Jpn. J. Cancer Res. 81, 902–908, September 1990

Interleukin-2 Receptor β-Chain (p70–75) Expressed on Leukemic Cells from Adult T Cell Leukemia Patients

Taiichi Kodaka,[1] Takashi Uchiyama,[1] Takayuki Ishikawa,[1] Masanori Kamio,[1] Rie Onishi,[1] Katsuhiko Itoh,[1] Toshiyuki Hori,[1] Haruto Uchino,[1] Mitsuru Tsudo[2] and Koichi Araki[3]

[1]*The First Division of Internal Medicine, Faculty of Medicine, Kyoto University, 54 Shogoin-Kawaramachi, Sakyo-ku, Kyoto 606,* [2]*Department of Immunology, The Tokyo Metropolitan Institute of Medical Science, Honkomagome, Bunkyo-ku, Tokyo 113 and* [3]*The Second Department of Internal Medicine, School of Medicine, University of the Ryukyus, Nishihara-cho, Okinawa 903-01*

To determine whether the interleukin-2 receptor (IL-2R) β-chain (p70–75) is expressed on leukemic cells from patients with adult T cell leukemia (ATL) as well as α-chain (p55, Tac), we performed radiolabeled interleukin-2 (IL-2) binding assay, chemical crosslinking of radiolabeled IL-2 and flow cytometric analysis using a newly-developed anti-IL-2R β-chain antibody. The results showed that leukemic cells from all the 12 ATL patients we examined expressed the IL-2R β-chain together with the α-chain, whereas there was no detectable β-chain expression on unstimulated peripheral blood CD4(+) T cells from healthy volunteers. Southern blot analysis revealed that this abnormal expression was not caused by the structural change of IL-2R β-chain gene. Though leukemic cells from all ATL patients examined expressed high-affinity IL-2Rs, leukemic cells from only 25% of all ATL patients proliferated in response to IL-2. These results showing abnormal expression of IL-2R β-chain on leukemic cells from ATL patients (ATL cells) suggest a close association between HTLV-I infection and abnormal β-chain expression as well as α-chain expression.

Key words: Adult T cell leukemia — Interleukin-2 receptor

Adult T cell leukemia (ATL)[1] is a leukemia of human T cell leukemia virus type I (HTLV-I)-infected peripheral mature T cells.[2] It has been demonstrated that leukemic cells from ATL patients constitutively express the interleukin-2 receptor (IL-2R) α-chain which is considered to play an important role in the leukemogenesis and the growth of leukemic cells.[3] The role of $p40^{tax}$, one of the HTLV-I viral products, in the activation of cellular IL-2 and IL-2R α-chain gene has been studied.[4] Recent studies from several laboratories revealed another IL-2 binding molecule, IL-2R β-chain (p70–75),[5-8] which may play a more important role in signal transduction. It remained unclear whether IL-2R β-chain is also abnormally expressed on leukemic cells from ATL patients. In the present study, we examined the expression of the IL-2R β-chain on ATL cells by radiolabeled IL-2 binding assay, chemical crosslinking of radiolabeled IL-2 and flow cytometric analysis using a newly-developed anti-IL-2R β-chain antibody. Furthermore, we discuss the relation between the IL-2R α-chain and β-chain expressions on ATL cells and the proliferative response to exogenous IL-2, and the possibility that an IL-2 autocrine mechanism is involved in the growth of ATL cells.

MATERIALS AND METHODS

Patients and diagnosis Twelve patients with ATL were studied (Table I). The diagnosis of ATL was made on the basis of clinical features, morphological characteristics, cell surface phenotypes of leukemic cells,[9] serum antibodies to HTLV-I-associated antigens[2] and HTLV-I proviral DNA integration in leukemic cells.[10]

Cell separation and culture Mononuclear cells were separated from heparinized peripheral blood of ATL patients and healthy volunteers by Ficoll-Conray density gradient centrifugation. The proportion of leukemic cells was more than 85% as determined by cell surface marker analysis. The non-B non-adherent cells were separated from normal peripheral blood mononuclear cells (PBMC) by passage through nylon wool columns at 37°C. Normal PBMC were cultured in RPMI 1640 medium containing 10% fetal calf serum (FCS) (Flow Laboratories, North Ryde, Australia) and 0.1% purified phytohemagglutinin-P (PHA-P) (Difco Laboratories, MI) at 37°C in 5% CO_2 for 7 days. These cells were then harvested and used as normal activated T lymphocytes (PHA blasts).

Flow cytometric analysis Cell surface antigens of leukemic cells and normal PBMC were detected using a FACScan (Becton Dickinson Immunocytometry Systems, CA) after direct immunofluorescence staining with fluorescein isothiocyanate (FITC)- or phycoerythrin (PE)-conjugated Leu3a (CD4), Leu2a (CD8), Leu19 (CD56) (Becton Dickinson Immunocytometry Systems) and FITC-conjugated anti-Tac (CD25)[11,12] monoclonal antibody (mAb) and/or indirect immunofluorescence

staining with biotinylated 2R-B mAb recognizing the IL-2R β-chain[13] plus PE-conjugated streptavidin (Biomeda Corp., CA) as described previously.[3]

Radiolabeled IL-2 binding assay Radiolabeled IL-2 binding assay was performed as described previously.[14] Recombinant human IL-2 (kindly provided by Takeda Chemical Industries, Osaka) was radiolabeled by the chloramine T method. The number of IL-2 binding sites and the affinity were estimated by Scatchard plot analysis.

Chemical crosslinking of ^{125}I-labeled IL-2 The cells (5–10×10^6) were incubated with 5 n*M* ^{125}I-labeled IL-2 at 4°C for 1 h. Subsequently, chemical crosslinking using disuccinimidyl suberate as a crosslinker was performed according to the method of Brenner *et al.*[15] with slight modifications. The cell extracts were analyzed by SDS-PAGE as described.

Proliferative response to IL-2 Proliferation of leukemic cells from ATL patients and normal PHA blasts in response to IL-2 was measured in terms of [^{3}H]TdR incorporation as described previously.[14] In brief, 1×10^5 cells in a total volume of 200 μl of RPMI 1640 medium containing 10% FCS were cultured in a 96-well flat-bottomed plate in the presence of various concentrations of recombinant human IL-2 for 72 h. Cells were pulsed with 18.5 kBq/well [^{3}H]TdR (Amersham International, Buckinghamshire) for the last 6 h and harvested on glass fiber filters. The radioactivity was evaluated by liquid scintillation counting.

Southern blot hybridization DNA (5 μg) extracted from each sample was digested with a restriction endonuclease (*Eco* RI, *Bam* HI, *Hinc* II, *Hind* III or *Pst* I) (Toyobo Co., Ltd., Osaka), electrophoresed in a 0.7% agarose gel, transferred to nylon membrane and baked. Hybridization was performed with the ^{32}P-labeled probe and, after washing, autoradiography was performed as described previously.[16] The IL-2R β-chain cDNA probes which encode nucleotide positions 118–2,330 and nucleotide positions 1,600–3,572 of the published sequence[17] were provided by Dr. Honjo (Kyoto University, Kyoto).

RESULTS

Radiolabeled IL-2 binding assay Radiolabeled IL-2 binding assay of ATL cells (Table I) demonstrated 150–2,500/cell high-affinity (*Kd*: 3.5–25 p*M*) and 1,600–21,000/cell low-affinity (*Kd*: 6.8–77 n*M*) IL-2 binding sites. In 7-day cultured PHA blasts, 2,800/cell high-affinity (*Kd*: 18 p*M*) and 19,000/cell low-affinity (*Kd*: 8.1 n*M*) IL-2 binding sites were demonstrated, although only 650/cell intermediate affinity (*Kd*: 0.82 n*M*) IL-2 binding sites were detected in normal resting non-B non-adherent cells. From the results of flow cytometric analysis as described below, cells expressing intermediate affinity IL-2 binding sites seem to be CD56(+) and/or CD8(+) cells which solely express the IL-2R β-chain.[18–21]

Table I. Clinical Data, IL-2 Binding Sites and Cell Surface Markers of ATL Patients

Cells		Age	Sex	WBC (/mm³)	IL-2 binding sites				Flow cytometric analysis			
					High		Low		Positive cells (%)			
					/cell	*Kd* (p*M*)	/cell	*Kd* (n*M*)	Tac	2R-B	CD2	CD4
ATL												
case	MK	76	M	23,300	250	8	10,000	25	81.3	55.8	92.8	88.8
	TT	54	M	107,400	1,200	3.5	21,000	12	86.6	95.2	99.9	98.7
	HM	78	F	66,900	400	10	14,000	20	98.1	88.1	99.3	98.8
	IK	60	M	151,500	600	21	4,000	77	81.9	42.8	99.3	97.8
	KT	46	M	35,000	500	25	2,700	13	18.3	33.8	94.4	89.3
	SK	44	M	44,000	1,500	3.8	20,000	74	77.2	33.8	97.2	93.1
	NK	65	M	52,800	560	13.3	10,800	30	47.9	33.9	98.7	98.4
	EH	66	M	93,900	600	11	7,650	17	39.4	ND	ND	90.8
	TI	62	F	119,500	150	18	5,850	20	85.5	2.4	92.4	90.1
	YS	35	M	249,200	180	ND	1,600	ND	76.4	10.6	92.7	89.2
	AN	59	M	51,100	480	12.9	6,200	6.8	94.5	48.6	96.1	96.2
	KH	35	F	14,300	2,500	8	15,000	10	86.9	78.3	96.7	92.2
Normal PBMC (n=2)						650/cell 0.82 n*M* (intermediate)						
PHA blasts (7-day-cultured, n=2)					2,800	18	19,000	8.1				

ND: Not done.

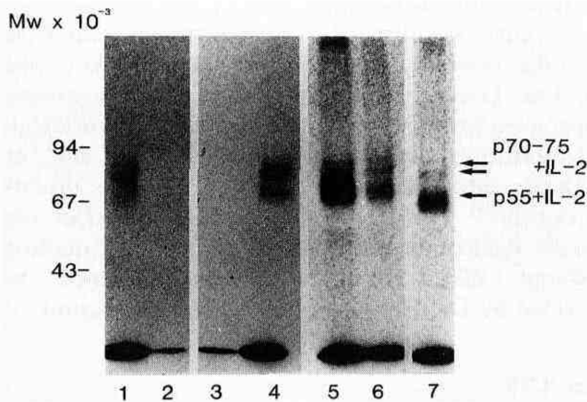

Fig. 1. SDS-PAGE of the cell extracts after radiolabeled IL-2 crosslinking to PBMC from patient KT (lanes 1, 2), patient IK (lanes 3, 4), patient TI (lane 5), PHA blasts (lane 6) and HTLV-I-infected cell line Hut102 (lane 7). Cells were incubated with radiolabeled IL-2 in the absence (lanes 1, 4, 5, 6, 7) or the presence (lanes 2, 3) of a 250-fold excess amount of unlabeled IL-2.

Chemical crosslinking of radiolabeled IL-2 To evaluate the molecular size of IL-2 binding proteins on ATL cells, ^{125}I-labeled IL-2 affinity crosslinking followed by SDS-PAGE analysis was performed in leukemic cells from 3 ATL patients (patients KT, IK, TI) (Fig. 1). Two major bands with apparent molecular weights (Mw) of 70,000–75,000 and 85,000–90,000 were visible in the 3 ATL cases examined and PHA blasts. The latter band seemed to be a doublet. These two bands represented the IL-2R α-chain (Mw 55,000) and the IL-2R β-chain (Mw 70,000–75,000) associated with an IL-2 molecule (Mw 15,500), respectively.

Flow cytometric analysis of normal PBMC and ATL cells Flow cytometric analysis of PBMC from 10 normal healthy volunteers was performed using Leu3a (CD4), Leu2a (CD8), Leu19 (CD56), anti-Tac (CD25) and 2R-B mAb. The IL-2R β-chain was expressed on 18.9–44.2% of unstimulated normal PBMC examined (mean$\pm$SD: 30.5$\pm$7.9%, n=10). Two-color immunofluorescence analysis demonstrated that 2R-B(+) cells

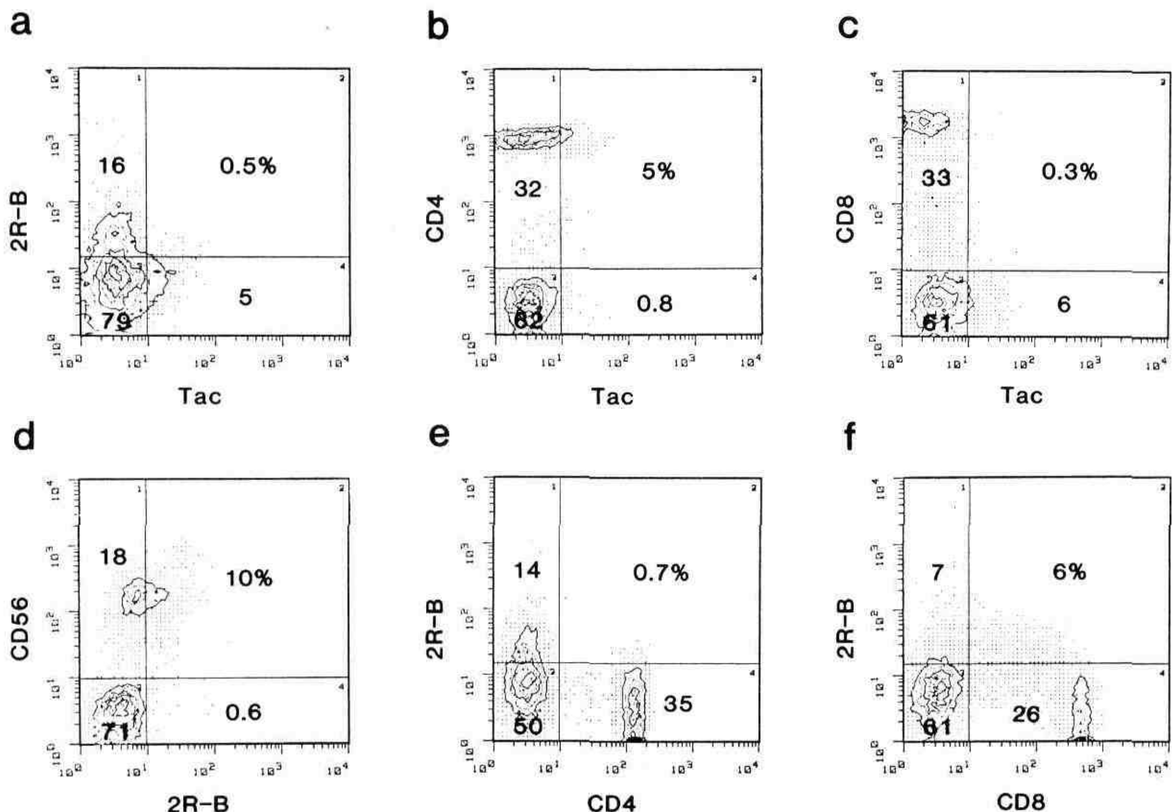

Fig. 2. Flow cytometric analysis of normal unstimulated PBMC following dual staining. PBMC from a healthy volunteer were stained with FITC-conjugated anti-Tac (a, b, c), CD4 (e), CD8 (f), PE-conjugated CD4 (b), CD8 (c), CD56 mAb (d) and biotinylated 2R-B mAb plus streptavidin-PE (a, e, f) or -FITC (d). Then cells were analyzed with the FACScan.

were contained mainly in CD56(+) and partially in CD8(+) cell population. The 2R-B(+) cell population did not seem to express the IL-2R α-chain (Fig. 2). A small number of unstimulated PBMC (3.7–15.5%, mean±SD: 8.25±3.27%, n=10) expressed the IL-2R α-chain. These Tac(+) cells were detected exclusively in CD4(+) cell population and did not express a detectable amount of IL-2R β-chain.

We previously reported that abnormally expanded large granular lymphocytes (LGL) constitutively expressed the IL-2R β-chain.[21] The present results suggest that unstimulated normal CD56(+) cells also constitutively express IL-2R β-chain although the expression level of IL-2R β-chain on abnormally expanded LGL is about 2 times higher than that on normal LGL as determined from the mean fluorescence intensity.

In contrast to normal CD4(+) T cells, various percentages of PBMC from ATL patients examined expressed IL-2R β-chain as shown in Table I. Two-color fluorescence analysis with FITC-Leu3a (CD4), FITC-

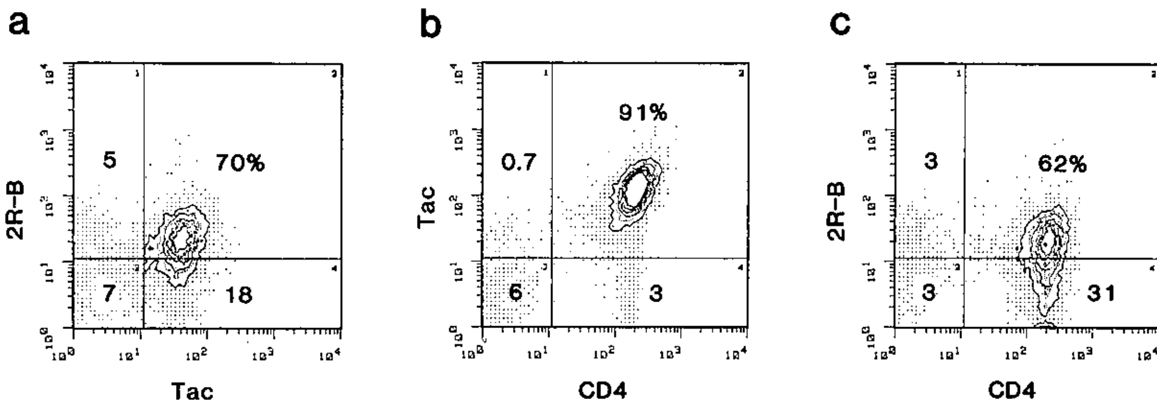

Fig. 3. Flow cytometric analysis of ATL cells following dual staining. PBMC from case KH were stained with FITC-conjugated anti-Tac (a, b), CD4 (c), PE-conjugated CD4 mAb (b) and biotinylated 2R-B mAb plus streptavidin-PE (a, c) and then analyzed with the FACScan.

Table II. Proliferative Response to IL-2 in ATL cells

Cells		[³H]TdR incorporation (cpm±SD)		
		Medium alone	0.5 n*M* IL-2	62.5 n*M* IL-2
ATL				
case	MK	6,393±966	13,151±262	11,648±501
	TT	1,847±336	1,825±92	545±70
	HM	382±24	6,870±976	7,422±31
	IK	796±74	1,292±33	1,550±155
	KT	474±169	735±32	931±142
	SK	147±24	767±125	695±234
	NK	1,446±449	2,446±130	2,550±176
	EH	473±252	4,636±1,545	1,579±103
	TI	268±58	1,042±97	1,092±184
	YS	154±9	785±123	709±38
	AN	455±252	450±46	1,390±52
	KH	2,030±581	12,068±2,811	10,645±2,639
7-day-cultured PHA blasts (n=7)		57±31	31,010±9,798	37,015±10,127

[³H]TdR incorporation of each case was examined at least at 6 different concentrations of IL-2 (0.01–62.5 n*M*). [³H]TdR incorporations at 2 concentrations (0.5 n*M* and 62.5 n*M*) of IL-2 are shown in this table.

anti-Tac and biotinylated 2R-B plus streptavidin-PE revealed that the IL-2R α-chain-bearing cells were CD4(+) and 2R-B(+) cells (Fig. 3). Thus, freshly isolated leukemic cells from ATL patients expressed both IL-2R α- and β-chain.

Proliferative response to IL-2 Table II shows the proliferative response of ATL cells to IL-2. Normal PBMC cultured with PHA-P for 7 days responded well to IL-2. [^{3}H]TdR incorporation of PHA blasts in the absence of IL-2 was 57±31 cpm (mean±SD, n=7) and it reached 37,015±10,127 cpm in the presence of 62.5 n*M* IL-2. In 6 of 12 ATL cases examined, [^{3}H]TdR incorporation of ATL cells in the presence of IL-2 was at most three times higher than the medium control. In three cases (SK, TI, YS), [^{3}H]TdR incorporation in the presence of IL-2 was about five times higher than that of the medium control. We did not regard these cases as responsive cases because the maximum incorporation of each case was less than 1,000 cpm and this level of maximum incorporation was very low compared with that of PHA blasts. In three other cases (HM, EH, KH), [^{3}H]TdR incorporation was more than five times higher than that of the medium control and maximum incorporation was more than 4,500 cpm. Thus, we regarded these three cases as responsive cases. As reported previously,[22] leukemic cells from about 25% of the ATL patients that we examined in the present study responded to IL-2. There was no correlation between the number of high-affinity IL-2Rs and the responsiveness to IL-2.

Southern blot hybridization Southern blot analysis was performed on leukemic cells from the 12 patients shown in Table I and an additional 50 ATL patients using *Eco* RI restriction endonuclease. Furthermore, DNAs from some cases were examined with other restriction endonucleases (*Bam* HI, *Hin*c II, *Hin*d III and *Pst* I). No gross rearrangement or obvious amplification of IL-2R β-chain gene was detected in any of these ATL patients (data not shown).

DISCUSSION

In the present studies we have demonstrated IL-2R β-chain expression on fresh leukemic cells from ATL patients by radiolabeled IL-2 binding assay, chemical crosslinking of radiolabeled IL-2 and flow cytometric analysis using 2R-B mAb, which recognizes IL-2R β-chain. Radiolabeled IL-2 binding assay showed both high-affinity and low-affinity receptors and chemical crosslinking studies also revealed two bands representing p70–75 and p55 (Tac) associated with radiolabeled IL-2, respectively. In addition, flow cytometric analysis using 2R-B mAb confirmed the expression of IL-2R β-chain on ATL cells. Recent studies have revealed another IL-2 binding protein (β-chain) which represents an intermediate-affinity IL-2 receptor, forms a high-affinity receptor together with α-chain (p55, Tac)[7, 8] and is considered to play an important role in signal transduction initiated by IL-2 binding to its receptor. A large intracytoplasmic domain of β-chain in contrast to only 13 amino acid residues of α-chain,[17] also suggested an important role of β-chain. Flow cytometric analysis using 2R-B mAb revealed that β-chain is mainly expressed on CD 8 (+) and/or CD 56 (+) unstimulated normal PBMC.

Expression of β-chain but not α-chain on LGL or LGL leukemia cells[18, 19, 21] and atypical CD8(+) T lymphocytes from infectious mononucleosis patients[23] was reported and the function in the early phase of the IL-2-dependent proliferation of such cells has been clarified. However, it has been unclear whether functionally important IL-2R β-chain molecule is expressed on ATL cells which abnormally express IL-2R α-chain. Our present studies have clearly demonstrated the expression of IL-2R β-chain on ATL cells. IL-2R β-chain expression on ATL cells is considered abnormal in the sense that normal unstimulated CD4(+) T cells do not express IL-2R β-chain and require stimulation to express a detectable amount of β-chain. Although IL-2R β-chain expression in ATL cells strongly suggests its close association with HTLV-I infection, it remains to be determined whether HTLV-I viral products, especially p40^{tax}, which can indirectly activate several cellular genes *in vitro* including IL-2, IL-2R α-chain, IL-3, IL-4 and GM-CSF genes,[24] are involved in the abnormal expression of IL-2R β-chain.

With regard to the possible role of abnormally expressed IL-2R β-chain in the leukemogenesis of ATL or neoplastic growth of ATL cells, we do not have any direct evidence. In spite of the expression of a considerable number of high-affinity receptors, leukemic cells from only 3 of 12 ATL patients examined proliferated in response to IL-2. In addition, there seemed to be no clear correlation between the number of high-affinity IL-2 receptors and the responsiveness of the leukemic cells to IL-2. These results indicate that the binding of IL-2 to its receptor does not lead to the induction of DNA synthesis, at least in most of the peripheral blood leukemic cells from the majority of fully developed ATL patients. IL-2R α- and β-chain on ATL cells may not produce and transmit signals although they can bind IL-2. Or, although they can produce signals, they may work negatively.[25] Thus, stimulation by IL-2 may not be apparent. Or, as cultured leukemic cells from some ATL patients may produce a small but sufficient amount of IL-2 for their growth,[26] growth of these cells may not be further stimulated by exogenous IL-2. Alternatively, there may be some abnormalities in intracytoplasmic

sequential events which are normally induced by IL-2 binding to its receptor and eventually lead to cell growth.[27] Abnormally expressed IL-2R β-chain as well as α-chain may be critically involved, instead, in the early stage of the development of ATL.

ACKNOWLEDGMENTS

This study was supported by Grants-in-Aid for Scientific Research from the Ministry of Education, Science and Culture, Japan.

(Received April 7, 1990/Accepted June 19, 1990)

REFERENCES

1) Uchiyama, T., Yodoi, J., Sagawa, K., Takatsuki, K. and Uchino, H. Adult T cell Leukemia: clinical and hematologic features of 16 cases. *Blood,* **50,** 481–492 (1977).
2) Hinuma, Y., Nagata, K., Hanaoka, M., Nakai, M., Mitsumoto, T., Shirakawa, S. and Miyoshi, I. Adult T-cell leukemia: antigen in an ATL cell line and detection of antibodies to the antigen in human sera. *Proc. Natl. Acad. Sci. USA,* **78,** 6476–6480 (1981).
3) Uchiyama, T., Hori, T., Tsudo, M., Wano, Y., Umadome, H., Tamori, S., Yodoi, J., Maeda, M., Sawami, H. and Uchino, H. Interleukin-2 receptor (Tac) expressed on adult T cell leukemia cells. *J. Clin. Invest.,* **76,** 446–453 (1985).
4) Inoue, J., Seiki, M., Taniguchi, T., Tsuru, S. and Yoshida, M. Induction of interleukin 2 receptor gene expression by $p40^x$ encoded by human T-cell leukemia virus type I. *EMBO J.,* **5,** 2283–2288 (1986).
5) Sharon, M., Klausner, R. D., Cullen, B. R., Chizzonite, R. and Leonard, W. J. Novel interleukin 2 receptor subunit detected by cross-linking under high-affinity conditions. *Science,* **234,** 859–863 (1986).
6) Tsudo, M., Kozak, R. W., Goldman, C. K. and Waldmann, T. A. Demonstration of non-Tac peptide that binds interleukin 2: a potential participant in a multichain interleukin 2 receptor complex. *Proc. Natl. Acad. Sci. USA,* **83,** 9694–9698 (1986).
7) Teshigawara, K., Wang, H. M., Kato, K. and Smith, K. A. Interleukin 2 high-affinity receptor expression requires two distinct binding proteins. *J. Exp. Med.,* **165,** 223–238 (1987).
8) Robb, R. J., Rusk, C. M., Yodoi, J. and Greene, W. C. Interleukin 2 binding molecule distinct from the Tac protein: analysis of its role in formation of high-affinity receptors. *Proc. Natl. Acad. Sci. USA,* **84,** 2002–2006 (1987).
9) Hattori, T., Uchiyama, T., Toibana, T., Takatsuki, K. and Uchino, H. Surface phenotype of Japanese adult T-cell leukemia cells characterized by monoclonal antibodies. *Blood,* **58,** 645–647 (1981).
10) Yoshida, M., Seiki, M., Yamaguchi, K., and Takatsuki, K. Monoclonal integration of human T-cell leukemia provirus in all primary tumors of adult T-cell leukemia suggests causative role of human T-cell leukemia virus in the disease. *Proc. Natl. Acad. Sci. USA,* **81,** 2534–2537 (1984).
11) Uchiyama, T., Broder, S. and Waldmann, T. A. A new monoclonal antibody (anti-Tac) reactive with activated and functionally mature human T cells. I. Production of anti-Tac monoclonal antibody and distribution of Tac(+) cells. *J. Immunol.,* **126,** 1393–1397 (1981).
12) Uchiyama, T., Nelson, T. D., Fleisher, T. A. and Waldmann, T. A. A monoclonal antibody (anti-Tac) reactive with activated and functionally mature human T cells. II. Expression of Tac antigen on activated cytotoxic killer T cells, suppressor cells, and on one of two types of helper T cells. *J. Immunol.,* **126,** 1398–1403 (1981).
13) Kamio, M., Uchiyama, T., Arima, N., Itoh, K., Ishikawa, T., Hori, T. and Uchino, H. Role of α chain/IL-2 complex in the formation of the ternary complex of IL-2 and high-affinity IL-2 receptor. *Int. Immunol.,* in press.
14) Hori, T., Uchiyama, T., Umadome, H., Tamori, S., Tsudo, M., Araki, K. and Uchino, H. Dissociation of interleukin-2-mediated cell proliferation and interleukin-2 receptor upregulation in adult T-cell leukemia cells. *Leuk. Res.,* **10,** 1447–1453 (1986).
15) Brenner, M. B., Trowbridge, I. S. and Strominger, J. L. Cross-linking of human T cell receptor proteins: association between the T cell idiotype β subunit and the T3 glycoprotein heavy subunit. *Cell,* **40,** 183–190 (1985).
16) Southern, E. M. Detection of specific sequences among DNA fragments separated by gel electrophoresis. *J. Mol. Biol.,* **98,** 503–517 (1975).
17) Hatakeyama, M., Tsudo, M., Minamoto, S., Kono, T., Doi, T., Miyata, T., Miyasaka, M. and Taniguchi, T. Interleukin-2 receptor β chain gene: generation of three receptor forms by cloned human α and β chain cDNA's. *Science,* **244,** 551–556 (1989).
18) Dukovich, M., Wano, Y., Thuy, L. T. B., Katz, P., Cullen, B. R., Kehrl, J. H. and Greene, W. C. A second human interleukin-2 binding protein that may be a component of high affinity interleukin-2 receptors. *Nature,* **327,** 518–522 (1987).
19) Tsudo, M., Goldman, C. K., Bongiovanni, K. F., Chan, W. C., Winton, E. F., Yagita, M., Grimm, E. A. and Waldman, T. A. The p75 peptide is the receptor for interleukin 2 expressed on large granular lymphocytes and responsible for the interleukin 2-activation of these cells. *Proc. Natl. Acad. Sci. USA,* **84,** 5394–5398 (1987).
20) Siegel, J. P., Sharon, M., Smith, P. L. and Leonard, W. J. The IL-2 receptor β chain (p70): role in mediating signals for LAK, NK, and proliferating activities. *Science,* **238,** 75–78 (1987).
21) Hori, T., Uchiyama, T., Onishi, R., Kamio, M.,

Umadome, H., Tamori, S., Motoi, T., Kodaka, T. and Uchino, H. Characteristics of the IL-2 receptor expressed on large granular lymphocytes from patients with abnormally expanded large granular lymphocytes: implication of a non-Tac IL-2-binding peptide. *J. Immunol.*, **140**, 4199–4203 (1988).
22) Uchiyama, T., Kamio, M., Kodaka, T., Tamori, S., Fukuhara, S., Amakawa, R., Uchino, H. and Araki, K. Leukemic cells from some adult T-cell leukemia patients proliferate in response to interleukin-4. *Blood*, **72**, 1182–1186 (1988).
23) Kamio, M., Uchiyama, T., Hori, T., Kodaka, T., Ishikawa, T., Onishi, R., Uchino, H., Yoneda, N., Tatsumi, E. and Yamaguchi, N. Selective expression of the p70 subunit of the interleukin-2 receptor on lymphocytes from patients with infectious mononucleosis. *Blood*, **75**, 415–420 (1990).
24) Miyajima, A., Miyatake, S., Schreurs, J., de Vries, J., Arai, N., Yokota, T. and Arai, K. Coordinate regulation of immune and inflammatory responses by T cell-derived lymphokines. *FASEB J.*, **2**, 2462–2473 (1988).
25) Sugamura, K., Nakai, S., Fujii, M. and Hinuma, Y. Interleukin 2 inhibits *in vitro* growth of human T cell lines carrying retrovirus. *J. Exp. Med.*, **161**, 1243–1248 (1985).
26) Arima, N., Daitoku, Y., Ohgaki, S., Fukumori, J., Tanaka, H., Yamamoto, Y., Fujimoto, K. and Onoue, K. Autocrine growth of interleukin 2-producing leukemic cells in a patient with adult T cell leukemia. *Blood*, **68**, 779–782 (1986).
27) Churilla, A. M. and Braciale, V. L. Lack of IL-2-dependent proliferation despite significant expression of high-affinity IL-2 receptors on murine cytolytic T lymphocyte clones late after antigenic stimulation. *J. Immunol.*, **138**, 1338–1345 (1987).