

CELL MEDIATED IMMUNITY IN PATIENTS WITH OSTEOSARCOMA

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Because of the difficulty of obtaining suitable material, earlier studies on cell mediated immunity in the radium patients failed to include positive controls. Recently we were fortunate in obtaining samples of lymphocytes from two suitable patients who had had amputations for spontaneous osteosarcoma six months previously. Lymphocytes from both of these patients showed cytotoxicity to cultured cells derived from a human osteogenic sarcoma but not to normal fibroblasts. These results help to validate our test for early detection of osteosarcoma in the radium patients using measurements of cytotoxicity.

Introduction

Earlier reports have dealt with tests for measuring cell mediated immunity in the radium patients. ^(1,2) Of the 20 patients studied with body burdens greater than $0.5 \mu\text{Ci } ^{226}\text{Ra}$, only one patient had lymphocytes which were shown to be cytotoxic to osteosarcoma cells in culture. This patient was suffering from an inflammation of the hip joint due to a foreign body reaction at the time of the test. Subsequent measurements on the same patient 6 months and 19 months after the inflammation had been treated showed no cytotoxicity.

Because osteosarcoma is a rare disease, and only samples of blood from patients who had not previously been treated with radiotherapy or chemotherapy were suitable as positive control samples for the test, no positive controls were included in the earlier studies. Recently we obtained lymphocytes from two patients, D and G, who had undergone amputations for spontaneous osteosarcomas six months previously.* The results of tests for cell mediated immunity in these patients are described in the present report.

Materials and Methods

The method for the separation of the lymphocytes using a Ficoll-Hypaque gradient has already been described. ⁽¹⁾ The lymphocyte to target cell ratio used throughout was 1000:1. In each test, media controls, as well

* Dr. Gerald Miller of the Mayo Clinic kindly provided us with lymphocyte samples from the two osteosarcoma patients.

as lymphocytes from a normal subject, were tested simultaneously with the patient's lymphocytes. Lymphocytes from the two patients, D and G, were interacted with target cells from two cultures. One of the cultures, LOS-28, was derived from an osteosarcoma, while the other, DF-550, was a normal fibroblast culture.

Results

As seen in Fig. 1, lymphocytes from both patients D and G were cytotoxic to the osteosarcoma cells of LOS-28 but not to the normal fibroblasts.

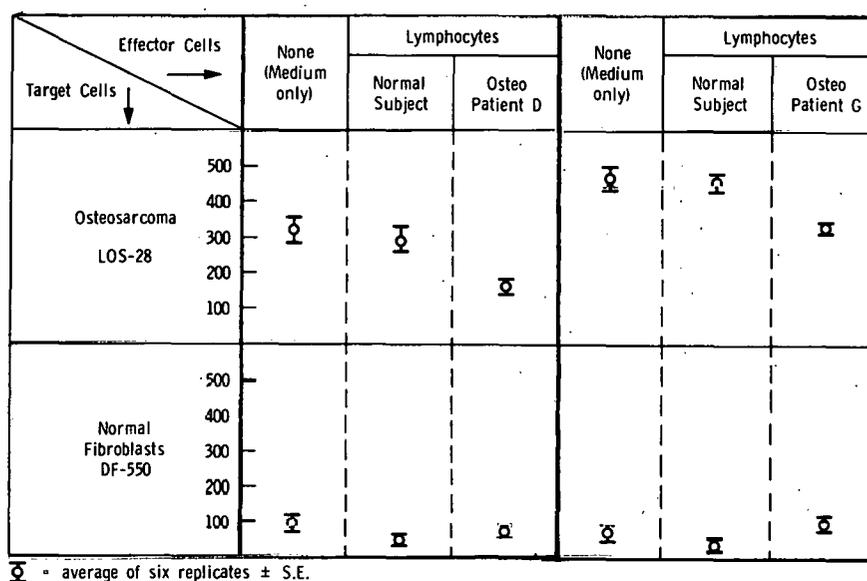


FIG. 1. --Number of target cells remaining after incubation with lymphocytes from two patients, D and G, who had amputations for osteosarcoma 6 months previously.

The points plotted show the number of target cells remaining after a 48-hr incubation period in each case. Each point on the figure shows the mean value and the standard error for 6 replicate samples. When normal control lymphocytes were used, the number of target cells of LOS-28 was the same within the standard error as when media alone was used. By contrast, the number of tumor cells remaining was reduced by 45% for patient D and by 27% for patient G, relative to the values obtained for the normal control subjects. With the normal fibroblasts, however, no difference was observed between the

effects of lymphocytes from the patients or the controls. These results go a long way towards validating our test for early detection of osteosarcoma in the high level radium patients. However, it is still necessary to test patients at various stages of their disease using a variety of osteosarcoma cell cultures at different passage levels for a more complete study.

References

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2. Menon, M., E. L. Lloyd, and J. L. Mitchen. Interrelationships of radiation, viruses, and the immune response in radium-induced tumors. Part III. Lymphocyte cytotoxicity to osteosarcoma cells in vitro. Radiological and Environmental Research Division Annual Report, Part II, July 1973-June 1974. ANL-75-3, Part II, pp. 21-25.