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Serum Concentration of Cobalt, Copper, Iron, Phosphorus, and Magnesium in Cattle Naturally Infected With *Theileria annulata* .

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Abstract: The present study was planned to evaluate serum albumin, glucose and macrominerals status Calcium(Ca), Cobalt(Co), Copper (Cu), Iron(e), Magnesium(Mg) and Phosphorus(P) in Cattle which naturally infected with *Theileria annulata*. The material of this study was a total of 15 Cattle with different ages, breeds and genders diagnosed as theileriosis according to clinical and microscopical examination and 10 clinically healthy animals without parasitaemia (according blood smears). According to statistical analysis of biochemical parameters;



Serum glucose, Ca, Co, Cu, Fe, Mg and P concentrations were detected significantly low in cattle with theileriosis ($P < 0.05$). In conclusion, mineral substance levels were altered in cattle naturally infected with theileria annulata. Additionally to the classical treatment of theileriosis, administering mineral supplements including Ca, Co, Fe, Cu, Mg and P to the animals is thought to be more useful in treatment.
Keyword: Thileriosis, Macrominerals, Cattle.

1 .Introduction:

Tick-borne diseases (TBDs) are a wide spread disease causing substantial economic losses and are a constant threat to the development of animal health(1). Bovine tropical theileriosis caused by the haemoprotozoan *theileria annulata*, is a tick-borne disease that transmitted to cattle by ixodid ticks of genus *Hyalomma*.(2). Two stages in the life cycle of the parasite are responsible for the pathogenesis of the disease. These are Schizont in mononuclear cells of the reticuloendothelial system and intraerythrocytic piroplasm (3,4).

The disease occurs in a large part of Asia, a wide zone of Africa and southern Europe(5).Blood biochemical alteration can provide valuable information about the severity of the infection and are considered to be good tools for the diagnosis, prognosis and evaluation of the therapy applied(5). Macro and micronutrient are inorganic substances essential to maintain the normal function and living status in domestic animals .These nutrients play a critical role in physiological processes related to health growth and reproduction, and the adequate function of the immune and endocrine system(6). Cobalt is the main component of vitamin B12 which plays an important role in energy metabolism ,haematological parameters and amino acid metabolism(5). Cobalt has been used for years to prevent unproductiveness with signs of anorexia, low body weight gain, emaciation, low milk yield and anemia (7). Copper (cu) is



a compound of enzyme catalysts and participates in important biochemical function; it reported that there is closely between concentration copper and health in the cows (8).

Iron (Fe) is essential to virtually as a component of hemoglobin and numerous others iron containing proteins in the body. (Fe) deficiency caused the incidence of infectious diseases and impairment of the activities of iron-containing enzymes in cells of the immune system in animals (9). Magnesium (Mg) is an important mineral for in animals, Mg should be sufficient to metabolic activity and protect animal health (10). In cattle with Theileriosis, there were decreased serum calcium and phosphorus concentration in cattle infected with *T. annulata*(11).

Minerals are the critical nutrients bearing significant effect in animal nutrition because both their excess and deficiency produce detrimental effects on the health and production of livestock (10, 11).

The objectives of the present study were:

1. Present changes in serum mineral concentration in cattle with theileriosis before and after treatment.
2. importance of these concentration changes in treatment and prognosis of the Theileriosis

2. Materials and Methods:

2.1 Animals and Parasitological examination: The materials of this study was a total of 15 cattle with different ages ,breed and genders suffering from theileriosis according to clinical and microscopical examination were selected from clinical cases of some farms in Garmut Ali city from September to distemper 2018. On the other hand, 10 clinically healthy animals without parasitaemia (according to the blood smears) located in the same farms served as control.



Clinical observation: 2.2

Clinical examination included history of ticks infestation, duration of illness, changes in manage mental and feeding practice, appetite of the animal, abnormalities in the behavior, gait, posture, rumination, defecation quantity, consistency and frequency), urination, examination of visible mucous membranes , eyes, skin and anus and general clinical examinations including rectal temperature, pulse, respiration and auscultation of heart and lungs. Clinical finding of the animals were evaluated systemically before treatment and at the 7th day after treatment. In order to perform microscopical examination. Single dose of buparvaquon (Butalex -ceva Dif) were administrated to infected animal with dose of 2.5mg/kg. live weight. Oxytetracyclin was administrated for 5 days intramuscularly in order to prevent secondary infection.

2.3 Blood collection:

Blood samples were obtained from v . jugulars before and at the 7th day of treatment and for once from the control group for haematological and biochemical analysis .blood sample were obtained in tubes containing EDTA to perform biochemical analysis. Serum was extracted from blood samples obtained to anticoagulant free tubes. Which were centrifuged on 3000 PRM (Rot fix 32-Heitachi) obtained serum samples were preserved in -20c until evolution, blood sample were referred to department of veterinary clinical pathology, Vet. College of Basra University.

2.4 Blood Smear Examination:

Blood smear were stained with 10% Giemsa stain and examine under 100x oil immersion objective using light microscope for the presence of theileria Spp. At least 50 microscopic fields were examined, and the presence of one or more piroplasm in erythrocytes we considered positive. Figure (1).

2.5 Hematological and Biochemical Examination:

Hematocrit (Hct), hemoglobin (Hb) concentration and white blood cell



count(WBC) were measured with hemogram device (QBC) vet autoreader – Idex). Serum cobalt (Ca), Copper (Cu), Phosphorus (P), Iron (Fe), Calcium (Ca) and Magnesium concentration were determined by using spectrophotometric method. Commercially available kits, (Dia Sys GmbH-German, and Centronic GmbH-Germany) were used. Serum glucose concentration was measured spectrophotometrically (Photometer 5010-Boehringer Mannheim). According to the test procedures.

2.6. Statistical analysis:

Statistical analysis was performed using SPSS 20 for windows. F-Test used to compare between groups. Clinical signs, hematological and biochemical parameters were stated as Mean, Standard Deviation, Minimum Maximum values were performed for comparing these attributes. Spearman-Correlation Coefficient was calculated in order to determine the relation between parameters in each group. Statistical significance level was taken as 5%.

3. Results:

3.1 Clinical Findings:

The clinical examination of cattle with theileriosis: Anorexia, dullness, swelling in superficial lymph nodes, hyperemia in visible mucosa and conjunctivas were observed. Anemia and petechial hemorrhage were observed in certain animal (table 1).

Table (1): Clinical finding in Cattle infected with *Theileria annulata* and healthy Cattle.

Parameters	Control n=10 mean ±SD	Before treatment n=15 mean ±SD	7 th day after Treatment n= 15 Mean± SD
Body Temperature	38.02±0.29a	41.30±0.87b	38.45±0.16c



Heart Rate (beat/min)	73.50±0.19a	84.24±0.84b	80.23±0.79c
Respiratory Rate (breath/min)	27.91±.028a	43.57±0.25b	35.74±0.23c

Importance of statistical difference in the same lines was determined as: $P < 0.05$.

3.2. Hematological Finding:

Hemoglobin concentration , hematocrite , and total leukocyte count were significantly lower in suffering from theileriosis than in the healthy controls ($P < 0.05$), despite treatment , while hematological parameters of cattle with theileriosis were detected close to same hematological parameters of healthy cattle;

Htc and WBC values were detected statistically lower than healthy cattle ($P < 0.05$) .Table (2)

Table (2): Hematological findings in healthy Cattle and Cattle with theileriosis.

Parameters	Control n=10 ± SD mean	Before Treatment n=15 mean± SD	7 th day after Treatment n=15 mean± SD
Hemoglobin Concentration(g/dl)	10.20±0.32a	6.08±0.31b	8.11±0.23c
Hematocrit (%)	29.67±0.45a	14.61±0.29b	24.54± 0.29c
Total Leukocyte /L)Count(× 10 ⁹	7.44±0.16a	4.89±0.62b	6.39±0.14c

Importance of statistical difference in the same lines was determined as : $P < 0.05$.

3.3 Biochemical Finding:

There is a decrease in serum albumin concentration in the cattle infected with *T. annulata*. There was , however no significant difference in the albumin ratio between infected and healthy cattle ($P > 0.05$). Serum glucose , P ,Ca, Co ,Fe ,Cu ,and Mg in cattle with theileriosis were detected



significantly lower than control group before treatment ($P < 0.05$). Despite treatment ;serum glucose($P < 0.05$) ,Co($P < 0.05$),

Cu($P < 0.05$) ,Fe($P < 0.05$) ,Mg($P < 0.05$) and P($P < 0.05$) concentration were detected lower in cattle with theileriosis at the 7th day of treatment than the same parameters of control group (table 3).

Table (3): Biochemical parameters in healthy Cattle and Cattle with Theileriosis.

Parameters	Control n=10 mean±SD	Before treatment n=15 mean±SD	7 th day after Treatment n=15 mean±SD
Glucose(mg/dl)	70.24±4.80a	44.12±5.28b	60.21±3.35c
Albumin(gr/dl)	2.12±0.30a	2.11±0.39a	2.60±0.24a
Ca(mg/dl)	7.98±2.24a	3.50±1.73b	6.77±0.69c
Co(mg/l)	49.46±6.10a	18.55±3.24b	44.89±4.22c
Cu(mg/l)	1119.21±246.40a	498.52±51.14b	758.65±101.25c
Fe(mg/l)	1.60±0.25a	0.36±0.12b	1.11±0.122c
Mg(mg/dl)	2.69±0.34a	1.33±0.42b	1.98±0.14c
P(mg/dl)	5.30±0.77a	2.70±0.65b	3.65±0.56c

Importance of statistical difference in the same lines was determined as: $P < 0.05$.

Discussion: 4.

The determination of blood biochemical parameters is an important part of the clinical laboratory diagnostic tools, available to veterinarians to assess health and disease in animal species. In this study Bovine theileria



lead to alteration in some plasma biochemical parameters. Significant changes in cattle infected with *theileria annulata* were reported in several studies (12 ,13 ,14).In the present study ;RBC count ,PCV and Hb concentration in cattle with theileriosis were significantly lower than healthy ones ($P<0.05$).With progression of parasitemia severity ,a significant decrease in WBC count, Pcv and Hb concentration was observed ($P<0.05$).These findings were similar to those of many researchers (12,14,15).The decline in RBC count ,PCV and Hb concentration can also be attributed to multiplication of Piroplasms in RBC which results in erythrolysis (16).Similar observation have been made by previous workers like (17, 18, 19) there were significant in the decrease in Hb, Pcv, and significant decrease leukocyte affected cattle. In the present study ,there was decrease in serum glucose concentration in cattle infected with *T. annulata* compared with control group .These observations are agreement with that of researches (20,13) as they reported that hypoglycemia could be due to the utilization of glucose by parasites and damage to the liver in cattle infected with *T. annulata* , but contradicts to that of (21) ,who showed anon-significant decrease in calves infected with *T.annulata*. Our finding are in accordance with reports made by (22, 23, 24) there were significant decrease in glucose, protein. This might be due to liver damage, and this in agreement with other studies conducted in different regions (25, 26, and 27) reported the lower serum total albumin concentration in cattle naturally infected with *T. annulata* was possible because of hypoalbuminaemia and hypoglobulinemia resulting from liver failure and sever lymphocytopenia. In current study serum albumin concentration in animal infected with *T. annulata* were found to be numerically lower concentration of healthy cattle. The decreased serum copper and iron concentrations in theileria-infected cattle could be attributed to the in ability of the damaged liver to synthesis ceruloplasmin and transferring, in this study, serum Fe and Cu concentration were detected significantly lower than control group($P<0.05$).Copper has roles on superoxide dismutase and cytochrome oxidize, myelin synthesis and keratinization(28). Cytochrome C oxidase enzyme which has important function in energy conversion is dependent on



copper (29).Therefore copper is an essential element and primarily has role in carbohydrate metabolism (30) .

In this study; serum Cu levels in healthy cattle were determined as 1130.50 ± 215.14 mg/dl, before treatment, serum Cu levels in cattle with theileriosis were determined as 513.52 ± 62.11 mg/dl .At the 7th day after treatment serum Cu level 828.77 ± 114.46 mg /dl.

Decrease in food intake resulted by anorexia lead to impairment of carbohydrate metabolism (6, 8, 28, 29) is also thought to cause decrease in Cu levels in cattle with theileriosis. In current study; before treatment, while average serum calcium concentration was detected lower in cattle with theileriosis than control group. Decreased calcium concentration in heileriosis could be resulted from decreased dietary intake, intestinal malfunction, kidney damage and decreased phosphorus concentration due to diarrhea and renal wasting (11, 13). Mg has great impact on basic metabolism rate in fat , carbohydrate metabolism and blood glucose regulation(30).Serum magnesium concentration of healthy cattle is reported as 2.29 ± 0.34 mg/dl(21).In the current study Mg levels in bovine with theileriosis was found lower($P < 0.05$)compared with control group.

In this study; the reasons of lower magnesium concentration are thought as increase in tubular resorption and loss of appetite due to gastrointestinal agony. Serum phosphorus levels in healthy cattle are reported as 5.30 ± 0.77 mg/dl. As reported , low food intake by these factors lead to phosphorus deficiency (22).Hypophosphataemia causes various clinical finding such as anorexia, pica, muscle weakness, intravascular haemolysis impaired liver function (31).In this current study ,the serum phosphorus concentration were significantly lower in cattle suffering from theileriosis than in the control group ($P < 0.05$), these dates supported the researches (31,32).In the study ,the decrease in concentration of Mg and P in cattle with theileriosis can be caused diarrhea and wasting(11).



5. Conclusions and recommendations:

Based on the information obtained from this study, serum Ca, Co, Cu, Fe, Mg and P in cattle with *Theileria annulata* were detected low before treatment. Additionally to the classical treatment of theileriosis, administering mineral supplements to the animals is thought to be more useful in treatment. Therefore there should be more studies performed about the contribution of minerals to the treatment of theileriosis. This study will provide an insight to the studies will be performed in this respect

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