Arch Dermatol Res (1998) 290 : 681–687

ORIGINAL PAPER

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Defective IL-2 receptor expression in lymphocytes of patients with arsenic-induced Bowen's disease

Received: 18 September 1997 / Received after revision: 19 August 1998 / Accepted: 27 August 1998

Abstract **The immune function of peripheral mononuclear cells (MNC) in patients with endemic arsenic-induced Bowen's disease (BD) was investigated. Many cytokines and immune-related factors were determined in the present study. Interleukin-1β and TNF-α production was used as an indicator of monocyte/macrophage function. Il-2 and sIL-2R production was used as an indicator of lymphocyte activation. The release of sCD4 and sCD8 was used as an indicator of activation of respective T-cell subpopulations. Production of IFN-γ and IL-2 reflected the cellular effector function of helper T-cells type 1. In vivo cell-mediated immunity was also assessed by estimation of the percentage of T-cells in peripheral blood MNC and the nonspecific delayed-type hypersensitivity (DTH) response to 2,4-dinitrochlorobenzene (DNCB). Both assays revealed depressed cell-mediated immunity in BD. Compared with healthy controls, spontaneous and PHA-induced IFN-γ and TNF-α production was significantly decreased in BD whereas spontaneous release of IL-2, sCD4 and sCD8 was significantly increased. Although PHA stimulation increased IL-2 release, the expression of IL-2R α and β chains and the release of sIL-2R were not proportionately increased in BD. In addition, IL-2-mediated [^{3}H]-thymidine incorporation by MNC in patients with BD was significantly decreased. These findings suggest that the defective cell-mediated immune function in BD is due to impairment of membrane IL-2R expression in lymphocytes after stimulation.**

Key words T lymphocytes · Tumor immunity · IL-2 receptor · Bowen's disease

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Introduction

An endemic occurrence of skin cancers due to high concentrations of arsenic in artesian well water in a limited area of the southwest coast of Taiwan has been reported [1]. Arsenical skin cancers in the endemic area include Bowen's disease (carcinoma in situ), basal cell carcinoma, squamous cell carcinoma and their various combinations. In animals, the carcinogenic nature of arsenic has not yet been reliably and consistently demonstrated [2]. Chronic arsenical poisoning in the inhabitants of an area such as the above may, however, provide a human model of naturally occurring chemical carcinogenesis. Our previous investigations have demonstated a decrease in PHA-induced [^{3}H]-thymidine incorporation by peripheral mononuclear cells (MNC) in arsenic-induced Bowen's disease (BD) [3] as well as a considerable decline in the density of Langerhans cells, as compared with normal epidermis [4]. These findings indicate defective cell-mediated immunity in these patients.

It is not inconceivable that the host response to malignant tumors is primarily cell-mediated. Delayed-type hypersensitivity (DTH) response is induced by helper T-cell type 1 (Th1) and the status of the nonspecific cell-mediated immune response has been measured in response to 2,4-dinitrochlorobenzene (DNCB) [5]. Patients with BD and squamous cell carcinoma, but not those with basal cell carcinoma, show an impaired response to DNCB [6], thus implicating depressed cell-mediated immunity in some

skin cancer patients. Cytokines of Th1 cells, such as IFN-γ and IL-2, have been shown to be essential modulators of the cell-mediated immune response. T lymphocyte activation, proliferation and differentiation are critically dependent on the presence of IL-1 and IL-2 [7]. TNF-α, released mainly from monocytes/macrophages, is a cytotoxic molecule for a broad spectrum of tumor cells [8, 9].

Defects in the production of certain cytokines have been reported in patients with various solid tumors such as melanoma [10], colorectal cancer [11], gynecological carcinomas [12], and nasopharyngeal carcinoma [13]. It is believed that certain soluble molecules of membrane proteins, such as sIL-2R, sCD4 and sCD8, are released by activated T lymphocytes and serve as significant immunoregulatory molecules [14–16]. Recent studies have revealed an increase in sCD4 and sCD8 production in nasopharyngeal carcinoma [13] and sIL-2R, sCD4 and sCD8 levels in melanoma [17]. Accordingly, release of both cytokines and soluble membrane molecules can be used as indicators of the status of the cellular immunity in tumor patients.

In the present study, to assess the immune status of BD patients, we monitored the DTH response after application of DNCB, and determined the plasma sIL-2R levels, the percentage of each lymphocyte population (T-cells, activated T-cells and B-cells) and T-cell subpopulation (T-helper cells and T-cytotoxic/suppressor cells), Th1 cytokines (IFN-γ, TNF-α, IL-2), the T-cell-activating monokine (IL-1 β), and soluble T-cell surface molecules (sIL-2R, sCD4, sCD8) in these patients, and evaluated the expression of IL-2R on lymphocytes. Our results suggest that a defect in IL-2 receptor expression plays an important role in the immune dysfunction of arsenic-induced BD.

Materials and methods

Selection of patients

All 16 patients with BD were selected from an endemic area in the southwest coast of Taiwan. The mean age of the patients was 68.4 ± 6.5 years (range 61–77 years). The mean age of onset in these patients was 62.8 ± 8.0 years (range 54–74 years) and the average number of lesions was 9.0 ± 4.7 (range 2–14). The exposure time to arsenic in these patients was estimated at more than 20 years and no extracutaneous tumors were found. The average estimated level of arsenic in the water ingest patients in the endemic area was 7.3×10^6 ppm. Groups of 16 age- and sex-matched healthy volunteers from the endemic area (N-BD controls) and a nonendemic areas (normal controls) were selected. The mean urinary arsenic levels in the BD patients and in the N-BD controls and normal controls were 201.1 ± 57.6, 75.5 ± 39.1 and 63.4 ± 29.7 ng/ml, respectively, as determined by atomic absorption spectrophotometry. None of the patients had been medically treated before the study.

Induction of contact sensitization

Measurements of DTH response were conducted using a previously reported method with modification [18, 19]. Briefly, DNCB (Sigma Chemical, St. Louis, Mo.) was diluted to 0.1% in acetone and applied to the upper back epicutaneously using Finn chambers (Epitest, Helsinki) for 24–48 h. After 14 days, the same skin site was challenged with 0.01% DNCB. Only intense cutaneous responses (such as erythema, edema and/or vesicle formation) at the treated site were considered to be positive.

Determination of sIL-2R levels in plasma

Plasma sIL-2R levels were determined using a sandwich enzyme immunoassay (T Cell Diagnostics, Cambridge, Mass.) according to the manufacturer's instructions.

Isolation of MNC

MNC were isolated from heparinized venous blood after centrifugation at 300 *g* for 30 min over a Ficoll-Hypaque cushion (specific gravity 1.077) as reported previously [20].

Flow cytometric determination of the percentages of lymphocyte populations and subpopulations

The percentages of lymphocyte populations and subpopulations in MNC were determined according to the method of Sindern et al. with modification [21]. Briefly, freshly obtained MNC (1.0×10^6 cells/ml) in 10% fetal bovine serum in RPMI-1640 medium (GIBCO, Gaithersburg, Md; 10% FBS-RPMI) were double-stained with antibodies against CD3-FITC/HLA-DR-PE (for activated T-cells) (Becton and Dickinson, San Jose, Calif.) or against CD4-FITC/CD8-PE (for T-helper cells and T-cytotoxic/suppressor cells) (Becton Dickinson) in an ice-bath for 30 min. After washing twice with PBS, pH 7.2, the percentages of positively stained cells were analysed in a flow cytometer (FACScan, Becton Dickinson).

Preparation of spontaneous and mitogen-stimulated MNC culture supernatants

The concentration of MNC was adjusted to 2.5×10^6 cells/ml in 10% FBS-RPMI. Then 0.5 ml 10% FBS-RPMI (spontaneous) or 0.4 ml 10% FBS-RPMI and 0.1 ml phytohemagglutinin L form (PHA, 10 μg/ml, Sigma) (PHA-induced) was added to each tube in triplicate. The mixture was incubated at 37° C in an atmosphere comprising 5% CO_2 and 95% air for 48 h, then centrifuged at 2000 rpm for 10 min and the cell-free supernatants stored at –20° C.

Measurement of cytokines and soluble T-cell surface antigens in culture supernatants

Cytokines and soluble T-cell surface antigens were determined using commercially available ELISA kits. They were assayed using colorimetric enzyme immunoassays according to the manufacturer's instructions. The cytokines and antigens determined included: IL-1β, IL-2, TNF-α and IFN-γ (Quantikine, R&D System, Minneapolis, Minn.), and sIL-2R (T Cell Diagnostics), sCD4 and sCD8 (T Cell Sciences, Woburn, Mass.).

Measurement of recombinant human IL-2-induced [^{3}H]-thymidine incorporation by MNC

The uptake of [^{3}H]-thymidine by MNC induced by recombinant human IL-2 (rIL-2, Biosource, Camarillo, Calif.) was measured using a modified version of a method reported previously [22]. Briefly, 0.05 ml of a suspension of MNC (2×10^6 cells/ml) was placed in each well of a microtiter plate in triplicate. Then 50 μl 10% FBS-RPMI or 50 μl rIL-2 (6 U/μl) was added to each well. The mixture was incubated at 37° C in an atmosphere comprising 5% CO_2 and 95% air for 72 h. After incubation, each microwell was pulsed with 0.5 μCi (specific activity 6.7 Ci/mmol) methyl[^{3}H]-thymidine (Dupont, Boston, Mass.) in a volume of 10 μl for 6 h. Then the cells were harvested and the radioactivity was measured in a liquid scintillation counter (Minax-B Tri-Carb 4000 series, Packard, Gowners Grove, Ill.)

Table 1 Percentages of T-cells, T-helper cells and B-cells in the BD patients and the two control groups

Group	T-cells	Activated T-cells	B-cells	T-helper cells	T-cytotoxic/ suppressor cells
Normal controls	63.1 ± 12.7	23.8 ± 9.4	17.7 ± 6.6	31.4 ± 5.7	31.1 ± 5.8
N-BD controls	68.6 ± 1.6	29.2 ± 9.3	17.2 ± 1.1	39.6 ± 1.8	28.5 ± 4.0
BD patients	46.9 ± 18.8*	26.1 ± 16.2	29.7 ± 14.5**	19.5 ± 5.5*	26.7 ± 20.7

* $P < 0.02$; BD patients vs normal controls and N-BD controls; ** $P < 0.05$, BD patients vs normal controls and N-BD controls

Table 2 Spontaneous and PHA-induced IFN-γ, TNF-α and IL-2 release in the BD patients and the two control groups

Group	IFN-γ (pg/ml)	TNF-α (pg/ml)	IL-2 (pg/ml)	IL-1 β (pg/ml)
Normal controls				
Spontaneous	126.1 ± 28.8	107.1 ± 21.5	5.4 ± 1.1	124.1 ± 25.3
PHA-induced	623.5 ± 94.9	239.3 ± 110.0	17.3 ± 4.0	165.7 ± 30.8
N-BD controls				
Spontaneous	108.0 ± 32.1	90.2 ± 27.6	6.1 ± 1.1	109.1 ± 31.1
PHA-induced	571.2 ± 101.1	257.3 ± 129.0	19.2 ± 5.1	125.1 ± 36.7
BD patients				
Spontaneous	7.8 ± 1.5*	68.7 ± 16.1*	14.4 ± 6.9*	119.9 ± 36.7
PHA-induced	22.9 ± 3.8**	84.1 ± 19.3**	73.9 ± 34.5**	151.1 ± 20.3

* $P < 0.005$, BD patients vs normal controls and N-BD controls (spontaneous release); ** $P < 0.005$, BD patients vs normal controls and N-BD controls (PHA-induced)

Determination of IL-2R α chain and β chain on MNC

The expression of IL-2R α chain and β chain on MNC was assessed according to the method of Kierszenbaum and Szteins [23]. Briefly, freshly obtained MNC in 10% FBS-RPMI (1.0×10^6 cells/ml) were stimulated with PHA (10 μg/ml) for 3 days. Then they were stained with antibodies against CD25-FITC (for p55) and against CD122-PE (for p75) (Becton Dickinson) in an ice-bath for 30 min. After washing twice with PBS buffer, the stained cells were determined in a flow cytometer. Mean fluorescence intensity (MFI) was used as a measure of the expression of IL-2R α and β chains on T cells.

Statistical analysis

All values presented are means ± SD. Statistical significance of differences was assessed using the unpaired Student's *t*-test between the different groups while the comparisons between the percentages of lymphocyte populations and subpopulations among the BD patients, normal controls and N-BD controls were analysed using Student's *t*-test. A *P*-value of < 0.05 was considered as statistically significant.

Results

Low DTH response after DNCB challenge was observed in BD patients

All of the studied subjects showed a positive response to the epicutaneous application of 0.1% DNCB. However, only BD patients showed a negative response to challenge with 0.01% DNCB. Apparently, the DTH response in BD patients was weaker than in controls.

Plasma sIL-2R levels in BD patients were similar to the levels in normal controls

There were no significant differences in sIL-2R levels in plasma among BD patients (61.3 ± 5.2 pmol/l), normal controls (57.4 ± 3.0 pmol/l) and N-BD controls (51.5 ± 5.3 pmol/l).

The percentages of T-cells and T-helper cells were decreased and the percentage of B-cells was increased in BD patients compared with controls

As shown in Table 1, the percentages of T-cells and T-helper cells (46.9 ± 18.8% / 19.5 ± 5.5%) in BD patients were significantly lower ($P < 0.02$) than in normal controls and N-BD controls (63.1 ± 12.7% / 31.4 ± 5.7% and 68.6 ± 1.6% / 39.6 ± 1.8%, respectively). The percentage of B-cells (29.7 ± 14.5%) in BD patients was significantly higher ($P < 0.05$, Table 1) than in normal controls and N-BD controls (17.7 ± 6.6% and 17.2 ± 1.1%, respectively). The percentages of activated T-cells and T-cytotoxic/suppressor cells were similar in BD patients (26.1 ± 16.2% / 26.7 ± 20.7%), normal controls and N-BD controls (23.8 ± 9.4% / 31.1 ± 5.8% and 29.2 ± 9.3% / 28.5 ± 4.0%, respectively).

Spontaneous and PHA-induced IFN-γ and TNF-α production were decreased and IL-2 release was increased in BD patients

The spontaneous and PHA-induced production of IFN-γ, TNF-α, IL-2 and IL-1 β in N-BD controls were not different from those in normal controls (Table 2).

A significant decrease in spontaneous IFN-γ production was observed in BD patients (7.8 ± 1.5 pg/ml) as compared with normal controls and N-BD controls (126.1 ± 28.8 pg/ml and 108.0 ± 32.1 pg/ml; $P < 0.005$; Table 2). PHA induced a significant increase in IFN-γ production in normal controls and N-BD controls (623.5 ± 94.9 pg/ml and 571.2 ± 101.1 pg/ml) compared with the level in BD patients (22.9 ± 3.8 pg/ml; $P < 0.005$, Table 2).

A significant decrease in spontaneous TNF-α production was found in BD patients (68.7 ± 16.1 pg/ml) as compared with normal controls and N-BD controls (107.1 ± 21.5 pg/ml and 90.2 ± 27.6 pg/ml; $P < 0.005$, Table 2). PHA stimulation increased TNF-α production in normal controls and N-BD controls (239.3 ± 110.0 pg/ml and 257.3 ± 129.0 pg/ml), but not in BD patients (84.1 ± 19.3 pg/ml). Thus, compared with controls, BD patients responded poorly to PHA stimulation as far as TNF-α production was concerned ($P < 0.005$).

A significant increase in spontaneous IL-2 production was seen in BD patients (14.4 ± 6.9 pg/ml) compared with normal controls and N-BD controls (5.4 ± 1.1 pg/ml and 6.1 ± 1.1 pg/ml; $P < 0.005$, Table 2). PHA stimulation increased the production of IL-2 in BD patients (73.9 ± 34.5 pg/ml) compared with normal controls and N-BD controls (17.3 ± 4.0 pg/ml and 19.2 ± 5.1 pg/ml; $P < 0.005$, Table 2).

The spontaneous production of IL-1β in BD patients was not different from that in normal controls and N-BD controls. Upon stimulation with PHA, levels of IL-1β in BD patients remained similar to those in controls (Table 2).

Spontaneous sCD4 and sCD8 production was increased and PHA-induced sCD4, sCD8 and sIL-2R release was decreased in BD patients

The spontaneous and PHA-induced production of sIL-2R, sCD4 and sCD8 in N-BD controls was not different from that in normal controls (Table 3).

Spontaneous release of sIL-2R was not significantly different among patients (3.2 ± 1.0 pmol/ml), normal controls (4.3 ± 2.3 pmol/ml) and N-BD controls (3.8 ± 2.4 pmol/ml). PHA stimulation increases sIL-2R release in normal controls and N-BD controls (120.8 ± 17.6 pmol/ml and 109.0 ± 21.4 pmol/ml), but only a modest increase in BD patients (8.1 ± 4.3 pmol/ml). PHA induced a statistically significant increase in sIL-2R production in normal controls and N-BD controls, as compared with BD patients ($P < 0.005$, Table 3).

A spontaneous increase in sCD4 production was observed in BD patients (19.7 ± 2.3 U/ml) as compared with normal controls and N-BD controls (4.5 ± 1.9 U/ml and 3.2 ± 2.1 U/ml; $P < 0.005$). PHA stimulation increased sCD4 production in the normal controls and N-BD controls (22.6 ± 3.4 U/ml and 24.1 ± 5.2 U/ml), but not in BD patients (16.4 ± 4.3 U/ml; Table 3).

A spontaneous increase in sCD8 production was observed in BD patients (265.2 ± 78.1 U/ml) as compared with normal controls and N-BD controls (132.7 ± 46.1 U/ml and 112.3 ± 52.1 U/ml; $P < 0.005$). PHA stimulation increased sCD8 release in normal controls (597.3 ± 119.0 U/ml), N-BD controls (539.4 ± 65.2 U/ml) and BD patients (340.1 ± 92.8 U/ml). PHA-stimulated sCD8 release was significantly lower in patients than in controls ($P < 0.005$; Table 3).

Table 3 Spontaneous and PHA-induced release of sIL-2R, sCD4 and sCD8 in the BD patients and the two control groups

Group	sIL-2R (pmol/l)	sCD4 (U/ml)	sCD8 (U/ml)
Normal controls			
Spontaneous	4.3 ± 2.3	4.5 ± 1.9	132.7 ± 46.1
PHA-induced	120.8 ± 17.6	22.6 ± 3.4	597.3 ± 119.0
N-BD controls			
Spontaneous	3.8 ± 2.4	3.2 ± 2.1	112.3 ± 52.1
PHA-induced	109.0 ± 21.4	24.1 ± 5.2	539.4 ± 65.2
BD patients			
Spontaneous	3.2 ± 1.0	19.7 ± 2.3*	265.2 ± 78.1*
PHA-induced	8.1 ± 4.3**	16.4 ± 4.3**	340.1 ± 92.8**

*$P < 0.005$, BD patients vs normal controls and N-BD controls (spontaneous release);
**$P < 0.005$, BD patients vs normal controls and N-BD controls (PHA-induced)

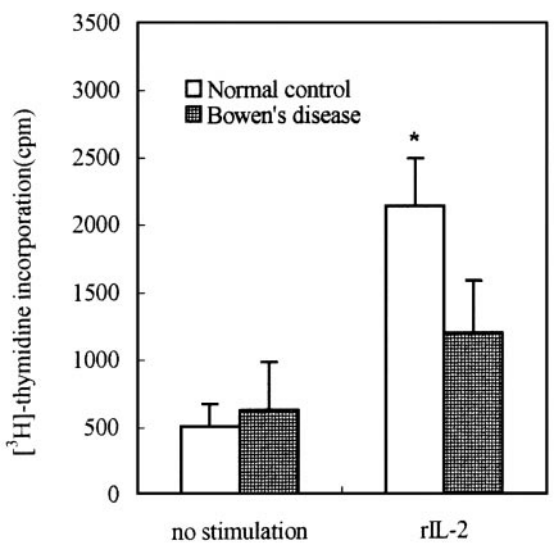

Fig. 1 Recombinant IL-2 (rIL-2) induced a significantly lower [^{3}H]-thymidine incorporation into MNC of Bowen's disease patients than into MNC of normal controls (*$P < 0.001$)

Recombinant IL-2-induced [^{3}H]-thymidine incorporation was lower in BD patients than in normal controls

As demonstrated in Fig. 1, the spontaneous incorporation of [^{3}H]-thymidine into MNC of BD patients (626 ± 353 cpm) and normal controls (504 ± 170 cpm) was not significantly different. However, after incubation with recombinant IL-2, [^{3}H]-thymidine uptake in BD patients (1191 ± 385 cpm) was significantly lower than in normal controls (2136 ± 355 cpm; $P < 0.001$).

PHA-induced IL-2R α and β chain expression on MNC of BD patients was lower than on MNC of controls

The expression of IL-2R α chain (343.1 ± 29.4 MFI) and β chain (204.2 ± 40.7 MFI) on T-cells of patients was not

Table 4 Spontaneous and PHA-induced expression of IL-2R α and β chains on T-cells in the BD patients and the normal control group (*MFI* mean fluorescence intensity)

Group	IL-2R α chain (MFI)		IL-2R β chain (MFI)	
	Spontaneous	PHA-induced	Spontaneous	PHA-induced
Normal controls	369.0 ± 80.6	465.0 ± 37.5*	225.1 ± 126.7	321.1 ± 233.5**
BD patients	343.1 ± 29.4	395.8 ± 30.5	204.2 ± 40.7	211.9 ± 15.7

* $P < 0.005$, normal controls vs BD patients (PHA-induced); ** $P < 0.001$, normal controls vs BD patients (PHA-induced)

different from that on T-cells of normal controls (369.0 ± 80.6 and 225.1 ± 126.7 MFI, respectively). However, PHA induced significantly lower IL-2R α chain (395.8 ± 30.5 MFI) and IL-2R β chain (211.9 ± 15.7 MFI) expression in BD patients than in normal controls (465.0 ± 37.5 and 321.1 ± 233.5 MFI; $P < 0.005$ and $P < 0.001$, respectively; Table 4).

Discussion

In the present study, we investigated cytokine production and immune responses in patients with BD and in healthy controls. The cutaneous response to nonspecific DTH antigen (DNCB) and the proportions of each lymphocyte population were determined as indicators of the in vivo immune status. Analysis of the results revealed depressed cell-mediated immunity in BD patients in the form of impaired DTH responses to DNCB and a reduction in T-cells and T-helper cell percentages, in comparison with controls. However, the proportion of B-cells was elevated in BD patients. This defective cell-mediated immunity in BD patients was consistent with a decreased release of IFN-γ which is produced by T-helper 1 cells. The plasma sIL-2R level is a sensitive and quantitative marker of circulating peripheral MNC activation [24] and also reflects the status of lymphocyte activation [25]. Thus, the similar percentage of activated T-cells among BD patients, normal controls and N-BD controls could, in part, explain the similar plasma sIL-2R level and spontaneous IL-2R release among the three groups.

PHA stimulation induced significant increases in sIL-2R, sCD4 and sCD8 in normal controls and N-BD controls, but not in BD patients. This suggests that T-cells in BD patients are relatively anergic in their response to mitogen stimulation compared with cells of the other two control groups. IL-1β and TNF-α production were used as indicators of monocyte/macrophage activation. sCD4 and sCD8 release were used as indicators of helper/inducer and suppressor/cytotoxic T-cell activation, respectively [13, 15–17]. IL-2 and IFN-γ production were taken as reflecting Th1 cell function. In this study, neither spontaneous nor PHA-induced IL-1β production differed among patients, normal controls or N-BD controls, but spontaneous and PHA-stimulated TNF-α production were decreased in BD patients. Thus, we believe that monocyte function may not be selectively defective in BD.

In BD patients, there was an increase in the spontaneous release of IL-2 (Table 2), sCD4 and sCD8 (Table 3) compared with the release in normal controls and N-BD controls, indicating a pre-excitation state of T lymphocytes in BD. In addition, PHA stimulated a marked increase in IL-2, but did not change the expression of membrane IL-2R (α and β chains, Table 4) and produced a negligible increase in sIL-2R production in BD patients compared with controls. Furthermore, IL-2-stimulated [^{3}H]-thymidine incorporation into MNC was also significantly decreased in BD patients compared with normal controls (Fig. 1). These findings suggest defective IL-2R expression and function in patients suffering from BD.

The soluble form of IL-2R is considered to be the product of a proteolytic cleavage of the membrane molecule. It can bind efficiently to IL-2 to maintain the homeostasis of the immune response [26]. Therefore, discrepancies in the interrelationships among IL-2 production, membrane IL-2R expression, and sIL-2R release upon PHA stimulation may indicate the intrinsic defects in lymphocytes that may lead to the impairment of IL-2-mediated function in these patients [27]. Defective lymphocyte function may also bring about a decline in spontaneous and PHA-stimulated IFN-γ and TNF-α production (Table 1).

Decreased IL-2R expression in BD patients leads to ineffective binding between IL-2 and IL-2R and depressed [^{3}H]-thymidine incorporation by MNC. IL-2R includes at least three distinct subunits, the α chain (55 kDa), the β chain (75 kDa) and the γ chain (64 kDa) [28]. Different combinations of the three subunits form the three classes of IL-2R. Low-affinity IL-2R consists of the α chain, intermediate-affinity IL-2R includes the β and γ chains, and high-affinity IL-2R contains all three chains. The IL-2R α chain can bind with IL-2 but has little to do with intracellular signal transduction. The IL-2R β chain plays a crucial role in intracellular signal transduction induced by IL-2. The IL-2R γ chain also participates in the mechanism of intracellular signal transduction once interaction with IL-2 has taken place. By measuring IL-2R α and β chains, we could evaluate the binding affinity between IL-2 and IL-2R [29]. Our findings show that the expression of IL-2R α and β chains in BD patients after PHA stimulation was less than that in normal controls. PHA is a macrophage-dependent T-cell activator, so the unchanged expression of IL-2R α and β chains after PHA stimulation indicates defective expression of IL-2R α and β chains by T-cells in patients with BD. This defect causes the deficient IL-2R function in patients with BD, which has not

been reported previously. From these findings we suggest that unsuccessful binding between IL-2 and IL-2R is caused by an abnormal expression of IL-2R on T-cells and brings about an immunological dysfunction in BD patients.

There are several possible explanations for the IL-2R defect in BD patients. First, long-term exposure of MNC to arsenic may impair IL-2R. Arsenic is known to exert a biphasic effect on the proliferation response of lymphocytes, that is, their proliferation is amplified at low arsenic concentrations and inhibited at high concentrations [30, 31]. Arsenic concentrations in urine, frequently used as an index of exposure [32], have been found to be significantly higher in BD patients from the endemic area than in healthy controls [33, 34]. Consequently IL-2R impairment in our BD patients may have been due to chronic arsenical poisoning in the endemic area. Second, certain immunosuppressive factors may be released in BD, impairing the synthesis of IL-2R in activated lymphocytes. Third, the cancer tissue may release certain IL-2R-specific binding factors or antagonists to block IL-2R on the surface of activated lymphocytes. However, the molecular mechanism of the IL-2R defect in BD remains to be studied.

Acknowledgements This study was supported by the National Science Council of the Republic of China (research grant NSC 84-2621-B-037-002-Z).

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