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UICC

FIGHT CANCER REGARDLESS

PROCEEDINGS OF THE

XVII INTERNATIONAL CANCER CONGRESS

New Delhi (India), October 30 - November 5, 1994

Editors
R.S. RAO, M.G. DEO and L.D. SANGHVI
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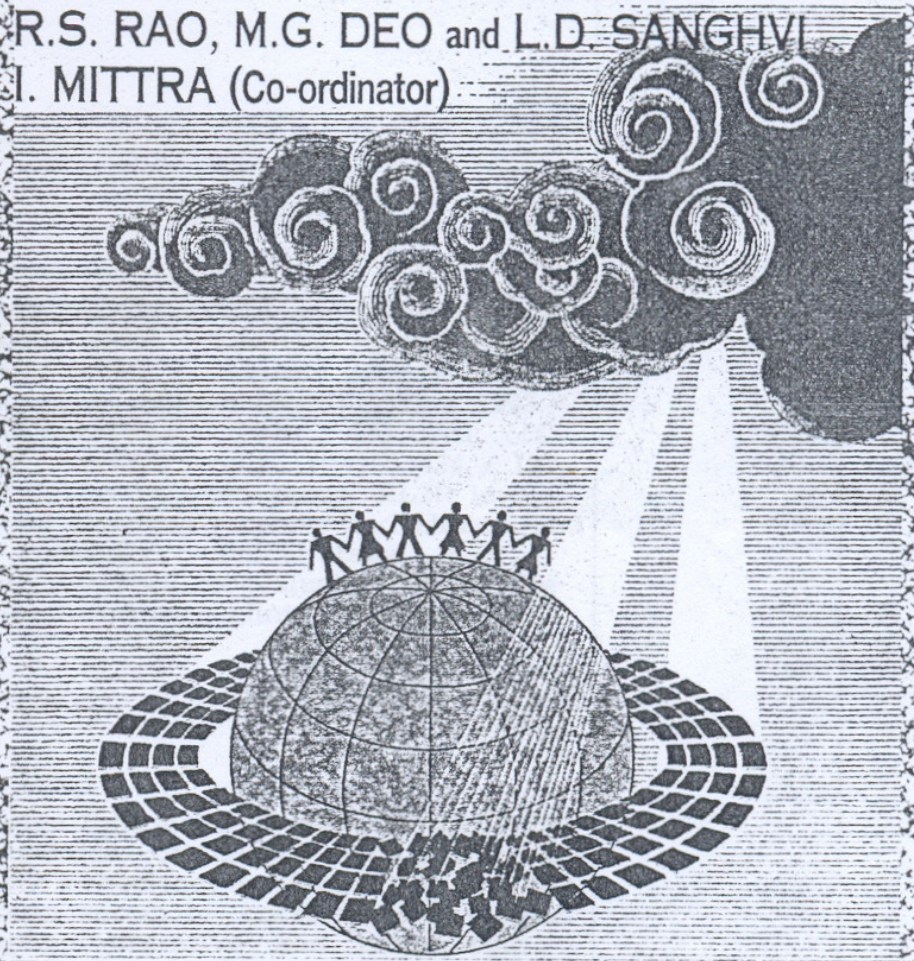

MONDUZZI EDITORE

INTERNATIONAL PROCEEDINGS DIVISION

XVI International
Cancer Congress
1994

New Delhi, India
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Serum soluble interleukin 2 receptor levels in children with Hodgkin's disease

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SUMMARY

Studies of peripheral blood lymphocytes (PBL) and serum from 35 children with Hodgkin's disease (HD) show that serum soluble interleukin-2 receptor (sIL2R) levels are closely related with disease status { patients with active disease (n=15) 3572 ± 1253 u/ml, on therapy, early remission (n=16) 1578 ± 480 u/ml, off therapy (n=4) 447 ± 163 u/ml, control group (n=15) 424 ± 137 u/ml ($p<0.001$)}, but no correlation between serum sIL2R levels and PBL CD25 expression. Some previously recognized clinical and laboratory variables such as histology, clinical stage, ESR, blood and hair zinc levels were analysed to evaluate whether the level of serum sIL2R was associated with clinical and pathologic parameters of HD.

INTRODUCTION

Increased serum levels of soluble interleukin 2 receptor(sIL2R), the truncated extramembrane portion of the p55 alpha subunit of the IL2R complex, detected by monoclonal antibodies to the Tac glycoprotein (CD25), have been shown to correlate with disease activity in a variety of lymphoproliferative and hematologic malignancies including Hodgkin's disease (HD) (1-6). Although the increases in serum levels may result from release by activated normal lymphocytes, they are more likely to derive from

neoplastic cells. Pizzolo et al. demonstrated the majority of Hodgkin and Reed - Sternberg cells expressed IL2R as evidenced by a strong cytoplasmic reaction with an anti-Tac antibody (7). Moreover, higher proportions of activated lymphocytes with surface IL2R were found in tissues involved by HD than in normal or reactive lymph nodes. Impaired cell - mediated immunity in untreated HD has long been recognised (8). The nature of the defect in HD remains enigmatic, although evidence points to immunosuppressive factors in serum or plasma of those patients. It is possible that the sIL2R itself is immunosuppressive. Since it is capable of binding IL2, it might compete with its counterpart on the surface of normal lymphocytes and may have a role in the immunosuppression characteristic of patients with advanced HD. On the other hand Zinc (Zn), which plays a crucial role in the development of normal cellular immunity, has been shown to be deficient in different parts of Turkey. Our previous studies have demonstrated a high frequency of HD and chronic Zn deficiency associated with this type of malignant lymphoma in Turkey (9,10). In the present study we aimed to determine sIL2R levels in the sera of patients with HD and to establish the possible correlation with disease features and Zinc (Zn) status of patients at presentation.

MATERIALS AND METHODS

Thirtyfive patients with biopsy-proven HD were included in the study. There were 27 boys and 8 girls with an age range from 3.5 to 17 years (median 8.5 years). In all patients the diagnosis was based on histological findings, the nodular sclerosis subtype found in 4 patients, mixed cellularity in 29, lymphocyte predominance in 2 patients. Of 35 patients 6 were in stages I and II, 8 were stages III and 21 were stage IV according to Rye classification.

ASSAY FOR CYTOKINE

Soluble IL2R was measured using stored frozen serum from patients with HD { active disease (n=15), on therapy, early remission (n=16) and off therapy (n=4) and control group (n=15)} in a sandwich ELISA technique (T Cell Sciences, Cambridge, MA, USA).

IMMUNOPHENOTYPING

PBL, were immunophenotyped using indirect immunofluorescence with anti (α) CD25 (IgG1, Cymbus) monoclonal antibody, with goat α mouse IgG1 FITC (Cymbus Bioscience Ltd, Southampton) as the first and second layers, respectively. Irrelevant isotype matched mouse IgG1 antibody (Serotec, Kidlington, Oxford, UK) was used as negative control.

Zn levels were measured in blood (plasma and erythrocyte) and hair samples of the patients by flame atomic absorption spectrophotometer (Perkin-Elmer Model) as described previously (8,9).

STATISTICAL ANALYSIS

Paired or unpairedm t testsm werem used to compare the data for significant differences ($p<0.05$). Analysis of correlations between ESR, blood and hair Zn levels and serum sIL2R were performed using Spearman rank's correlation test.

RESULTS AND CONCLUSIONS

Results of the plasma sIL2R levels of patients with HD in remission or active disease and control group are summarised in table 1. Active HD was associated with a higher serum sIL2R levels, when compared with normal volunteers or patients in remission, off therapy ($p<0.001$).

The results of Zn analysis of blood and hair are shown in table 2. The mean Zn levels in blood (plasma, erythrocyte) and hair were found to be significantly decreased in patients with active HD when compared to controls ($p<0.001$, $p<0.001$ and $p<0.001$, respectively). Using Spearman rank test, we observed no correlations between serum sIL2R levels and PBL CD25 expression, ESR or blood and hair Zn levels (coefficients between -0.322 and 0.699; p values over 0.50).

Table 1-ELISA serum sIL2R levels in patients with HD vs controls

Source of serum	number	sIL2R U/ml (mean±SD)	t test
Normal volunteers	15	424 ± 137	
Hodgkin's disease Active	15	3572 ± 1253	p<0.001
Remission (off therapy)	4	447 ± 163	unpaired

Table 2-Zinc Status in Pediatric Hodgkin's Disease

	Mean ± Sx		
Group	Plasma Zn (µg/dl)	RBC Zn (µg/ml)	Hair Zn (µg/g)
Patients	72.9 ± 6.06 (10)*	11.17 ± 0.59 (10)	109.57 ± 9.69 (9)
Controls	109.8 ± 12.3 (83)	15.8 ± 2.8 (34)	184 ± 19.36 (97)
p	<0.001	<0.001	<0.001

* No. of studied subjects is shown in parenthesis

Serum levels of soluble IL2R are easily quantitated by an ELISA. Soluble IL2R levels have been found to be higher in HD patients with active disease than in serum from remission group and normal children. The increased serum soluble IL2R levels in our patients with active disease could reflect greater release of the receptor from either malignant cells or activated normal lymphocytes. We favor the first explanation, because soluble IL2R levels are not correlated with PBL cell surface IL2R (CD25) expression. Furthermore with in vitro assays, a lack of functional IL2R expression on PBL from patients with cancer have been demonstrated (6,11). The functional significance of sIL2R is unknown. Since soluble IL2R is capable of binding interleukin 2, it may have an immunoregulatory role by competing with cellular IL2R for the ligand and thus down-regulating the immune response (12). In this regard, the soluble IL2R has been suggested to be blocking factor produced by the malignant cells which, by binding to the host's growth factors, may inhibit the normal immune response attempting to eliminate those tumor cells (3,4,6,12). We conclude that sIL2R levels in children with HD appear to be a rapid, reliable, and noninvasive measure of disease activity. Understanding the biological significance of sIL2R will render to clinical applications of IL2R directed therapy representing a new perspective for the treatment of certain neoplastic diseases including pediatric lymphoid tumors.

Supported in part by Turkish Scientific Research Council(TÜBITAK

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