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Research Article

Clinical and Hematological Profiles due to Cases of Minerals Deficiency in Local Ewes at Basra, Iraq

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Abstract | Normal development and cellular maintenance of living organisms need mineral as the essential nutritional element. Evaluation the status of the trace minerals is profitable for determining the potential sources of livestock problems. This present study was conducted in the Basra Province during January to August 2017, in which 478 sheep were examined for signs of mineral deficiency and/or poor body condition score. Blood samples were taken to estimate serum biochemistry parameters minerals such as copper, zinc, calcium, magnesium and cobalt. Complete blood count parameters were included in this study. Results reveal that 153 (32%) sheep exhibited signs of mineral deficiency such asalopecia (47%), parakeratosis (19%), diarrhea (14%), pale mucous membrane (88%), lachrymation (9%) and ataxia (8%). There were no marked changes in body temperature between affected sheep and control, while the means of respiratory and pulse rate were significantly elevated in affected sheep. A significant decrease in body condition score and hair coat condition score were recorded. The study also showed that RBC, hemoglobin and pack cell volume was markedly low in the affected sheep than control, while WBC shows no significant changes. RBC indices of the affected sheep such as MCV, MCH and MCHC were not significantly different.The study also revealed that there was decreased (p>0.05) in mean values of copper, zinc, cobalt, magnesium, and calcium of the tested ewes as compared with the control animals. In conclusion, a considerable correlation between the lower mineral levels in the blood of ewes and the general health status leading to reduce the production and subsequently economic losses was established.

Keywords | Body condition, CBC, Clinical signs, Mineral biochemistry, Sheep.

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INTRODUCTION

Mineral deficiency usually involves several minerals as well as other conditioning factors; however, the deficiency symptoms of one mineral may predominate and affect the performance in the ruminant. Trace elements function probablyact as an activator of enzymes, co-factors and a stabilizer of the secondary molecular structures (Aitken, 2008; Anderson & Rings, 2008). Their study has evolved from the recognition of their essential function in cell metabolism. Ruminants often are subject to severe deficiency of the trace elements such as: zinc, cobalt, manganese, selenium and copper.

Living organisms need mineral as well as an essential nutritional element to continue their normal development (Aitken, 2008). In a successful surveillance management, assessment the levels of the trace minerals is beneficial for detecting the main sources of the diseases/problems of the livestock (Anderson & Rings, 2008).

The essential element in sheep is copper; it is easily available in diet and rapidly be absorbed through the abomasum the duodenum. When copper incorporate into specific cuproenzymes .It serves as a critical co-factor, catalyzing the facile electron transfer reactions required for cellular respiration, iron oxidation, pigment formation, neurotransmitter biosynthesis, antioxidant defense, peptide amidation, and connective tissue formation (Balamurugan & Schaffner, 2006). Lack or low concentration of copper in animal feed may cause a deficiency of this micro-element (Suttle, 2010). Copper deficiency is taken to be a severe nutritional problem in tropical regions (including Basra), because of the low copper concentration in animal's diet and/or because of high concentrations of other elements that are antagonistic towards copper such as: molybdenum, iron and sulfur (Cardoso et al., 1997; Sousa et al., 2012).

Zinc is fundamental for the functioning of more than (200) enzymes (Keen et al., 2003), it containing enzymes that found in all of the major metabolic pathways involved in proteins, lipids, nucleic acids and carbohydrates metabolism (O'Dell & Sunde, 1997; Keen et al., 2003). Zinc also plays an essential role in maintaining hoof tissues by stimulation of growth of epithelial cells, production of keratin, improved wound healing and improved cellular integrity (Brown et al., 2007). Cobalt is a dietary essential element for ruminants, allowing synthesis of vitamin B12. It is a cofactor for two enzymes, methyl malonyl – CO A mutase and methionine synthase. Ruminant normally do not have any dietary source of vitamin B12, therefore their supply of this vitamin must be ensured by continues the adequate supply of dietary cobalt. Therefore, Vitamin B12 deficiency can be induced by long-term consumption of Co-inadequate diets (Suttle, 2010; Keady et al., 2017).

The clinical signs of cobalt deficiency are non-specific but lambs are particularly susceptible, exhibiting loss of appetite, growth retardation, debility, emaciation and a watery discharge from the eyes (Keady et al., 2017). Mineral deficiencies may cause clinical disorders that have dramatic effects on the health and survival of sheep, or marginal deficiencies that result in subtle and often undetected effects on productivity (Balamurugan et al., 2017).

This current study was therefore conducted to estimate the clinical, hematological and serum level of copper, zinc, cobalt, magnesium and calcium in ewes suspected to be suffering trace minerals deficiency in Basra province.

MATERIAL AND METHODS

This study was conducted in Basra province of Iraq during January August 2017 in different regions of Basra province. Four hundred and seventy-eight (478) dairy ewes were examined, amongst which a total of 153 were suspected to be affected with mineral deficiency, 177 had poor body/ hair coat condition score and 148 clinically healthy ewes were used as a control. Clinical examination was applied to all sheep which included the determination of body temperature, respiratory rate, heart rate with body condition and hair coat condition score.

Body Condition Score (BCS)

Body condition scoring (BCS) is a quick and easy method of describing how fat or thin ewes are. This was achieved by using a numerical score from 1 (very thin) to 5 (very fat) based on the level of muscling and fat deposition around the loin region (Villaquiran et al., 2004; Phythian et al., 2012). Ewes may be given a half score such as 2.5 if it between BCS 2 and BCS 3. An accurate BCS could not be done by looking at the ewe; one must feel the muscle and fat cover. An appropriate BCS range for ewes was from BCS 2 to BCS 4, as seen on the reverse side.

Hair Coat Condition Score (HCS)

The quality of the hair coat condition as animal-based welfare indicator in different condition was examined in all animals throughout the study. Two main points were considered in the scoring of the hair coat condition; normal hair coat (NH) and poor or rough hair condition (RH). The same visually resident identified the animal with a rough hair coat in all groups, which defined as: rough or scarify hair coat, uneven, matted and frequently longer than normal (Battini et al., 2016) while ewes with shiny, homogenous and adheres to the body hair coats were classified as normal. Ewes may reveal rough hair coat on all body or they might only show part of the body covered by the rough hair coat, both categorized as RH.

Blood collection

Jugular blood was collected with an anticoagulant tube for haemogram and leukogram analysis while the tubes without anticoagulant used for trace minerals analysis. All variables of complete blood were determined. Serum samples were analyzed for copper, zinc, cobalt, magnesium and calcium levels using an atomic absorption spectrophotometer. This technique was preferred because it was rapid, simple and accurate.The absorbance of copper, zinc, cobalt, magnesium and calcium in standards and samples solutions were measured by flame atomic absorption spectrophotometer (FAAS) in the research unit laboratory, College of Agriculture, University of Basra, Basra, Iraq under conditions as described by (Ji & Ren, 2002).

Statistical Analysis

Student t-test was performed, and the statistical significance level of sheep parameters were ($p<0.05$). SPSS software "version 11.5" has been used in the analysis of all data of the current study.

RESULTS

Body temperature, respiratory and heart rates were recorded accordingly. The mean of body temperature was similar in both control and mineral deficiency group. While the means of respiratory and pulse rate were significantly increased in the affected animal group as compared to the control ($p<0.05$) (34.15 ±4.68, 87.17±0.57) respectively (Table 1).

Table 1: Temperature, respiratory and heart rate in sheep affected with mineral deficiency

Criteria	Control (Mean ±SE) (NO.= 148)	Affected Sheep (Mean ±SE) (NO.= 330)
Temp(C^0)	39.47±0.09[a]	39.46±0.04[a]
P.R. /Min	77.25± 0.74[b]	87.17±0.57[a]
R.R./ Min	28.75± 1.34[b]	34.15±0.46[a]

[a,b,] values in different superscripts within row were significantly different at ($p<0.05$)

A total of 153 (37.5%) animals showed the following clinical signs of mineral deficiency in sheep (Table 2).

The mean of body condition score of all sheep is detailed in (Table 3). The results of BCS showed significant decreased ($p<0.05$) in body score of affected sheep below the normal range of healthy animals, unlike control sheep which did not show any significant decreased in body condition scoring. A significant change in the condition of the hair coat of affected ewes was noticed. Poor hair coat was observed on 289 ewes (88%) in affected ewes, this poor hair condition includes: uneven, shaggy, matted and rough or scarify hair coat that almost were not involved all parts of the body but categorized as a rough hair condition. Control group had normal hair coat condition (Table 3).

Results of the present study have indicated that there were significant changes in values of RBC count, hemoglobin concentration and PCV in the affected sheep when compared with clinically healthy sheep. WBC was similar in control sheep and sheep affected with mineral deficiency, however, there was no significant variations observed (Table 4).

However, there were no significant differences in values of MCV, MCH and MCHC in the affected and control sheep.These findings suggest that the type of anemia in the affected sheep was normocytic normochromic.

Generally, estimation of the serum levels of the main trace elements in the current study showed marked changes in the ewes with the poor body and hair coat condition score when compared with the clinically healthy ewes .The present study revealed that the values of copper, zinc, cobalt, magnesium and calcium in the serum of affected sheep were markedly low in affected ewes compared to healthy-ewes (Table 5).

DISCUSSION

Several studies have been documented the alteration of the levels of the trace elements of the local domestic animals in Iraq. To our knowledge, the current study is the first survey that covers the clinical manifestations and the hematological valuation that can be related to the main mineral deficiencies in the local sheep in Basra. This study also provides the latest profile of the serum values of the main trace elements in local sheep.

As expected, due to the absence of the inflammatory reaction and the chronicity nature of the minerals deficiency cases, there were no significant differences in temperature between control and affected sheep. Results of the present study also revealed that respiratory and pulse rates were higher in affected sheep than in control. This result is in tandem with documented reports of (Abd El-Raof & Ghanem, 2006). Previous kinds of literatures (Kusiluka & Kambarage, 1996) had explained that the increase in respiratory and pulse rates with mineral deficiency occur to compensate the hypoxia and anemia conditions which resulted from copper or zinc deficiency so as to avail cells with oxygen. The effect of hypoxia decreased RBCs thereby giving an appearance which is pale mucous membrane and the loss of natural light pink color as a result of anemia associated with copper deficiency (Aitken, 2008; Hefnawy & El-khaiat, 2015; Naji, 2017).

A total of 153 (37.5%) animals showed various clinical signs of mineral deficiency in sheep such as: alopecia, parakeratosis, diarrhea, pale mucous membrane, lacrimation and ataxia. Other previous related studies (Abd El-Raof & Ghanem, 2006; Aitken, 2008; Constable et al., 2016) have been documented similar findings.Primarily, rough hair coat condition and/or wool abnormalities were usually related to deficiency of copper, zinc and cobalt (Pond et al., 2004). Alopecia and steely wool might be attributed to defective keratinization. The pathway of alopecia in zinc deficient animal mayattribute to the reduction of the follicular epithelium capacity to produce fiber (Radostits et al., 2006). Alopecia was the most common symptom that has observed in the current study especially in ewes with low zinc levels. "Parakeratosis" as highlighted in the current study might be related to the importance of zinc as an activator of some enzymes requisite in carbohydrate, lipids, protein and nucleic acid metabolisms (Suttle, 2010). Clinically observed parakeratosis and hyperkeratosis in some cases are the main lesions. These pathological changes have been imputed to the involvement of zinc in cell replication of the skin (Wright & Spears, 2004). It is evident from the ongoing research that diarrhea occurs when levels of copper, zinc and cobalt fall to (0.13 ppm, 0.77 ppm, 0.39 ppm) respectively. Diarrhea is usually a major clinical finding in secondary copper deficiency associated with molybdenosis and may also occur due to the atrophy of intestinal villi. The inhibitory role of copper in the regulation of intestinal motility leads to disturbances in the gastrointestinal motility (Kaneko et al., 2008).

Table 2: Clinical signs of sheep affected with Cu, Zn, Co, Mg and Ca deficiency.

Sings	% of affected sheep	Cu Mean ± S.E	Zn Mean ± S.E	Co Mean ± S.E	Mg Mean ± S.E	Ca Mean ± S.E
Alopecia	47%	0.15±0.03	0.71±0.03	0.52±0.01	0.71±0.06	14.35±1.25
Parakeratosis	19%	0.14±0.00	0.77±0.05	0.43±0.00	0.58±0.04	12.98±2.01
Diarrhea	14%	0.13±0.00	0.77±0.06	0.39±0.00	0.65±0.07	15.03±2.14
Pale M.M	88%	0.15±0.00	0.73±0.03	0.56±0.01	0.68±0.05	13.56±3.08
Lacrimation	9%	0.12±0.00	0.82±0.09	0.36±0.00	0.59±0.03	14.24±1.02
Ataxia	8%	0.12±0.01	0.76±0.12	0.35±0.00	0.62±0.03	13.86±2.13

$p > 0.05$

Table 3: Body Condition and Hair coat condition score of all sheep

Criteria	Affected Sheep (NO.=330)	Control (NO.= 148)
Body condition score Mean ± S.E	2.12±0.04^{b}	2.92±0.05^{a}
Rough hair coat condition (%)	88	0

^{a,b,} values in different superscripts within row were significantly different at ($p<0.05$)

Table 4: Hematological values of control ewes and affected with minerals Deficiency

Criteria	Control (Mean ±SE)(NO.= 148)	Affected Sheep (Mean± SE)(NO.= 330)
RBC10^6/ml	7.63±.35^{a}	6.42±0.14^{ab}
PCV. %	25.40±.58^{a}	19.94±0.36^{b}
Hb g/L	8.75±.41^{a}	6.76± 0.14^{b}
MCV fl	35.06±1.73^{a}	31.99±0.72^{ab}
MCH pg	11.92± 0.63^{a}	10.70±0.23^{ab}
MCHC%	35.41±1.84^{a}	33.8±0.49^{ab}
WBC10^3/ml	6.73±.23^{a}	6.26± 0.16^{ab}

^{a,b,} values in different superscripts within row were significantly different at ($p<0.05$)

Table 5: Mean value of trace elements in all examined ewes

Minerals	Control (Mean± SE)	Affected Sheep (Mean± SE)
Cu ppm	0.45± 0.01^{a}	0.13±0.01^{b}
Zn ppm	1.35±0.59^{a}	1.13±0.07^{b}
Co ppm	0.78±0.02^{a}	0.42±0.019b
Mg ppm	1.63±0.04^{a}	0.58±0. 024^{b}
Ca ppm	20.81±0.33^{a}	13.97±0.49^{b}

^{a,b,} values in different superscripts within row were significantly different at ($p<0.05$)

Furthermore, affected animals showed signs of ataxia when level of copper in serum was as low as 0.12±0.01. Enzootic ataxia, lack of balance and atrophy of hind legs with the evidence of the neurological signs of lambs were mentioned by (Rizwana et al., 2016). Thus, the deficiency of copper in animal diet causes damages to nerves due to its role in the synthesis of phospholipids. Ataxia also occurs due to deficient myelination in the spinal cord followed by lesion appearance in parts of the cerebral white matter and brain stem (Silva et al., 2014).

The decrease in body condition score recorded in the affected sheep compared to the control group could be due to a loss of appetite which eventually led to less feed consumption and consequently loss in body weight. The results of hair coat condition score were often related to those observed in body condition score. All affected sheep had poor hair condition score. This notion achieved consensus among recent studies (Battini et al., 2014). Thus, symptoms like poor hair coat could be attributed to a mineral deficiency that subsequently led to induced nutritional and health problems. The researchers have documented that the main causes of the poor hair coat in sheep and goats are associated with health and nutritional disorders (Smith & Sherman, 2011; Lengarite et al., 2012).

Results of the blood picture from the present study indicated a significant variation of erythrocyte count and significant decrease values of packed cell volume and hemoglobin. This decrease might be due to disturbance in the regular metabolism of iron as copper deficiency decreases the absorption of iron, releasing of iron from body stores and utilization in hemoglobin synthesis (Pond et al., 2004; Abd El-Raof & Ghanem, 2006). However, this might be associated with a reduction of ceruloplasmin enzyme in serum which transports iron from stores cells in intestine & liver to transferase in plasma. Transferase is an enzyme which transports iron to bone marrow to synthesis Hb, and additionally reduce the liberation of iron from normally broken erythrocytes (Sharma et al., 2005; Kaneko et al., 2008). In the current study, normochromic normocytic anemia has been detected in the blood of the affected sheep.

The low serum values of copper suggest that the ewes were supplemented by a low concentration diet or (Pugh & Baird, 2012; Smith, 2014). Moreover, increase intake of calcium and phosphorus is the main factor that subsequently leads to cause zinc deficiency in sheep by decreasing the absorption of zinc. High calcium diet (enriched in legumes), high phosphorus (grain) feed supplements, increasing of soil pH higher 6.5 and elevation of the soil fertilization by phosphorus and nitrogen also be attributed to induce a zinc deficiency (Abd El-Raof & Ghanem, 2006). Based on the clinical investigation in the present study, most farmers provide high concentration of grains only to sheep, particularly barley, and this could subsequently induce a zinc deficiency. Deficiency of cobalt in sheep is attributed to the deficient level of cobalt in the diet especially when livestock are depending on forages coupled those originally receiving a limited amount of cobalt from soil (Aitken, 2008). Result of means value of minerals in the present study is in tandem with the result reported by (Pugh & Baird, 2012).

Overall, the low serum levels of mg and ca in the affected ewes indicate a poor feed supplementation of ewes with Ca and Ca+Mg in conjugation with consuming high levels during late gestation and early lactation period. Mineral supplementation had a beneficial effect on energy balance regulation in ewes at lambing (Ataollahi et al., 2018). Furthermore, the low mineral concentrations in blood of ewes suggest a considerable number of animals in Basra region might be at the risk of a subsequent sub-clinical or clinical hypocalcaemia and hypomagnesaemia. Elements concentration in soil did not give an indicator of the spreading of calcium and magnesium deficiency; although such pasture might the susceptibility of these cases (Edwards et al., 2018). Moreover, forages had a complex minerals compositions meaning that grazing ewes may have an increased risk of direct or induced hypocalcaemia or hypomagnesaemia (Masters et al., 2017).

CONCLUSION

The serum levels of mineral bio-markers were more sensitive to the reproductive stage than to the level of concentrate supplementation. In general, the use of supplementation is recommended to support the maintenance of mineral status in homeostasis and the reproductive performance of ewes in Basra. Thus, the supplementation is a viable and extremely important tool, primarily in periods of high nutritional requirements of the animals, such as during the final trimester, at birth and in early lactation. More studies should be attempted to evaluate the role of the trace minerals concentration in oxidative stress of the local small ruminants.

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CONFLICT OF INTEREST

No conflict of interests is declared by author for the contents in this manuscript.

AUTHORS CONTRIBUTION

Wessam Monther Mohammed Saleh has designed and carried out the experiment and prepared the draft manuscript.

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