

Detection of Interleukin-2 Receptor (IL-2r)  
by Indirect Immunofluorescence with Anti-Tac  
Monoclonal Antibody on the Surface of T Lymphocytes  
from Patients with Lepromatous Leprosy

TO THE EDITOR:

We have recently published that lymphocytes from patients with lepromatous leprosy (LL) stimulated with concanavalin A (ConA) are deficient in the synthesis of interleukin-2 (IL-2); however, the cells possess receptors for IL-2 (IL-2r) in such a manner that when activated lymphocytes are cultured *in vitro* and exogenous IL-2 is added, they recover their capacity to proliferate as demonstrated by the incorpora-

tion of tritiated thymidine (<sup>3</sup>). A demonstration of the physical presence of IL-2r on the surface of T lymphocytes activated with phytohemagglutinin (PHA) by means of indirect immunofluorescence (IIF) using an anti-Tac monoclonal antibody (<sup>4, 8, 9, 11</sup>) is reported here. (The anti-Tac monoclonal antibody was kindly provided by Dr. T. A. Waldmann, Metabolism Branch, National Cancer Institute, National Institutes of Health, Bethesda, Maryland, U.S.A., and we thank him for his courtesy.)

Five women and eight men between the ages of 20 and 60 years old were studied at the Instituto Dermatológico de Guadalajara, Jalisco, Mexico. All of them had nodular or diffuse LL, a positive bacilloscopy, and had been receiving irregular treatment with 100 mg of dapsone daily for from 2 to 15 years. The control group consisted of eight healthy persons, of similar sex and age, not related to the patients.

The mononuclear cells from heparinized venous blood were obtained by means of Ficoll-Hypaque and resuspended in RPMI 1640 medium supplemented with 10% fetal calf serum (3). The IL-2r was studied and detected by IIF (1) on the membrane of the lymphocytes using anti-Tac monoclonal antibody; for this purpose two groups of cells were studied. The first group (To) corresponds to lymphocytes without any stimulation; the second group (T<sub>72</sub>) consisted of lymphocytes previously activated with PHA for 72 hr. Cells ( $1 \times 10^6$ ) obtained from experiments To and T<sub>72</sub> were incubated for 30 min at 4°C with anti-Tac monoclonal antibody diluted 1:500. The cells were then washed and again incubated for 30 min at 4°C with anti-mouse IgG antibody labeled with fluorescein isothiocyanate (FITC) (Sigma Chemical Co., St. Louis, Missouri, U.S.A.), and were analyzed by fluorescence microscopy. Data were obtained by count-



THE FIGURE. Tac+ cells revealed by indirect immunofluorescence. Anti-Tac monoclonal antibodies are bound to IL-2r and demonstrated by fluorescent anti-mouse IgG antibodies.

ing 300 cells and the percent of Tac+ cells was calculated for each of the studied groups.

The data obtained by IIF show that both the cells from the group of LL patients and those of the control group not activated with PHA (To) had practically no Tac+ cells (The

THE TABLE. Presence of IL-2r on the surface of T lymphocytes by IIF with anti-Tac antibody.

Case	LL cases (N = 13)		Control group (N = 8)	
	To <sup>a</sup>	T <sub>72</sub> <sup>b</sup>	To <sup>a</sup>	T <sub>72</sub> <sup>b</sup>
1	1.0	72.0	0.0	70.3
2	1.0	63.1	0.0	60.0
3	1.5	51.1	3.0	80.0
4	4.7	57.9	0.0	64.4
5	6.4	22.2	2.0	54.7
6	0.6	69.3	1.0	51.4
7	0.0	70.2	0.3	76.8
8	1.0	64.8	2.0	65.0
9	0.9	80.1		
10	0.0	63.0		
11	1.0	62.0		
12	0.0	69.3		
13	0.0	53.0		
Mean ± S.D.	1.3 ± 1.8	60.0 ± 13.4 <sup>c</sup>	1.04 ± 1.08	65.3 ± 9.4 <sup>c</sup>

<sup>a</sup> Mononuclear cells from peripheral blood taken from LL patients and controls without stimuli.

<sup>b</sup> Mononuclear cells cultured 72 hr with PHA (10 µg/ml).

<sup>c</sup> Difference not statistically significant; Student's *t* test.

Table). However, in both groups after activation with PHA for 72 hr ( $T_{72}$ ) the percentages were  $60.8 \pm 13.4$  for cells from LL subjects and  $65.3 \pm 9.4$  for the control group. These results were not different statistically when an independent Student's *t* test was performed.

This study demonstrates the physical presence of the IL-2r by the specific union of anti-Tac monoclonal antibodies to the 55-kDa chain of the high-affinity IL-2r<sup>(6-9)</sup>, and confirms that T cells of LL patients truly possess IL-2r but are incapable of proliferating adequately due to the deficiency of IL-2 biosynthesis<sup>(2-4)</sup>. It should be mentioned that Mohaghehpour, *et al.* found similar results using ConA<sup>(5)</sup>, but the percentages of T cells expressing IL-2r (Tac+) were lower than those found by us; this difference is probably due to the fact that PHA is a better inducer of IL-2r<sup>(11)</sup>.

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