

# **SUPPLEMENTATION IN LOW-COPPER DIET: ANALYSIS OF HUMERUS BIOMECHANICAL RESPONSE IN A RAT MODEL STUDY**

*Siemowit Muszyński<sup>1</sup>, Katarzyna Ognik<sup>2</sup>, Małgorzata Kwiecień<sup>3</sup>, Piotr Dobrowolski<sup>4</sup>, Ewelina Cholewińska<sup>3</sup>, Agnieszka Tomczyk<sup>5</sup>, Katarzyna Kwiatkowska<sup>4</sup>, Natalia Kowal<sup>1</sup>, Marta Ejtel<sup>1</sup>, Agnieszka Leus<sup>1</sup>, Estera Kamińska<sup>4</sup>*

<sup>1</sup>Department of Physics, Faculty of Production Engineering, University of Life Sciences in Lublin, Akademicka 13, 20-950 Lublin, Poland

<sup>2</sup>Department of Biochemistry and Toxicology, University of Life Sciences in Lublin, Akademicka 13, 20-950 Lublin, Poland

<sup>3</sup>Institute of Animal Nutrition and Bromatology, University of Life Sciences in Lublin, Akademicka 13, 20-950 Lublin, Poland

<sup>4</sup>Department of Comparative Anatomy and Anthropology, Maria Curie-Skłodowska University, Akademicka 19, 20-033 Lublin, Poland

<sup>5</sup>Department of Animal Physiology, University of Life Sciences in Lublin, Akademicka 12, 20-950 Lublin, Poland

Copper is an essential metal micronutrient linked to bone metabolism. Copper-poor diet leads to bone loss and reduced bone mass, resulting in a reduction of its mechanical strength and increases the risk of fractures. A traditional dietary form of Cu presents in supplemental mineral mixture is inorganic copper sulfate or carbonate. Recently, the use of elements in form of nanoparticles has been considered as an alternative to inorganic forms. Copper nanoparticles at smaller concentrations can promote animal growth and performance. However, there are no scientific analyses involving the use of Cu nanoparticles in animal feeding and their effect on the bone mechanical properties.

The objective of the present study was to investigate the geometrical, biomechanical and densitometric traits of the humerus in growing male rats given Cu in two different chemical forms (carbonate and nanoparticles) at the reduced level of 50% of daily recommended dose ( $3.25 \text{ mg} \cdot \text{kg}^{-1}$  of the premix). It is hypothesized that the use of Cu in a more assailable form of nanoparticles might improve the development of the skeletal system, even if it is administered at the reduced level.

Twenty four healthy male, albino Wistar rats were randomly allocated to three dietary treatments: the control CONT group fed without additional Cu

supplementation in premix, the CuSalt group fed with lowered level of inorganic copper as cooper carbonate in premix and the CuNano group fed with lowered level of copper as nanoparticles. The experiment lasted 4 weeks.

Our study showed that supplementation with  $3.25 \text{ mg}\cdot\text{kg}^{-1}$  of Cu irrespective of its source decreased bone length, bone tissue density, Young modulus of elasticity and increased work to fracture, toughness, yield strain, ultimate strain and yield stress as compared to the rats whose diet was devoid of copper (the control CONT group). There were no changes in bone mineral density, midshaft geometrical properties, ultimate load and ultimate stress. The altered values of Young modulus of elasticity, yield strain and yield stress prove that there was a change in bone elastic properties after copper inclusion. However, bone mineral density, an important indicator of bone strength, did not differ between copper-supplemented and Cu-deprived control animals. Thus, alteration in bone susceptibility to the elastic deformation could be related to alteration in bone collagen matrix structure. Furthermore, there were also some alteration in other calculated bone parameters which were Cu-source-depended. The most significant were an decrease of the bone weight, yield load and stiffness in CuSalt group when compared to the CONT group.

In summary, in our study, a selected biomechanical indicators showed that Cu given at reduced dose increased mechanical endurance of bone, however, more significant improvement was observe when cooper was supplemented in form of nanoparticles than in traditional form of cooper carbonate.

**Keywords:** COPPER, NANOPARTICLES, HUMERUS, MECHANICAL TESTING, RAT.