Determination of trace elements in organs and tissues of Zn-deficient mice by instrumental neutron activation and PIXE analyses ~Determination of trace elements in soluble proteins separated by two-dimensional electrophoresis~

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Abstract

The concentrations of trace elements in hepatic subcellulars and cytosolic protein of zinc deficient mice were determined in order to investigate the behavior and role of zinc and other trace elements.

Eight-week-old male mice of ICR strain were divided into two groups; one was fed with zinc deficient diet ($<1 \mu g/g Zn$), the other with control diet ($30 \mu g/g Zn$). After 3 weeks of treatment periods, their livers were removed. Two types of experiments were performed. In the first experiment, the liver samples homogenized with HEPES buffer which adjusted to pH 7.4 with KHCO₃ were centrifuged under differential conditions in order to separate into cellular fragments and 5 subcellular fractions, such as nuclear, mitochondrial, lysosomal, microsomal and cytosolic fractions. Each fraction was freeze-dried for instrumental neutron activation analysis (INAA). Concentrations of 11 elements, Na, Mg, Cl, Mn, Fe, Co, Cu, Zn, Se, Br, and Rb, were determined by INAA. In the second experiment, sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE) and two-dimensional electrophoresis (2-DE) were performed for cytosolic fraction of other mice. After electrophoresis, the gel was cut into protein bands and subjected to PIXE analysis.