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Assessment of Copper Deficiency In Lambs: A Study From Salah Al-Din City

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ABSTRACT

Copper is a trace element that contributes to metabolical and neurological actions of metalloenzymes. and metalloproteins. Deficiency can lead to severe health issues, including anemia and neurological disorders, with two types identified: In atherosclerosis low dietary. copper provokes Primary Copper Deficiency (PDC) and Secondary Copper Deficiency (SDC) acts as a result of interaction. with other minerals. Copper transporters, when underregulated, lead to disparate overall health. Copper deficiency decrease production. in sheep in a big way thus needs to be checked regularly and the animals supplemented. with the mineral. Early detection and management of the issues are underlined by this study, especially targeting the soil and forage mineral content. This study assessed 150 lambs in Salah al-Din city from June 2 to October 15, 2024, categorized. into three groups: a control group (50 healthy lambs), a subclinical group (50 lambs with insufficient copper), and a clinical group (50 lambs with copper deficiency). Blood samples were collected via the external jugular vein and centrifuged to prepare. serum, which was stored at -20˚C. Biochemical analysis measured. levels of copper, iron, calcium, phosphorus, superoxide dismutase. (SOD), and ceruloplasmin. Thus, the means of copper, iron, calcium, phosphorus, ceruloplasmin, SOD in the studied lamb groups were significantly different, p<0.000. The clinical group consistently exhibited the lowest levels for these parameters: copper – 0,38±0,193 µg/L; iron – 105,39±24,462 µg/dL; calcium – 4,76±2,076 mg/dL; phosphorus – 4,56±0,559 mg/dL; ceruloplasmin – 6,60±4,180 mg/dL; and SOD – 34,20±11,938 UI The post hoc tests also revealed that there was significant difference between the groups. which showed the serious effects of copper deficiency in growing lambs. this study, conclude the most numerous nutrition problems of lambs in Salah Al-Din are deficiency. of copper, which reflected the overall health and productive performance and cause anemia. and developmental disorders

Keyword: *congenital diaphragmatic hernia, newborns, surgical correction, minimally invasive methods, prognosis, intensive therapy, neonatal surgery*

INTRODUCTION

Copper is an essential microelement. for animals, since it is necessary for the function and synthesis of metalloenzymes and metalloproteids. that are necessary for metabolism, target tissular formation, and neurological processes; deficiency of this element refers to grave dangerous states, due to its negative influence on all the animal's vital. functions (Gale and Aizenman, 2024). Connective tissue and bone formation, stimulation. of lysyl oxidase enzyme which are required for cross-linking of collagen and elastic fibers,Copper is also involved in the neuronal function, myelination and neurotransmitter. synthesis;Copper deficiency and excess leads to serious neurological problems like Menkes and Wilson's diseases respectively (Aggett, 2023) (Gale and Aizenman, 2024). There are two types of copper deficiency: Primary Copper Deficiency (PDC) from low dietary copper derived from .copper-deficient soils leads to anemia and neurological disorder. Secondary Copper. Deficiency (SDC) occurs from reduced copper .bioavailability resulting from dietary interactions with minerals and other factors which interfere with the efficiency of copper .activity and may cause grave blood conditions (Yilmaz and Atan, 2024)(Gupta, 2024). ABC transporters like ATP7A and ATP7B maintain specific copper .concentrations and dysregulation has damaging effects; shellfish, nuts, grains, and other sources are significant for copper uptake which is .regulated by other nutrients such as zinc (Locatelli and Farina, 2025). In sheep rearing, copper deficiency leads to significant production losses that .require close monitoring to avoid deficiency and toxicity, .Copper sulfate supplementation improves the health and fertility of rams (Borobia et al., 2022). Cu deficiency in lambs creates serious neurological .disorders, indistinguishable at times from other poor dietary element deficiencies, and is exacerbated by factors in the soil that reduce Cu ,bioavailability. Soil and forage mineral consultation together with early diagnostics and intervention is crucial to avoid destructive outcomes and enhance results in affected species(Hill and Shannon, 2019). The study definitively focuses on identifying copper-related elements by measuring Fluorine, Magnesium, Zinc, Phosphorus, and .Calcium, evaluating complete blood count, and assessing enzymes such as ceruloplasmin and superoxide dismutase (SOD)

MATERIAL AND METHOD

Experimental design:

This study rigorously examined 150 lambs across Salah al-Din city (Tikrit - Al-Alam - Samarra - Baiji) from June 2 to October 15, 2024, including fifty in a control group, fifty with subclinical copper insufficiency, and a .clinical group with copper deficiency, with ages from one day to one year and weights ranging from 2 to 10 kg. Three groups of 50 lambs each: Control (C) group: 50 healthy lambs; Subclinical group: 50 lambs with insufficient copper; Clinical group: 50 lambs with copper deficiency.

Samples Collection:

Blood samples was taken from the .external jugular vein using a syringe to determine absolute .biochemical profile of the lambs; serum was prepared by centrifuging the blood at 3000 rpm for 15 min and was stored in 0.5 mL Eppendorf tubes at -20˚C till further use.

Boichemical Assy analysis

in vitro analysis was determined by .measuring copper, iron, calcium, Phosphorus, SOD and Ceruloplasmin.

Data analysis

Data collected in this study was well .aggregated, synthesized in the Microsoft Excel 2010 and analyzed by IBM-SPSS 26. To confirm normality, the Shapiro-Wilk test was employed and as a result, a parametric test was applied which used mean and standard deviations with one-way .ANOVA with post hoc test at a significance level of P value ≤ 0.05.

Results

The comparison of the .cupper mean levels among the studied groups was demonstrated in table (1) which revealed a statistically significant difference (p=0.000). The mean cupper level among the clinical group (0.38±0.193 µg/L) was the lowest in comparison to the other groups and the real difference was found between each groups

Table (1): Cupper

Cupper µg/L	Controls	Subclinical	Clinical	p-value*
	Mean ±SD	Mean ±SD	Mean ±SD	
	16.40±4.033 A	3.82±1.881 B	0.38±0.193 C	**0.000**

**One-Way ANOVA; Post hoc test =Similar letters means no significance while different letters means significant difference*

. Table (2), which compared the Iron mean levels among the groups under study, showed a statistically significant difference (p=0.000). When compared to the other groups, the clinical group's mean Iron level (105.39±24.462 µg/dL) was the lower , and there was a noticeable difference between each group.

Table (2): Iron

Iron µg/dL	Controls	Subclinical	Clinical	p-value*
	Mean ±SD	Mean ±SD	Mean ±SD	
	170.03±31.885 A	138.15±29.175 B	105.39±24.462 C	**0.000**

**One-Way ANOVA; Post hoc test =Similar letters means no significance while different letters means significant difference*

When comparing the mean Calcium levels among the groups being studied, table (3) reveals a statistically significant difference (p=0.000). The clinical group had the lowest mean Calcium level (4.76±2.076 mg/dL) compared to the other groups, and each group differed noticeably from the others

Table (3): Calcium

Calcium mg/dL	Controls	Subclinical	Clinical	p-value*
	Mean ±SD	Mean ±SD	Mean ±SD	
	10.42±0.836 A	7.02±3.267 B	4.76±2.076 C	**0.000**

**One-Way ANOVA; Post hoc test =Similar letters means no significance while different letters means significant difference*

The difference in the mean Phosphorus level among the studied groups was statistically significant at p=0.000. By performing the post hoc test for the significant difference, it was found that the levels among the clinical (4.56±0.559 mg/dL) and subclinical (4.65±0.675 mg/dL) were significantly lower than that among the control group, but with no difference between them, as shown in table (4).

Table (4): Phosphorus

Phosphorus mg/dL	Controls	Subclinical	Clinical	p-value*
	Mean ±SD	Mean ±SD	Mean ±SD	
	5.92±0.875 A	4.65±0.675 B	4.56±0.559 B	**0.000**

**One-Way ANOVA; Post hoc test =Similar letters means no significance while different letters means significant difference*

There was a statistically significant difference (p=0.000) in the Cupper in soil mean levels across the groups under investigation, as shown in Table (3.5). The clinical group had the lowest mean Cupper in soil level (0.92±0.505 mg/kg) when compared to the other groups. The post hoc test showed a significant difference between each group

Table (7): Ceruloplasmin mg/dL

Ceruloplasmin (mg/dL)	Controls	Subclinical	Clinical	p-value*
	Mean ±SD	Mean ±SD	Mean ±SD	
	27.20±4.184 A	18.82±11.138 B	6.60±4.180 C	**0.000**

**One-Way ANOVA; Post hoc test =Similar letters means no significance while different letters means significant difference*

The comparison of the SOD mean levels among the studied groups was demonstrated in table (8) which revealed a statistically significant difference (p=0.000). The mean SOD level among the clinical group (34.20±11.938 UI/g) was the lowest in comparison to the other groups and the real difference was found between each groups.

Table (8): SOD UI/gram

SOD UI/gram	Controls	Subclinical	Clinical	p-value*
	Mean ±SD	Mean ±SD	Mean ±SD	
	126.00±17.448 A	86.62±48.037 B	34.20±11.938 C	**0.000**

**One-Way ANOVA; Post hoc test =Similar letters means no significance while different letters means significant difference*

DISCUSSION

Copper is vital for animal health, supporting growth, pigmentation, and immune function, and its levels can be easily evaluated through blood tests(Borobia et al., 2022).The low copper levels can lead to deficiencies, impacting health and productivity. The following sections explore the implications of low copper levels, the effects of copper toxicity, and the balance required for optimal copper levels in sheep.

The results of the current study found The clinical group's copper levels were significantly lower than those of the other groups. This findings agree with The study conducted by Ježek et al., 2024 found that low copper levels correlate with reduced concentrations of important biochemical parameters such as beta-hydroxybutyrate, calcium, and albumin, which are essential for maintaining health. In regions with extensive pastures, sheep often suffer from mineral deficiencies, including copper, due to inadequate supplementation. This deficiency can be exacerbated by the lack of awareness among farmers about the need for mineral and vitamin supplements. Also agree with study conducted in Bahia, by Fontes et al., 2020 sheep affected by enzootic ataxia exhibited significantly reduced serum copper levels, highlighting the critical role of copper in preventing this condition. simlarity Helmer et al., 2021 found Clinical signs of copper deficiency in lambs include emaciation, anemia, growth retardation, and wool discoloration. These symptoms were observed in a study of a small sheep herd in Germany, where copper deficiency was compounded by other mineral deficiencies.

In this study, it was found that lambs with copper ,deficiency had iron deficiency compared to normal lambs this agree with Jensen et al., 2019 and Ježek, Starič, Veren, et al., 2024 found Copper deficiency is associated with significant .changes in hematological and biochemical parameters. In sheep, low copper levels correlated with reduced concentrations of hemoglobin, albumin, and white blood cells, indicating anemia and compromised immune function .Copper deficiency can lead to anemia, which is often characterized by low iron levels in the blood. Also Klevay, 2019 and Wu, Xiong and He, 2021 reported Copper plays a role in the absorption and transport of iron. It is a component of ceruloplasmin, a protein that .oxidizes iron to its ferric form, which is necessary for binding to transferrin and subsequent transport in the bloodstream. Similarity Jayalakshmi et al., 2020 showed that copper deficiency can lead to .hypoferremia (low iron levels in the blood), as seen in cases of haemonchosis in sheep, where both copper and iron levels were significantly reduced. As well as Jensen et al., 2019 suggested the deficiency of copper can lead to mitochondrial dysfunction in erythropoietic cells, affecting the differentiation of red blood cells and contributing to anemia.

In this study found that lambs with copper deficiency .had low calcium levels compared to normal lambs. This findings consistent with study conducted by Meng et al., 2024 found that copper supplementation improved the apparent absorbability of calcium, suggesting that copper deficiency might impair calcium .absorption and metabolism. Also Ježek, Starič, Veren, et al., 2024 found that lambs with low copper levels also exhibited significantly lower calcium concentrations in their blood serum. This suggests a potential link between copper deficiency and reduced calcium levels. As well as Jin et al., 2023 reported that sheep on a copper-deficient diet showed deficiencies in other minerals, including calcium, which could adversely affect organ function and energy metabolism. Similarty Shen and Song, 2021 found that copper deprivation in sheep not only affects copper levels but also disrupts other physiological and biochemical parameters, potentially impacting calcium metabolism.

Additionally, the results of the current study found that lambs with copper deficiency had low phosphorus levels compared to normal lambs. This agree with Pereira et al., 2019 conducted a study on Somali lambs found that mineral requirements, including phosphorus, are crucial for growth and maintenance. Copper deficiency could potentially alter these requirements by affecting the overall mineral balance . Also Meng et al., 2024 demonstrated that a copper-deficient diet significantly decreased the apparent absorbability of phosphorus, among other minerals. This suggests that copper plays a role in facilitating the absorption of phosphorus in the digestive system.

Ceruloplasmin serves as the primary transport protein for copper in the bloodstream, produced by .both copper and proteins in the liver. It exhibits a comparable function to superoxide dismutase, effectively neutralizing superoxide anion free radicals and preventing lipid peroxidation(Liu et al., 2022). Copper is a crucial trace element involved in various biological processes, including the .functioning of enzymes like ceruloplasmin and SOD, which are essential for antioxidant defense and iron metabolism. Copper deficiency disrupts these processes, leading to reduced enzyme activity and associated physiological impairments(Tsang, Davis and Brady, 2021).

The .results of the current study found that lambs with copper deficiency had low Ceruloplasmin level compared to normal lambs. This results consistent with Copper deficiency leads to significantly lower ceruloplasmin levels in affected animals compared to healthy ones. This is evident in Min et al., 2022 and Shen & Song, 2021 on Kazakh sheep and Chinese Merino sheep, where .serum ceruloplasmin was notably reduced in copper-deficient groups. Additionally study conducted in iran by Moazzemi et al., 2023 indicated a strong positive correlation, signified a substantial association whereby increased in copper levels correspondingly elevated ceruloplasmin levels.

Oxygen free .radicals, constituting 95% of total free radicals in the animal body, necessitate a dynamic homeostasis between .their production and elimination to maintain health; an excess over the scavenging capacity disrupts redox equilibrium, causing macromolecular damage and potentially leading to disease .progression (Sadiq, 2023). The antioxidant system is crucial for neutralizing harmful oxygen free radicals, encompassing both enzymatic and non-enzymatic mechanisms that safeguard against oxidative damage. Copper is vital for animal growth and metabolism, significantly. boosting the antioxidant function through its role in various enzymes(Kotha et al., 2022). The findings of this study clearly indicated that the levels of SOD in the blood of clinical group was markedly lower to those in control group. This agree with Wu et al., 2021 found lambs with copper deficiency had low SOD levels. Also Emam et al., 2025 found that elevating the Cu levels in vivo significantly enhanced synthesis and improved SOD production. Smilartity Conti et al., 2023 reported that the combination of copper supplementation with organic sulfur sources in lambs resulted in increased serum CP activity, suggesting that the source of Cu and its interaction with other minerals can influence CP activity.

CONCLUSION

This study has been able to provide an understanding of the nutritional problem of copper deficiency disease in lambs. within Salah Al-Din city. The study thus supports the fact that copper deficiency is a common problem affecting the health and performance of lambs with attendant clinical signs, including anaemia, weakened immunity and developmental abnormalities.

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