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EVALUATION OF SURFACE MARKERS IN CHILDHOOD ACUTE LYMPHOCYTIC LEUKEMIA BEFORE AND AFTER THERAPY.

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INTRODUCTION

According to the investigations which have been done, childhood acute lymphocytic leukemia (ALL) is a heterogenous disease. (1,2,3,4,7). A substantially better prognosis in non-T, non-B or null cell origin ALL has been reported by some groups 3,8. The present study was undertaken to clarify the origin of the cells involved in ALL in Iranian children who were affected by the disease. This study also re-evaluates the surface marker while patients were on complete remission to discover any changes which might happen to surface markers with therapy.

MATERIALS AND METHODS

Patients: 16 children who hospitalized in the hematology department of Sharazad Children's Hospital with

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the diagnosis of ALL were selected for our study. The diagnosis of ALL in them was based on clinical pictures, CBC and bone marrow examination by the hematologist in charge of the department. The patients aged from 15 months to 14 years with a mean of 5.4 years. From each patient 7 ml of peripheral blood was sent to the Immunology department of Tehran University for surface markers, study 1) prior to any therapy and 2), 2-4 months after therapy when they were considered on complete remission.

TESTS

Peripheral lymphocytes were separated according to the method of Thorby and Bratile, 1970.(9)T-lymphocytes were assayed for their spontaneous rosette formation with sheep erythrocytes (E) and B-lymphocytes for their complement receptors by antigen-antibody complement rosette formation. Tests for E(T-lymphocytes) and EAC(B-lymphocytes) rosettes were performed as previously described(6). For E rosettes, the lymphocytes were mixed with an equal volume of 0.5% sheep erythrocytes(E).The mixed cell suspensions after centrifugation were incubated for one hour at 4^{o}C. Pellets were resuspended by gentle shaking and rosettes counted using a hemocytometer. Only lymphocytes with three or more E were considered rosettes. For EAC rosettes, complement receptors were visualized by using sheep erythrocytes(SRBC)presensitized with rabbit anti-SRBC and mouse complement. Null cells were cells which did not have either T or B cell markers.

Total and differential white blood cell counts, as well as total lymphocytes/cmm of blood, were done on all blood samples.

RESULTS

Prior to therapy WBC(total white blood cells/cmm of blood)was from 5500 to 444/800 with a mean of 79, 792;the percentage of lymphocytes in the peripheral blood was from 35-100 with a mean of 88.6%(Table 1).In the study of surface markers on the peripheral leukemic cells, prior to any therapy T-lymphocytes were from 4 to 76% with a mean of 23.6%; B-lymphocytes from 1 to 30% with a mean of 11.42%; null cells were from 1 to 84% with a mean of 63.5%(Table 2). In the initial study 13 of our study patients (or 80% of them)had predominantly non T non-B or null cell leukemia. Three of them had less than 27% null cells in the initial study. In these three patients the percentages of T-lymphocytes were 76,46 and 67 and the percentages of B- lymphocytes were 24,36 and 7, respectively. After therapy while on complete remission, there was tremendous decrease in the percentage of null cells (in null cell leukemia) and increase in the percentage of T and B-lymphocytes(Table 4), which approached data of age and sex matched normal control subjects(Table 5).

On remission there have been tremendous decrease in WBC for most of the patients. Counts of WBC on remission were from, 14/300-3/400cmm with a mean of 7,490 (Table 3).

Three of the patients who had less than 27% null cells in the initial study died in a period of less than one year after diagnosis. One of the null cell leukemia patients with WBC 28,500 in the initial study while being on bone marrow remission in re-evaluation of surface markers still showed null cells predominant

Table I

PRIOR TO THERAPY

	WBC	PERCENTAGE OF LYMPHOCYTES	ABSOLUTE NO OF LYMPH/CMM OF BLOOD
RANGE	5500 - 444800	35-100	3500-400320
MEAN	79792	88.6	33173.9

Total white blood cell counts/cmm (WBC) Percentage of lymphocytes and absolute no of lymphocytes / cmm of blood prior to therapy

Table 2 — PRIOR TO THERAPY

	ABSOLUTE NO OF T LYMPHOCYTES/CMM OF BLOOD	ABSOLUTE NO OF B LYMPHOCYTES/CMM OF BLOOD	ABSOLUTE NO OF NULL CELLS/ CMM OF BLOOD
RANGE	385-30243	595-92073	2520-254870
MEAN	34395.2	10047.7	30173.9

Absolute No of T, B Lymphocytes and Null cells /cmm of blood prior to therapy

Table 3

AFTER THERAPY IN REMISSION

WBC	PERCENTAGE OF LYMPHOCYTES	ABSOLUTE NO OF LYMPHOCYTES/ /CMM OF BLOOD
RANGE 3400-14300	29-66	1520-6578
MEAN 7690	40.2	3700

Total white blood cell counts/cmm (WBC), percentage of lymphocytes and absolute no of lymphocytes/cmm of blood after therapy in remissin

Table 4 AFTER THERAPY IN REMISSION

	PERCENTAGE OF T LYMPHOCYTES	PERCENTAGE OF B LYMPHOCYTES	PERCENTAGE OF NULL CELLS
RANGE	32 - 75	11 - 32	9 - 23
MEAN	61. 3	20 . 75	18

Percentage of T,B lymphocytes and null cells after therapy in remission

Table 5

NORMAL CONTROLS

WBC	PERCENTAGE OF LYMPHOCYTES	ABSOLUTE NO OF LYMPHOCYTES/CMM OF BLOOD
RANGE 5700-13200	23 - 62	1380 - 7524
MEAN 8827	44 . 7	4000

Total white blood cell counts/cmm(WBC), Percentage of lymphocytes and absolute no of lymphocytes/cmm of blood in normal controls.

Table 6

NORMAL CONTROLS

	PERCENTAGE OF T LYMPHOCYTES	PERCENTAGE OF B LYMPHOCYTES	PERCENTAGE OF NULL CELLS
RANGE	50 - 76	18 - 33	0 - 29
MEAN	61 . 1	24 . 2	14 . 7

Percentage of T, B lymphocytes and Null cells in normal controls

similar to his initial study. Shortly after this reevaluation he developed CNS relapse followed by bone marrow relapse and death.

Twelve of our patients who had null cell leukemia are alive and well.18 months to twenty months after initiation of therapy.

DISCUSSION

The majority of the reports from surface markers study, in the childhood acute lymphocytic leukemia(ALL) indicates that All is a heterogenous disease. A better prognosis in non T and non B or null cell origin All has been reported by some groups.

In our study of surface markers of ALL in children, the majority (80%)had null cells In their peripheral blood in the initial study. Our study confirmed the findings of others that non-T, non-B, or null cell leukemias have better prognosis. From this study one could assume that re-evaluation of lymphocytes surface markers during the course of ALL might be helpful in predicting the effect of therapy, impending relaps and prognosis. As indicated by 1) the number of T and B lymphocytes in peripheral blood coming close to normal controls for null cell leukemia patients during remission,and 2)shortly after a reevaluation study, the development of bone marrow and CNS relapse resulting in death for one patient whose null cells remained high while on remission.

SUMMARY

In our study of surface markers of Iranian children affected by acute lymphocytic leukemia, the majority(80 %)had non-T, non-B, or null cell leukemia. The null cell

leukemia had a better prognosis as was confirmed by others. Twelve of our patients who had null cell leukemia are alive and well 18 months to 20 months after initiation of therapy. Three of the patients who had less than 27% null cells in the initial study died in a period of less than one year after diagnosis.

After therapy while on remission, there was a decrease in the percentage of null cells(in null cell leukemia) and an increase in the percentage of T and B lymphocytes in the peripheral blood which approached the data on the age and sex-matched normal control subjects.

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