ELEMENTAL ANALYSIS IN LIVER, ASCITES, AND BLOOD OF TUMOR-BEARING MICE

Y.-Y. Wei,* C. Chung

Institute of Nuclear Science, National Tsing Hua University, Hsinchu, 30043 (Taiwan, R. O. C.)

(Received June 1, 1992)

The contents of twelve minor and trace elements in liver, ascites, and blood of sarcoma-180 tumor-bearing mice were determined by instrumental neutron activation analysis at various tumor growing stages. Biological samples were irradiated by reactor neutrons and subsequently subject to direct analysis using a high-resolution HPGe γ-spectrometer. A statistically significant difference for the concentration of Na and Cl in all tumor samples relative to those in a normal control group is found. During the tumor growing period, concentration of Mg, K, Fe, Se, Rb, Cu, Zn, and Mn in liver and ascites of tumor mice vary with respect to the tumor size. Each elemental concentration and its biological function in the tumor mice is discussed and possible correlation to humans is analyzed.

The biological role of minor and trace elements in cancer has been extensively investigated in recent years. Some reports have explored the relations of trace elements with cancer in terms of carcinogenesis, indicating that some trace metal ions can interact with nucleic acids to influence base-pairing and conformation.1-4 Because the majority of trace elements are cofactors of enzymes, with magnesium, manganese, and zinc as examples in RNA and DNA polymerase, the deficient effects of these elements may cause somatic mutations, a consequence of base-pairing errors of frame-shift mutations by deletion, eventually leading to permanent cellular transformation.4

On the other hand, several investigations have also reported that sodium plays a role in mitogenesis and oncogenesis.5-9 Sparks et al. point out that cancer cells have a high intranuclear level of sodium compared to those in normal cells; they postulated that such an unusual level of sodium may be involved in cancer growth.10

It is possible that the concentration of minor and trace elements in biological fluids or tissues can be used as diagnostic or prognostic indicators for cancer bearing subjects. Statistically significant differences from the normal distribution of Ca, Cu, Fe, Mg, Mn, Ni, K, Rb, Se, Na, V, and Zn have been reported to occur in patients and animals with various forms of cancer, indicating that these minor and trace elements play an essential role in tumor pathology, although no explicit correlation in other types or classes of tumor were concluded.1,11-13

*Present address: Division of Radioisotopes, Nuclear Science and Technology Development Center, National Tsing Hua University, Hsinchu 30043, Taiwan ROC.
The presence of minor and trace elements in serum is routinely checked up clinically. In previous animal experiments, the investigating period is limited to only a part of life span of tumor-bearing subjects. Unfortunately, these results show only a partial evaluation that does not reflect the change of minor and trace elements in tumor during the whole life span of subjects. Thus, the problem of elemental concentration in tumor at various growing stages is worth investigating.

In this work, instrumental neutron activation analysis (INAA) is used as a quantitative method to determine elemental concentration in biological samples. It has two advantages in determining elemental concentration of analyzed samples — without chemical treatment of samples and simultaneous determination of multielement concentrations. In this study, we sampled the whole life span of sarcoma-180 tumor-bearing mice and utilize INAA method to analyze elemental concentration in the biological samples. The relation and effect between elemental concentration and tumor at various growing stages of tumor-bearing mice are correlated to tumor pathology; possible correlations of tumor-bearing mice with humans is discussed.

**Materials and methods**

*Tumored mice.* Male ICR mice about six to eight weeks old were purchased from Animal Resource Center of the College of Medicine, National Taiwan University, Taiwan. Water and food were fed ad libitum. The 0.1 ml Sarcoma-180 (S-180) tumor cells, supplied by the National Tsing Hua University, were routinely propagated by intraperitoneal injection every seven days. Ascites tumor of experimental animals were induced by injecting 0.1 ml S-180 tumor cells from 10-day old tumored mice. The life span of S-180 tumor-bearing mice is about 25–30 days. Six mice were assigned in each experimental and control group in this investigation.

*Sample preparation.* To avoid the physiological stress by environmental change, it is necessary to feed the mice in normal way for one week after purchase. Because the life span of S-180 ascites tumor-bearing mice is short, the collection of samples was arranged frequently from immediately after tumor transplantation to the very end of their life span. Healthy mice were also sampled simultaneously as a control group; the animals were killed after diethyl ether anaesthesia and dissected prior to sampling; the liver, ascites, and blood were singled out and put into 20 ml polyethylene vials. Samples were crushed in polyethylene vials with a pyrex glass pestle after freeze-drying for further homogenization. All glassware, polyethylene vials, and bags used in this work were precleaned with MERCK 4N nitric acid overnight followed by rinsing several times with deionized water to prevent material contaminations during pretreatment.