrolyte and trace element analysis

Concentrations of serum electrolytes and trace elements Ca, Cu, Na, Li, K and Fe were estimated through atomic absorption spectrophotometer. Acid digestion was used to prepare samples for chemical analysis of Ca, Cu, Na, Li, K and Fe. These levels were determined by using Atomic Absorption spectrophotometer (Varian AA-1275). The air-acetylene gas mixture was used as fuel for flame production. For each element, a specific hollow cathode lamp was used. The properly diluted standard solutions were individually aspirated, and absorbance was recorded to draw calibration curve for each mineral. Subsequently, concentrations of each element (ppm) in samples were determined by taking absorbance and comparing it with calibration curve.

Absorption of blanks was also determined to rule out any contamination in reagents and glassware and their values were subtracted from samples.

Blood Parameters

Hematology was performed through hematological analyzer. (Diatron company: abacus model)

Statistical Analysis

Analysis of variance tests (ANOVA) was used to compare serum electrolytes, trace elements and clinical signs after diarrhea and t-test was used to compare diarrheic and non-diarrheic sheep with serum electrolytes and trace element. Duncan's test applied for significant variables.

3. RESULTS

Serum samples were obtained from 100 diarrhea (n=50 sheep; n=50 goat) suffering from clinical diarrhea irrespective of cause of diarrhea was included in this

study. In the serum sample of diarrheic as well as non-diarrheic group of both sheep and goat, the concentration of sodium, potassium, calcium, copper and iron were evaluated. The results of the measurement of the serum electrolytes and trace elements of sheep are shown in Table 1.

The serum values of Sodium and Potassium were increased significantly ($P < 0.05$) while Calcium was decreased significantly ($P < 0.05$) in diarrheic sheep as compared to non-diarrheic group (table 1). Trace elements such as Copper and Iron are required in a minute concentration for various physiological functions. Serum copper and iron were also measured in both diarrheic and non-diarrheic groups of sheep. The serum values of copper and iron were also decreased non-significantly ($P > 0.05$) in diarrheic sheep as compared to non-diarrheic (table 1).

In diarrheic goats, the serum values of Sodium and Potassium were increased significantly ($P < 0.05$) whereas Calcium was decreased significantly ($P < 0.05$) as compared to non-diarrheic group (table 2). Serum copper and iron values were also measured in both diarrheic and non-diarrheic groups of goats. The serum copper and iron concentration were also decreased non-significantly ($P > 0.05$) in goats having diarrhea as compared to non-diarrheic group (Table 2).

The values of PCV were significantly upregulated as compared to normal range in both sheep and goat due to diarrhea. This condition leads toward dehydration and consequently PCV (%) increased. Serum copper and PCV had a significant correlation in the case of diarrheic sheep and goat. Copper deficiency is a significant feature of anemia so that serum copper concentration has a direct link with PCV. Iron also has a strong correlation with copper because it is required for utilization, mobilization and absorption of iron in animal body.

4. DISCUSSION

Neonatal diarrhea is a significant cause of economic loss in ruminants. The etiology of diarrhea can be infectious or non-infectious (Elhassan *et al.*, 2011). Overfeeding, overpopulation, cold temperature, bad hygiene, artificial feeding, and colostrum deprivation are all predisposing factors which can be important in the complex etiology of the disease (Hemashenpagam *et al.*, 2009). There are different causes of diarrhea due to some infectious agents like viruses, bacteria or fungi (Chafupnik *et al.*, 2012). In early age, groups of animals are more prone to diarrhea than adults. Causes of diarrhea may include uptake of toxins in small ruminants, as well as grazing at green or damp fodder in sheep and goats. Effects of diarrhea in sheep and goat is anorexia, lethargy, depression, acidosis,

dehydration, and gastroenteritis (Lorenz, 2003; Lorenz, 2007; Gidudu *et al.*, 2011).

In the present study, diarrheic ruminants without any treatment were clinically examined and their fecal consistency, age and days between disease onset and sampling were recorded. Alteration occurs in serum level of Na and K during diarrhea. In our study, the serum level of Na and K increase while Ca level decreases in small ruminants in case of diarrhea, along with significant correlation of PCV and copper. The findings of our study congruent with the findings of Fazal *et al.*, (2019) who reported increased Na and K levels in diarrheic calves. In our study Na and K exhibiting the non-significant difference when co-relate with breed, age, and sex. Although Na level significantly different when co-related with sex in our study. Serum copper concentration had a significant correlation with PCV. In present study electrolytes were quantified with correlation of different breeds, age and sex in sheep as well as co-relations of Cu and PCV that relates with the study of (Groutides & Michell, 1990). These findings are also significant when the co-relation between PCV and copper was measured. There is also a little variation in the level of other electrolytes in serum such as calcium (Ca). From present study Ca level in serum of diarrheic sheep shows the significant difference between breeds, non-significant with age as well as non-significant difference with sex. Laboratory tests are required for proper diagnosis of diarrhea. Hypocalcaemia has been diagnosed as a cause of hypothermia in ruminants, which may explain the observed relationship between Ca and diarrhea in the current study which shows that decrease in Ca in diarrheic animals. Diarrheic sheep and goats also show a significant difference between breeds, non-significant in age groups and non-significant in groups classified based on sex. Changes in trace elements were also studied during diarrhea. Concentration of Fe was also measured in diarrheic sheep and goats during our study. The results showed that Fe has non-significant difference when compare with breed, age and sex. Similarly results of my study showing that the Cu level in serum has significant relation with the value of PCV in sheep. The results of present study also correlate the results of Fazal *et al.*, (2019).

In a previous study, a comparison between the concentrations of Cu and Zn in case of ruminants suffering with diarrhea to the non-diarrheic ruminants has been done and findings show the decrease in value (Arora *et al.*, 2007). The results of current study showed that the serum concentration of Fe and Cu had no significant correlations with clinical signs. Serum copper concentration had a significant correlation with PCV ($P < 0.05$). The results of my study matched with the findings of the study of Rucker *et al.* 2008 as it indicated

that serum Cu level has significant correlation with PCV. Packed cell volume (PCV) is demonstrated as an index for the estimation and quantification of hydration status in the calves (Constable et al. 1998). PCV is the best indicator for the diagnosis of changes in hydration status (Haenlein & Anke, 2011) so there is decrease in hydration status of small ruminants which is coincide with the study of Elhassan et al., (2011). Higher PCV means more severe dehydration, which is due to the prolonged water loss (Sadeghian et al., 2011).

5. CONCLUSION

Trace elements and serum electrolytes take part in growth, reproduction, and productivity of household animals. Loss of minerals suffers major losses mutually in production and economy. This study highlights the importance of electrolyte and trace elements in diarrheic sheep and goats. This study will help veterinary practitioners in the field regarding diagnosis and treatment of diarrhea.

6. CONFLICT OF INTEREST

All authors have declared that there is no conflict of interests regarding the publication of this article.

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Table 1: Serum concentrations of electrolytes and trace elements in diarrheic and non-diarrheic sheep

Elements	Diarrheic sheep	Non-diarrheic
Sodium	2972.78±29.9	2433.78±24.9
Potassium	277.848± 12.8	241.4± 11.35
Calcium	68.1± 2.69	118.6± 8.65
Copper	4.44± 1.4	5.8± 1.46
Iron	4.46± 0.50	4.2±0.40

Table 2: Serum concentrations of electrolytes and trace elements in diarrheic and non-diarrheic goats

Elements	Diarrheic goats	Non-diarrheic
Sodium	2833.78±29.9	2572±76.1
Potassium	279.848± 14.8	241.4± 9.3
Calcium	88.1±8.6	118.6± 9.6
Copper	4.6± 0.4	4.8± 0.8
Iron	4.5± 0.30	4.4±0.5