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Concentration of Rare Earth Elements, As, and Th in Human Brain and Brain Tumors, Determined by Neutron Activation Analysis

GUISUN ZHUANG,*[,1] YUNLU ZHOU,[2] HUIJUAN LU,[2] WEIDONG LU,[2] MANFANG ZHOU,[2] YINSONG WANG,[1] AND MINGGUANG TAN[1]

[1]*Shanghai Institute of Nuclear Research, LNAT, Academia Sinica, PO Box 800-204, Shanghai 201800, P. R. China; and* [2]*General Research Institute of Non-ferrous Metals, Beijing 100088, P. R. China*

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ABSTRACT

Toxic elements As and Th, six rare-earth elemental profiles of brain tumor tissues from 16 patients of astrocytomas (grade I–III), and normal human brain tissues of 18 male, age-matched autopsies serving as controls have been studied by radiochemical neutron activation analysis. P-204 [di(2-ethylhexyl) phosphate] extraction chromatography column was used for group separation of rare-earth element (REE) by one step. Compared with the normal brain tissues, the analytical results showed that the concentrations of Th, La, Ce, Gd, and Lu were significantly higher in tumor tissues ($P < 0.01$ or 0.001). The possible effects of REE on tumor cell were discussed.

Index Entries: Rare earth; human brain; brain tumor.

INTRODUCTION

In recent decades, heavy-metal pollution of the biosphere and the biological role of trace elements in human health and disease have attracted considerable attention in cancer research. Although the levels of rare-earth elements (REE) in normal brain tissues and their role in etiology

*Author to whom all correspondence and reprint requests should be addressed.

and pathogenesis of patients with brain tumor are unknown, the biological role of REE has been noted recently *(1)*. Abnormal REE accumulation in lung tissue of exposed workers was reported *(2)*. The effect of compounds of lanthanum, samarium, europium, and ytterbium on Hela culture has been studied *(3)*. The correlation between the accumulation of ^{147}Pm in bone and its possible mutagenic effect in organism has also been observed *(4)*. Trace element concentrations in human brain and the brain of patients with neurological diseases have been determined by NAA *(5–6)*. Arsenic has been implicated as a carcinogenic or cocarcinogenic agent in skin cancer and lung tumors. Low concentrations of arsenic in the form of arsenite may abolish the cancer-protecting effect of selenite *(7)*. ^{232}Th is an important naturally occurring radionuclide. It is radiotoxic as well as chemically toxic to human cell when present even at low levels *(8)*. Quite recently, the authors carried out investigations on the occurrence of minor and trace elements in human hair, tissues, and subcellular components of patients affected by brain tumor *(9,10)*. Simultaneous determination of REE, As, and Th in biological samples is often a difficult task because of their very low concentrations, usually at ng/g level or below, and because of potential matrix interferences. Radiochemical neutron activation analysis is, at the moment, the only technique capable of meeting the sensitivity requirements for REE assay in biological samples. In this paper, a previously reported radiochemical procedure of REE separation was partially modified and applied to REE determination in brain and brain-tumor tissues *(11)*.

MATERIALS AND METHODS

Collection and Pretreatment of Samples

Sixteen male malignant brain tumor tissues (astrocytomas grade I–III) were taken at neurosurgery in Hua Shan Hospital, Shanghai. The mean age was 48 yr (range 35–64 yr). Eighteen normal male brain tissues (range 35–60 yrs), obtained from autopsies after accidental death of a person who was proven to be in good health before the accident, served as controls. The cerebral cortical sections containing both gray and white matter were collected from the left hemisphere. To avoid external contamination, the surface of all tissue samples were cut off using a titanium knife; the samples were diced into 1–2 cm cubes on a clean bench and stored in a freezer below –30°C until taken out for analysis. All tissue samples were freeze-dried for 48 h, homogenized by brittle fracture technique in liquid nitrogen, and ground into fine powder. The ratio of freeze-dry-to-wet wt for each subject was determined.

Irradiation and Measurements

Usually 100–300 mg of dry tissue samples, blank and comparator standard prepared by high-purity analytical reagents and biological stan-

Table 1
Nuclear Data of Investigated Elements and the Applied Method

Elements	Radionuclide formed	Half-life	Energy of measured gamma-ray, kEv	Methods
La	^{140}La	40.2 h	1596	INAA
Ce	^{141}Ce	32.5 d	145.5	RNAA
Sm	^{153}Sm	46.8 h	103	INAA
Gd	^{153}Gd	236 d	98	RNAA
Yb	^{169}Yb	32.6 d	177	RNAA
Lu	^{177}Lu	6.74 d	208	RNAA
As	^{76}As	26.3 h	559	INAA
Tb	^{233}Pa	27 d	312	RNAA

dard reference material Chinese Human hair (GBW 09101), were sealed in polyethylene vials and wrapped in high-purity aluminum foil for irradiation. The samples and standards were packed together and irradiated with a neutron flux density of 3.7×10^{13} n/cm^2 s for 9 h in the heavy water reactor at Beijing Atomic Energy Institute, China. After a delay time of 9 d the samples were counted to determine As, La, and Sm by gamma-ray spectrometry using a high-efficiency Ge (Li) detector coupled to a CANBERRA 90 multichannel analyzer. The energy of emitted gamma rays utilized in the present paper for REE and element As and Th were reported in Table 1.

Radiochemical Procedure

The irradiated samples, after the delay time of 2 wk, were transferred to a beaker and known amounts of the elements to be determined were added as carriers. Samples were then dissolved with a mixture of 14*M* nitric acid and hyperchloric acid solutions (v/v 7:3). The solvent was evaporated to dryness. The residue was dissolved in 10 mL 0.1 mol/L HCL and quantitatively transferred to a P-204 extraction chromatography column (100 mesh, 15 cm height, and 0.8 cm I.D. [inside diameter]) for adsorption of REE and Pa. It was washed afterward with 25 mL 0.1 mol/L HCL to remove non-REE including Hg, Cr, Se, Sc, Rb, Fe, Zn, Co, and Sb. The contents of element Ce, Gd, Tb, Lu, and Th were determined directly from the resin phases. The chemical yield of REE and Th measured by radioisotope tracer was 96–100%.

RESULTS AND DISCUSSION

The analytical results presented in this study are given in Table 2. The mean ratio of freeze-dry-to-wet wt for 18 normal brain tissues 0.245 (0.012) is consistent with the literature value 0.211 (0.004) presented by Ehmann *(6)*. More content of water was observed from 16 brain tumor tissues that may be related to the porous sponges structure of tumor tis-

Table 2
Element Contents in Normal Brain and Brain-Tumor Tissues[a]

Elements	A Normal brain tissue (18)[b], ng/g dry wt	B Brain tumor tissue (16)[b], ng/g dry wt	Ratio B/A	*P* value
As	55 (42)[c]	115 (123)	2.09	<0.05
Th	1.32 (0.62)	5.84 (4.11)	4.42	<0.001
La	13 (7)	43 (32)	3.31	<0.01
Ce	20.5 (9.0)	89 (57)	4.34	<0.001
Sm	3.9 (1.9)	4.5 (2.6)	1.15	>0.05
Gd	1.8 (0.71)	5.67 (3.65)	3.15	<0.001
Yb	2.51 (1.71)	4.24 (2.62)	1.69	>0.05
Lu	1.11 (0.51)	1.56 (0.65)	1.40	<0.05

[a]The mean ratios of freeze dry/wet for the brain and brain tumor tissues were 0.245 (0.012) and 0.175 (0.043), respectively.
[b]Number of cases.
[c]Standard deviation.

sues. The radiochemical neutron activation analysis method adopted in the present work for REE, As, and Th assay has shown to be quite capable of yielding reliable results. The reliability of the analytical techniques has been checked using the biological standard material Chinese human hair (GBW 09101). The analytical values coincided with the certified values. The relative standard deviation for the elements determined was less than 5–10%. The biological significance of REE for the brain tumor tissue is not yet well understood. Study on non-REE multielement contents of normal brain and brain tumor tissues determined by RNAA *(12)* and brain-tumor tissues determined by PIXE technique *(13)* and NAA *(9)* have been done. The levels of elements Ca, Mn, Fe, Cu, Se, Zn, Br, Hg, and trivalent element Sc in the brain tumor tissues showed a significant elevation compared with normal brain. The occurrence of element La was sought in the neutrophils and the malignant tissues from breast cancer patients *(14)*. The high levels of Sb, As, Cd, La, and Pb in lung tissue of smelter workers who died from lung cancer were also observed *(15)*. The effects of rare earth compounds, ytterbium and lanthanum trichloride, on malignant tumor cell in mice by cytochemistry and microscopic techniques have shown that the contents of succinic acid dehydrogenase, cytochrome oxidase, alkaphosphatase, and acid ATPase in malignant tumor cells were changed. The mitochondria and endoplasmic reticulum in malignant tumor cells were damaged *(16)*. The results presented in this study showed that the brain-tumor tissues exhibit statistically significant higher REE, As, and Th levels with respect to normal brain tissues. In the cases of La, Ce, Gd, and Th, the levels of brain tumor were observed to have a very significant increase after using *t*-testing ($P < 0.001$). Therefore, it is worthwhile to note the abnormal accumulation of

REE, As, and Th in the brain-tumor tissues, even if the contents of the elements are at ng/g level. A multifactorial genesis for the development of brain tumor was suggested from the results. Further investigations are now in progress to ascertain the clinical significance and their potential protection usefulness.

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