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Magnesium contents of leukemic lymphocytes

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In this study, magnesium concentrations were measured in lymphocytes from patients with acute myeloblastic leukemia (AML), chronic megalositer leukemia (KML) and acute lymphoblastic leukemia (ALL) before and after chemotherapy management, and results were compared with those of control subjects. Magnesium concentrations were higher in the patient groups compared with control values. However, no meaningful differences were found among magnesium concentrations of the patient groups themselves. Similarly, no statistically meaningful differences were found between lymphocyte magnesium concentrations before and after chemotherapy management in the patient groups. In the inter-correlation analysis, we observed no correlations between pre- and post-magnesium concentrations in patients' lymphocytes. It has been suggested that magnesium concentrations of leukemic lymphocytes might increase due to the high ATP requirement of the leukemic cells since magnesium is known to play an important part as a cofactor in most of the energy-producing reactions.

Keywords: leukemia, magnesium

Introduction

There are several publications on the elemental content of leukemic lymphocytes (Andronikashvili & Mosulishvili 1980, Carpentieri *et al.* 1984, 1988). In these publications it is suggested that some metals, such as iron, copper, zinc etc., play an important part in lymphocyte maturation and regulation of immune function (Chandra *et al.* 1977, Allen *et al.* 1981). Several studies have been performed. In one study, it was found that the zinc concentration was lower and the copper and iron concentrations were higher in lymphocytes from children with acute lymphocytic leukemia (ALL) compared with controls (Carpentieri *et al.* 1984). Similar results were obtained by several research groups, but the only information available was limited to the low zinc concentrations of the lymphocytes from leukemic patients (Carpentieri *et al.* 1984).

Magnesium is one of the physiologically important elements, functioning in various cellular processes. Several researchers measured magnesium concentrations in serum, cancerous tissues and cells. In cancerous breast, stomach and colon tissues, magnesium concentrations were found to be lower than those in normal tissues (Ranade & Pandey 1985); however, in oral, bone marrow and esophageal cancers, no differences were found between normal and cancerous tissue magnesium concentrations (Ranade & Pandey 1985). Magnesium concentrations in lymph nodes were found to be increased in non-Hodgkin's lymphoma but unchanged in Hodgkin's lymphoma (Krishna & Ranade 1985). Although it is suggested that the effects of iron, copper and zinc on human lymphocytes are related and interdependent, and require the presence of adequate levels of magnesium, no documented information exists regarding the magnesium concentrations of lymphocytes from patients with AML, KML and ALL.

The purpose of this study is to determine magnesium concentrations of leukemic lymphocytes and to compare the results with those of control subjects, and to investigate possible effects of chemotherapy management on the magnesium concentrations of leukemic lymphocytes.

Materials and Methods

Peripheral blood human lymphocytes were obtained from 10 AML, five KML and five ALL patients, and from 11 healthy subjects. Healthy subjects ranged in ages from 23 to 36 years (mean ± SD 28.2 ± 7.9), AML patients from 18 to 48 years (33.5 ± 13.9), KML patients from 32 to 59 years (49.2 ± 11.9) and ALL patients from 6 to 31 years (20.3 ± 8.8). In the control group, six subjects were male and five subjects were female. In the patient groups, five

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subjects were male and five subjects were female in the AML group, two subjects were male and three subjects were female in the KML group and, three subjects were male and two subjects were female in the ALL group. All patients were chosen from among individuals who were first diagnosed as leukemic and no previous leukemia history was present for the patients studied.

Patients were under combined chemotherapy management (given below). In the AML group, cytosine arabinoside, daunorubicin; in the ALL group, vincristine, daunorubicin, prednizon, L-asparaginase, cytosine arabinoside, 6-mercaptopurin, methotrexate; and in the KML group, hydroxyurea and busulfan were used.

Second samples were obtained after the first remission period of chemotherapy management (range: from 15 days to 2 months, mean ± SD 32.5 ± 7.2 days). Leukemia diagnosis and remission criteria were made by clinical examination, microscopic investigation of peripheral blood cells and bone marrow material, and histochemical staining techniques and routine hematologic analyses in the Hematology Department.

After fasting, blood samples were obtained at the morning, lymphocytes were isolated on Ficol-Hypaque, and red blood cells and platelets purified (Carpentieri *et al.* 1984). After contamination was tested by morphology, the amount of protein was determined by the method of Lowry (1951). Then, samples were digested in a nitric acid:perchloric acid (5:1, v/v) mixture by heating mildly. After digestion, samples were dissolved in a definitive amount of water. Magnesium analysis was performed on the last clear solution by using an atomic absorption spectrophotometer (Varian Techtron Model 1200 AAS). In the analysis, a standard addition technique was used (Kirkbright 1980).

Results

Magnesium concentrations of lymphocytes from controls and leukemic patients are given in Table 1, and Student's *t*-test analysis results are given in Table 2. Magnesium concentrations were expressed as $\mu g\ Mg\ g^{-1}$ protein due to the suggestion that the $mg\ g^{-1}$ protein estimate is more reliable and reproducible than the $\mu g\ Mg\ cell^{-1}$ estimate.

As seen from the results, the magnesium concentrations of leukemic lymphocytes were meaningfully higher than control values. However, there were no differences between the magnesium concentrations of leukemic lymphocytes themselves. Similarly, there were no significant differences between pre- and post-magnesium concentrations of patient lymphocytes.

Table 1. magnesium concentrations of lymphocytes from the patients and controls

Group	Magnesium concentrations before chemotherapy management ($\mu g\ g^{-1}$protein)	Magnesium concentrations after chemotherapy management ($\mu g\ g^{-1}$protein)
Control (n = 11)	1169.2 ± 201.9	—
AML (n = 10)	2427.3 ± 649.6	3035.5 ± 852.4
KML (n = 5)	3222.0 ± 457.3	2835.2 ± 585.4
ALL (n = 5)	3029.3 ± 630.0	2826.5 ± 536.1

Table 2. Student's *t*-test results

Groups	P
Control–AML[a]	< 0.05
Control–AML[b]	< 0.05
Control–KML[a]	< 0.05
Control–KML[b]	< 0.05
Control–ALL[a]	< 0.05
Control–ALL[b]	< 0.05
AML[a]–AML[b]	NS
KML[a]–KML[b]	NS
ALL[a]–ALL[b]	NS

NS, non-significant ($P < 0.05$).
[a]Magnesium concentrations before chemotherapy management.
[b]Magnesium concentrations after chemotherapy management.

Discussion

There are several studies on the elemental contents of lymphocytes from patients with various diseases. In particular, zinc, copper and iron concentrations were measured in leukemic lymphocytes but no coincidal results were generally found (Andronikashvili & Mosulishvili 1980, Allen *et al.* 1981, Carpentieri *et al.* 1984, 1988) for some elements except zinc, which was usually found to be decreased in leukemic lymphocytes (Carpentieri *et al.* 1984, 1988). Several researchers measured magnesium concentrations of human mononuclear blood cells and lymphocytes (Girardin & Paunier 1985, Hnang *et al.* 1988, Urdal & Landmark 1989, Yang *et al.* 1989, Geven *et al.* 1990). However, no data were available for the magnesium contents of various types of leukemic lymphocytes.

When our lymphocyte magnesium levels were compared with those of several studies, a general agreement was seen for the control group (Girardin & Paunier 1985, Urdal & Landmark 1989, Geven *et al.* 1990); however, we were not able to compare magnesium concentrations of leukemic lymphocytes with those of literature because no data were available in this respect.

We thought that establishment of magnesium concentrations of leukemic lymphocytes might be helpful in the elucidation of the elemental status of the leukemic lymphocytes because magnesium was mainly an intracellular cation and its concentration in the cells might give an important insight into the element metabolism of the leukemic cells. In fact, in a study carried out by Carpentieri *et al.* (1988) it was pointed out that the effects of iron, copper and zinc on human lymphocytes were related and interdependent, and required the presence of adequate levels of magnesium. Looking at our results in view of the information given above, we realized that magnesium concentrations increased in lymphocytes from patients with AML, KML and ALL, and that no differences existed between the values determined before and after

chemotherapy. In some publications, however, the magnesium contents of some cancerous tissues and cells were found to be unchanged or decreased compared with control counterparts (Carpentieri *et al.* 1984, Rande & Panday 1985). There were also some publications in which increased magnesium concentrations were reported in malignant lymphomas (e.g. Krishna & Ranade 1985).

Although various hypotheses might be put forward to explain the results of the study presented here, we think that increases observed in magnesium concentrations in leukemic lymphocytes arise from increased energy (ATP) requirements of the leukemic cells since magnesium is an important cofactor for the reactions by which ATP is synthesized (Murray *et al.* 1991). The increase in magnesium concentrations might affect the levels of other elements, including iron, copper, zinc, calcium, etc., which thus cause changes in the normal element metabolism of the cell. All of these changes may provide a selective advantage to cancer cells.

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