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Micronutrient Changes in Some Tissues of Copper Deficient Rats

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Abstract: The present study was carried out to investigate the effect of copper deficiency on tissue level of some minerals (Na, K, Ca, P, Cu, Fe, Mg, Zn, Mn) in the growing male rats divided into two groups of rats fed either a copper deficient (0.06mgCu/Kg) or a Copper adequate diet. Both groups of rats were fed for six weeks. Copper deficiency decreased the final body weight of the rats by 21% compared to copper- adequate control rats. The haematological parameter i.e packed cell volume (PCV) and white blood cell (WBC) count of the copper deficient rats were significantly reduced compared to the control rats. Tissue content of nine elements (potassium, sodium, calcium, magnesium, manganese, copper, zinc, iron and phosphorus) were measured in the liver and the kidney of both group of rats. In copper deficiency, however, significant increase ($p<0.05$) in the tissue content of some elements (Zn, Ca, Fe, Na, K, Mn) were observed compared to the control, while Cu, Mg and phosphorus level were lowered compared to the control. The result implies that copper deficiency in rats could be detrimental to growth, mineral metabolism and hence the overall well being of the rat.

Key words: Copper deficiency, rats, haematological parameters, minerals

Introduction

The role of essential trace elements in post-natal development is receiving increased attention. They are required for the proper functioning of the body (Underwood, 1981). Copper is essential for plant and animal nutrition. It is a component of many protein and also plays a vital role in many enzyme system.

Copper deficiency symptoms in animals include low plasma copper and ceruloplasmin levels, anaemia, demyelination, skeletal defects (McDonald *et al*., 1995) It has been previously reported that copper deficiency is also associated with decreased cytochrome- c- oxidase and supper oxide dismutase in the heart and liver of rats. (Rossi *et al*., 1998).

Copper deficiency during the critical postnatal period of development could have adverse effect on micronutrient metabolism which may exert a direct effect on metabolic processes. An additional information is sought on the interplay between micronutrients in copper deficiency and on mineral content of tissues in copper deficiency. The experiment described here is designed to examine the micronutrient levels in some tissues of growing rats.

Materials and Methods

Ten male white albino rats *(Rattus norvegicus*) were divided into groups each containing five animals and housed in plastic cages of stainless steel wire top and bottom. The animals were acclimatized for 24 hours before introducing the experimental diets. The two groups were maintained on (a) Control diet containing (adequate copper) and (b) Copper deficient diet. The composition of the diets was as described by Rossi *et al*. (1998). All reagents were of analytical grade and products of BDH Limited and Sigma chemical company, England. The feeding trial lasted six weeks.

Tissue preparation: At the end of the feeding period, rats were anaesthesized and sacrificed by cervical dislocation. Blood was drawn from the heart by cardiac puncture using a sterile syringe and needle. Blood was drawn into heparinized capillary tube for PCV analysis. Blood serum was obtained and stored for haematological parameters. The liver and kidneys (decapsulated) were removed, drained of blood and weighed. Each tissue was ashed and dissolved in 10 percent HCl and made upto 100ml standard flask with distilled water. The mineral content of each tissue were analyzed using atomic Absorption spectrophotometer AOAC (1990).

Phosphorus was determined colorimetrically using vanadonomolybdate method.

Results and Discussion

Table 1 shows the mean initial and final body weight of rats in the control and copper deficient groups.

In the present study, the average weight gained by the copper deficient rat was significantly (21%) less than that of the control. The reduction in weight could be due to depigmentation of fur of the rats observed in this study. Copper has been shown to be necessary for normal pigmentation of fur, wool and hair, Tyrosinase, the enzyme required for the incorporation of hydroxyl into melanin pigment is activated by copper (Underwood, 1981; Thompson and Fowler, 1990).

Table 1: Average mean initial and final body weight of animals

	A	B
Final body weight(g)	88.84±2.32	70.01±1.15
Initial body weight (g)	40.73±4.31	37.24±2.51

Table 2: Haematological parameters

	Packed Cell Volume (PCV)	White blood Cell(WBC) count
A[1]	46.50±0.21	2950±70.71
B[2]	41.50±0.71	2650±21.21

A[1] ----------> Control rats.

B[2] ----------> Copper Deficient rats

Table 3: Mineral content of the kidney

Minerals	Control (ppm)	Copper deficient (ppm)
Iron	40.13±1.80[a]	45.06±0.61[b]
Zinc	229.42±13.97[a]	358.77±34.04[b]
Manganese	26.12±8.020[a]	3.68±0.87[b]
Copper	28.55±1.87[a]	24.29±0.99[b]
Magnesium	53.27±3.24[a]	49.11±2.33[b]
Calcium	41.47±3.58[a]	82.43±8.13[b]
Potassium	411.43±7.49[a]	434.93±7.83[b]
Sodium	497.43±76.85[a]	605.08±16.81[b]
Phosphorus	165.40±4.50[a]	164.00±0.30[b]

Values are means of 5 determinations ± SD.

Means not bearing the same superscript are different significantly ($p<0.05$).

Table 4: Mineral contents of the liver

	Control (ppm)	Copper-deficient (ppm)
Iron	16.34±2.24[a]	19.80±0.44[b]
Zinc	90.07±21.32[a]	153.4±29.17[b]
Manganese	7.43±3.08[a]	8.80±1.58[b]
Copper	11.90±0.08[a]	9.23±0.69[b]
Magnesium	41.96±7.10[a]	51.37±4.47[b]
Calcium	6.03±3.98[a]	41.05±11.90[b]
Potassium	161.9±26.30[a]	192.97±5.22[b]
Sodium	183.93±2.73[a]	212.85±53.18[b]
Phosphorus	105.72±1.16[a]	142.59±4.51[b]

Values are means of 5 determinations ± SD.

Means not bearing the same superscript are different significantly ($p<0.05$).

Table 2 shows the haematological parameters. A significant decrease ($P<0.05$) in the PCV value of copper deficient rats compared to the control was observed. Copper has been found to be essential for the absorption of iron and for haemoglobin formation. Hence too low a copper status results in iron deficient anaemia which is characterized by a low Haemoglobin level (Maurice *et al.*, 1997). It may be that copper is needed for the formation of normal bone marrow necessary for the formation of red blood cells. The white blood cell (WBC) count for the copper deficient is significantly lower ($p<0.05$) than that of the control. It has been reported that white blood cell count is four times richer in copper than red blood cells. The reduced WBC count herein observed in this study may cause infection (Percival, 1998).

Table 3 and 4 shows the mineral content of the liver and kidney of the copper deficient and control rats. The Iron status of the copper deficient rat is significantly higher than that of the control in the kidney and the liver. Copper is needed to facilitate iron absorption and mobilization; therefore, reduced copper in the diet may hinder absorption and hence increased level of iron in these tissues.

Similarly, significant increase was observed in the calcium of the liver in copper deficiency compared to the control. This may be because of increased absorption of calcium, serum cholesterol has been found to be increased in copper deficiency. (Maurice *et al.*, 1997). This is carried via the blood plasma to the liver and kidney where it is converted into 1, 25, dihydoxy, cholecalciferol which facilitates calcium absorption (Nelson and Cox, 2000).

The phosphorus status in the liver is significantly higher than the control and this maybe due to the fact that calcium and phosphorus metabolism are interwoven. Since calcium and phosphorus are required for bone mineralization. Excess phosphates has been shown to hinder iron absorption (Channey *et al.*, 1979) which may probably account in part, for the high content of iron observed in this study.

The balance between calcium, phosphorus and Iron is very important in micronutrient metabolism. The potassium and sodium concentration in the liver and kidney of copper deficient rats were significantly higher than the control. Aldosterone synthesis had been reported to increase in copper deficiency(Bell and Sly , 1976).

This increase in aldosterone may therefore increase the reabsorption of sodium and potassium from the kidney tubule. Thus facilitating the secretion of sodium and potassium into the intestine at high rate. This may adversely affect acid-base balance and hence osmotic regulation. It has been postulated that calcium interacts with membrane phospholipids leading to an increase in membrane permeability to sodium and potassium (Turnlund, 1999).

The Magnesium and manganese content of the copper deficient rats varies in the organs studied. These two elements act as activators of many enzymes involved in energy production, and protein metabolism.

In the kidney, there was no significant difference in the Magnesium content of the copper deficient rat while a low manganese content was observed compared to the control and this reduction could affect the activity of those

enzymes that requires manganese for activation. The copper status in the copper deficient diet is significantly low compared to the control in the liver and kidney due to the low dietary intake.

The zinc status is higher in these tissue in copper deficiency due to the fact that reduced copper in the diet enhanced zinc absorption. A high intake of zinc induces the synthesis of copper binding ligand, metallothionein in the mucosal cell. This protein sequesters copper and hence decreases its absorption.

Low copper content of tissues may cause oxidative stress due to the reduction in activity of superoxide dismutase an enzyme whose role is vital in respiration. In summary, this study shows that copper deficiency in rats affects mineral content in tissues and since these minerals acts as activators of enzymes in most biochemical reactions, this deficient state could have adverse effect on mineral metabolism and hence could be detrimental to health.

References

AOAC, 1990. Official methods of Analysis 15th ed. Washington D.C: Association of official Analytical Chemists.

Bell, F.R. and J. Sly, 1976. The assessment of sodium appetite in calves using operant conditioning procedures. J. Physiol., 263: 178-179.

Channey, M.S., M.L. Ross and J.C. Witschi, 1979. Nutrition; minerals, 9th ed., pp: 139-227.

Maurice, E.S., A.O. James and S. Moshe, 1997. Specific dietary component In: Modern Nutrition in Health and disease (Williams and Wilkins Ed.) USA Vol. 1, 8th ed., pp: 156-238.

McDonald, P., R.A. Edwards, J.F.D. Greenhalgh and R.A. Morgan, 1995. Animal Nutrition; Minerals; 5th ed., pp: 97-121.

Nelson, D. and M.M. Cox, 2000. Lehninger Principle of Biochemistry 3rd ed. Worth Pub. New York. pp. 611.

Percival, S.S., 1998. Copper and Immunity. Am. J. Clin. Nutr., 56: 1064S-1068S.

Rossi, L., G. Lippe, E. Marchese, A. De-Martino, L. Marvelli, G. Rotillo and M.R. Ciriolo, 1998. Cytochrome-c-oxidase problem in heart. Mitochondria of copper deficient rats. Biometals, 11: 207-212.

Thompson, J.K and V.R. Fowler, 1990. The evaluation of minerals in the diets of farm animals. In feed stuff evaluation. Wiseman & Cole (eds.) London, Butterworth, pp: 235-259.

Turnlund, J.R. 1999. In Shils, M.eds.Nutrition in Health and Disease ,9th edn. Baltimore:Williams and Wilkins, pp: 241-252.

Underwood, E.J., 1981. The mineral nutrition of domestic animals. Nat. Acad. Sci., 41: 291-310.