ANNALES
UNIVERSITATIS MARIE CURIE-SKŁODOWSKA
LUBLIN-POLONIA
VOL. LVIII, N 2, 89 SECTIO D 2003

Chair and Department of Dermatology, Medical University of Lublin

GRAŻYNA CHODOROWSKA, DOROTA CZELEJ,
MARTA NIEWIEDZIOŁ

Interleukin-2 and its soluble receptor in selected drug-induced cutaneous reactions

In the course of many drug-induced adverse cutaneous reactions the presense of T lymphocytes and macrophages in lesional skin as well as manifestations of their effector activities toward epidermal cells can be found out.

Interleukin-2 (IL-2) connected with the function of both these cell types, belongs to the most crucial cytokines in immune response. IL-2 is released by activated T cells, mostly Th1, but also, although to a lesser degree, by cytotoxic T lymphocytes (10). The key role of IL-2 is due to the fact that it is not only the growth factor for T cells but also the potent stimulator of their immunological function (7,9). IL-2 affects proliferation of T lymphocytes, their differentiation into cytotoxic cells, and it also takes part in thymocytes maturation in thymus (7). The prominent influence of IL-2 on the NK cells leads to their increased proliferation and promoting of the LAK activity (9). Moreover, other immune system cells can also be directly or indirectly stimulated by IL-2, among them B cells. Growth and differentiation of these cells is induced both through IL-4 and directly through IL-2 receptor present on the B lymphocyte surface (5). IL-2 is capable of enhancing the main biological function of B cells, which is releasing antibodies and autoantibodies (5,7). It means that IL-2, being the powerful mediator of cell-dependent immunity, may also affect the humoral type of immune response. It seems that in pathogenic events of cutaneous drug-induced reactions, activating influence of IL-2 on releasing of IFN-γ (by T lymphocytes) and TNF-α (by T cells, NK cells and monocytes/ macrophages) may be of special interest (5,7,9,12). These activities of IL-2 indicate that this cytokine can influence (directly and indirectly) the most important phenomena in skin inflammation such as: T cell activation, antigen presentation, recruitment of inflammatory cells to skin, macrophage cytotoxicity against other cells (13). What is more, IL-2 stimulating the IL-6 release by T lymphocytes and keratinocytes can indirectly affect the mobilization of the acute phase response (14).

IL-2 exerts its biological influence through the specific receptors present on the activated T and B cells, monocytes, NK cells and endothelial cells (12). During activation IL-2 receptor can be released from the cell surface by the proteolytic enzymes and it can constitute the soluble receptor (sIL-2R). Soluble IL-2R binds interleukin-2 with affinity similar to the membrane receptor (2,7,12). The physiologic function of sIL-2R is complex. This protein may be regarded as negative regulator of immune IL-2-related response through: 1) decreasing the density of membrane receptor and 2) blocking IL-2 by competition with membrane receptor (7,12). Thus, in its both roles, sIL-2R may act as a competitive inhibitor of IL-2. It is believed that sIL-2R is released proportionally to receptor expression on the surface of activated mononuclear cells (7,12). Due to these characteristics, sIL-2R is regarded as a sensitive and

early marker of immune cell activation, preceding lymphocyte proliferation and apearance of other determinants on the cell membrane (7,12). Elevated sIL-2R levels were observed in Hodgkin disease, T-cell leukemia, infections, rheumatoid arthritis, SLE, Sjogren syndrome, systemic sclerosis, AIDS, GVHD, atopic dermatitis, lichen planus, cutaneous lymphomas and psoriasis (2,3,5,6,7,8,10,12,13,15).

Measurement of sIL-2R level in peripheral blood may be thus useful in monitoring of diseases pathogenically connected with lymphocyte activation (3,7,10).

MATERIAL AND METHODS

Drug-induced cutaneous reactions were diagnosed in 126 patients included into the study. Among them were 61 women and 65 men, aged from 18 to 77 years, mean age 41.5 years. The control group consisted of 30 healthy volunteers at appropriate age.

All patients were subdivided into 6 following groups: 1) maculopapular eruptions (ME) – 40 patients, 2) drug-induced urticaria (DU) – 33 patients, 3) erythema multiforme (EM) – 24 patients, 4) erythema multiforme + erythema nodosum (EMN) – 6 patients, 5) hyperergic vasculitis (HV) – 14 patients, 6) Stevens-Johnson syndrome and toxic epidermal necrolysis (SJS/TEN) – 9 patients.

Blood samples were taken from all the patients: a) during the acute stage of disease, before the treatment was administered; b) after clearing of skin lesions following the effective treatment.

Measurement of IL-2 and soluble IL-2 receptor concentrations. An enzyme-linked immunosorbent assay (ELISA) was used to detect and quantify the presence of IL-2 and sIL-2R in plasma. The kits for ELISA were provided by Endogen Inc. USA. The measurements were done in duplicates according to the instructions included in the assays. The obtained data were put to statistical analysis. Average (M), median (Me), standard deviation (SD), the mean error of the average (SE) and variation coefficient (V%) were evaluated. Significance of differences between the average values was tested by the Student's t-test and Mann-Whitney's test.

RESULTS

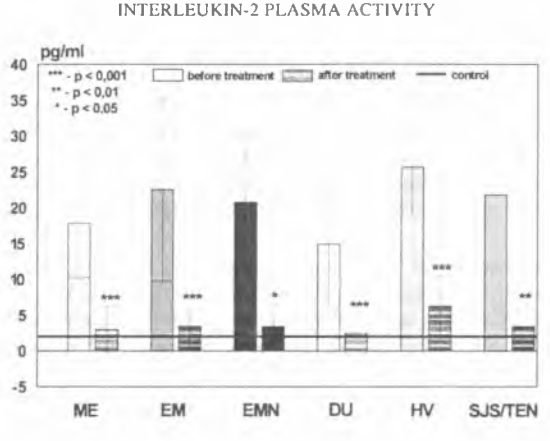

Fig. 1. Plasma concentrations of IL-2 in 126 patients with drug-induced skin reactions before and after treatment

Table. 1. Plasma concentrations of IL-2 (pg/ml) in 126 patients with drug-induced skin reactions

Patients	Group	Statistical characteristics						Comparison		
		n	M	SD	Min	Max	V%	with control		after vs before
								p	lg%	p
ME	C	30	2.08	1.95	0.00	8.00	93.90			
	P_1	40	17.85	7.61	3.60	39.10	43.08	< 0.001	2.93	< 0.001
	P_2	40	2.97	3.26	0.00	16.00	109.83	> 0.05	2.15	
EM	C	30	2.08	1.95	0.00	8.00	93.90			
	P_1	24	22.56	12.83	7.90	69.80	56.87	< 0.001	3.03	< 0.001
	P_2	24	3.40	3.03	0.00	12.20	97.17	> 0.05	2.21	
EMN	C	30	2.08	1.95	0.00	8.00	93.90			
	P_1	6	20.83	9.38	8.00	34.00	45.05	< 0.001	3.00	< 0.05
	P_2	6	3.38	3.37	0.00	7.30	99.49	> 0.05	2.21	
DU	C	30	2.08	1.95	0.00	8.00	93.90			
	P_1	33	14.93	8.62	0.00	35.80	57.76	< 0.001	2.85	< 0.001
	P_2	33	2.48	2.87	0.00	12.40	115.48	> 0.05	2.07	
HV	C	30	2.08	1.95	0.00	8.00	93.90			
	P_1	14	25.67	6.65	12.40	36.00	25.88	< 0.001	3,09	< 0.001
	P_2	14	6.26	4.21	0.80	16.00	67.14	< 0.001	2.47	
SJS/TEN	C	30	2.08	1.95	0.00	8.00	93.90			
	P_1	9	21.81	9.51	3.20	34.00	43.61	< 0.001	3.02	< 0.01
	P_2	9	3.41	3.33	0.00	10.00	97.85	> 0.05	2.21	
All patients	C	30	2.08	1.95	0.00	8.00	93.90			
	P_1	126	19.28	9.72	0.00	69.80	50.44	< 0.001	2.97	< 0.01
	P_2	126	3.34	3.41	0.00	16.00	102.10	> 0.05	2.20	

C – control, P1 – patients before treatment, P2 – patients after treatment

Table. 2. Plasma concentrations of sIL-2R (U/ml) in 126 patients with drug-induced skin reactions

Patients	Group	Statistical characteristics						Comparison		
		n	M	SD	Min	Max	V%	with control		after vs before
								p	lg%	p
ME	C	30	253.33	106.35	96.00	420.00	41.98			
	P_1	40	2289.88	1420.50	535.00	6446.00	62.03	< 0,001	2.95	< 0.001
	P_2	40	1030.58	784,97	184.00	340.4	76.17	< 0,001	2.60	
EM	C	30	253.33	106.35	96.00	420.00	41.98			
	P_1	24	1969.58	1012.77	431.00	3708.00	51.42	< 0.001	2.89	< 0.001
	P_2	24	793.04	493.44	150.00	2082.00	62.22	< 0.001	2.49	
EMN	C	30	253.33	106.35	96.00	420.00	41.98			
	P_1	6	2532.33	1103.92	1655.00	4160.00	43.59	< 0.001	2.99	< 0.05
	P_2	6	1012.00	608.59	415.00	1830.00	60.14	< 0.001	2.60	
DU	C	30	253.33	106.35	96.00	420.00	41.98			
	P_1	33	1839.61	1057.10	245.00	5520.00	57.46	< 0.001	2.86	< 0.001
	P_2	33	758.70	452.88	234.00	2402.00	59.69	< 0.001	2.47	
HV	C	30	253.33	106.35	96.00	420.00	41.98			
	P_1	14	2963.86	979.11	885.00	4426.00	33.04	< 0.001	3.07	< 0.001
	P_2	14	801.64	237.91	509.00	1297.00	29.68	< 0.001	2.50	
SJS/TEN	C	30	253.33	106.35	96.00	420.00	41.98			
	P_1	9	2323.78	1280.17	555.00	4020.00	55.09	< 0.001	2.96	< 0.01
	P_2	9	1268.78	763.89	555.00	2708.00	60.21	< 0.001	2.69	
All patients	C	30	253.33	106.35	96.00	420.00	41.98			
	P_1	126	2199.79	1215.12	245.00	6446.00	55.24	< 0.001	2.93	< 0.01
	P_2	126	904.82	609.90	150.00	3404.00	67.41	< 0.001	2.55	

C – control, P1 – pateints before treatment, P2 – patients after treatment

In the acute stage of drug-induced skin reactions highly significantly elevated mean levels of IL-2 ($p<0.001$) in comparison with normal values were found in all six groups of patients (Tab. 1). After clearing of clinical symtoms, IL-2 mean plasma levels, considered both in the whole group of 126 patients and in relation to the particular type of drug-induced reaction, were decreased to values not significantly different from the control (Tab. 1). Moreover, clinical recovery as a result of the treatment was connected with significant (EMN, SJS/TEN) or highly significant (ME, EM, DU, HV) decrease of IL-2 mean plasma levels in comparison to the values measured before treatment (Fig. 1).

SOLUBLE IL-2 RECEPTOR PLASMA LEVELS

In the examined group of 126 patients highly significant elevation of sIL-2R mean plasma concentrations in the acute stage of drug-induced skin reactions ($p<0.001$) in comparison with the control was observed (Tab. 2). After clearing of symptoms, mean plasma levels of sIL-2R, despite a decrease, were still highly significantly elevated in all examined groups of patients, when compared with the control. Comparison between before- and after treatment values showed, however, that clinical recovery was connected with highly significant (ME, EM, DU, HV) or significant (SJS/TEN, EMN) decrease of sIL-2R mean levels (Fig. 2).

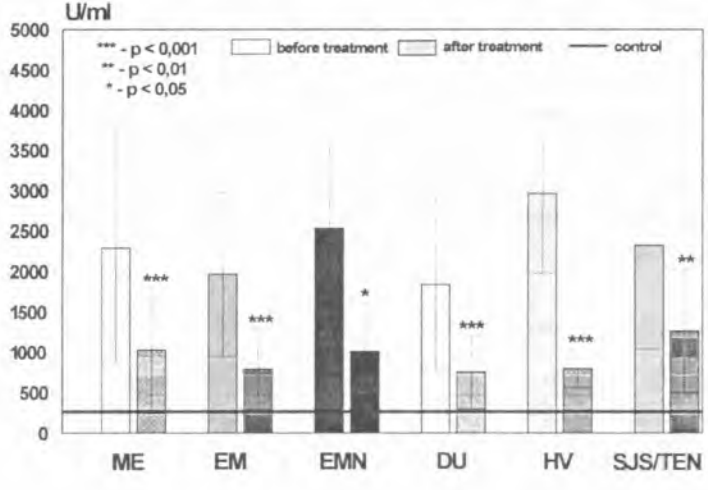

Fig. 2. Plasma concentrations of sIL-2R in 126 patients with drug-induced skin reactions before and after treatment

DISCUSSION

The results indicate that changes in the clinical course of drug-induced skin reactions can be followed by changes in plasma activity of interleukin-2 and its soluble receptor. In the healthy control subjects, the measurable amounts of both IL-2 and sIL-2R were found, which is in accordance with the literature data (1,4,7,11). It is believed that slight sIL-2R serum concentration in healthy adults expresses the basic level of immune activation induced by normal physiologic stimuli (2,3,12). Comparing differences of cytokine concentrations among

the six examined groups, the lowest mean plasma level, both before and after treatment, was observed in DU patients, and the highest in patients with HV. These results seem to reflect a lower influence of IL-2 on the immediate type reactions in urticaria, and a potentially powerful influence, through induction of adhesion molecules and proinflammatory cytokines, upon the inflammation of vessel walls.

It is worth to stress that plasma activity of sIL-2R was parallel to plasma activity of the cytokine in the acute clinical stage of the selected cutaneous drug-induced reactions. When recovery was achieved due to efficient treatment, the lowest sIL-2R mean level was found in DU but the highest in SJS/TEN patients. Finding the sIL-2R plasma concentrations persistent still highly significantly higher than in healthy subjects, despite the fact that IL-2 level returned to the control limits, is our the most striking result. It may suggest that sIL-2R is a more sensitive indicator of immune response and disturbed homeostasis, but on the other hand, it may also express mobilization of compensatory mechanisms extinguishing the cell-type immune reaction, because sIL-2R and IL-2 in some conditions act antagonistically towards each other (7).

The results support the belief that activated T lymphocytes take part in pathogenic events of cutaneous drug-induced diseases and that measurement of IL-2 and its receptor can be regarded as useful reliable parameters of the clinical course in these adverse reactions.

CONCLUSIONS

1. In the course of drug-induced skin reactions plasma activity of IL-2 and its soluble receptor is highly elevated, which indirectly suggests participation of activated T cells in the pathogenic events.

2. Persistent elevated sIL-2R plasma levels mean that immune activation is a more prolonged phenomenon than clinical symptoms of cutaneous drug-induced reactions.

REFERENCES

1. Alpsoy E. et al: The levels of plasma Interleukin-2 and sIL-2R in Behcet's disease: a marker of disease activity. J. Dermatol., 25, 513, 1998.
2. Bernengo M.G. et al.: Soluble IL-2 receptor in Sezary syndrome. Br. J. Dermatol., 128, 124, 1993.
3. Bruns M. et al: Serum levels of soluble IL-2 receptor, soluble ICAM-1, TNF-α , IL-4 and IL-6 in scleroderma. JEADV, 8, 222, 1997.
4. Chibowska M. et al: Internal organ lesions in acrosclerosis patients: IL-2 and soluble receptor for IL-2 plasma levels. JEADV, 14, 234, 2000.
5. Chodorowska G.: Badania zachowania się wybranych cytokin i białek ostrej fazy w łuszczycy. Praca habilitacyjna, AM w Lublinie, 1999.
6. Kemmet D. et al: Serum soluble IL-2 receptor in psoriasis. Acta Derm.Venereol (Stockh.), 70, 264, 1990.
7. Kozioł-Montewka M.: Ocena systemu cytokin, rozpuszczalnych receptorów i powierzchniowych cząsteczek sygnalizacyjnych w aspekcie zaburzeń immunologicznych u chorych z przewlekłą niewydolności nerek. Praca habilitacyjna, AM w Lublinie, 1997.
8. Lis A. et al: Serum level of soluble interleukin-2 receptor – a possible index of disease prognosis in systemic sclerosis. Clin.Exp.Dermatol., 22, 60, 1997.

9. Naume B., Espevik T.: Immunoregulatory effects of cytokines on natural killers. Scand.J.Immunol, 40, 128, 1994.
10. Ohashi Y. et al.: Serum levels of sIL-2R in patients with perennial allergic rhinitis before and after immunotherapy. Ann. Allergy Asthma Immunol., 77, 203, 1996.
11. Rubin L.: Soluble IL-2 receptor in rheumatic disease. Arth. Rheum., 33, 1145, 1990.
12. Rubin L. A., Nelson D.L.: The soluble IL-2 receptor: biology, function and clinical application. Ann. Int. Med., 113, 619-627, 1990.
13. Valsecchi R., et al: Soluble IL-2 receptor in lichen planus. Br. J. Dermatol., 125, 4, 403, 1991.
14. Wong R.L. et al: The mechanism of action of cyclosporin A in the treatment of psoriasis. Immunol.Today, 14, 69, 1993.
15. Yamamoto T. et al: Beneficial clinical effects of cyclosporin A on severe psoriasis. Arch.Dermatol.Res, 286, 57, 1996.

SUMMARY

Plasma concentrations of interleukin-2 (IL-2) and its soluble receptor (sIL-2R) were examined in 126 patients with drug-induced skin reactions: maculopapular eruptions (ME), erythema multiforme (EM), erythema multiforme coexisting with erythema nodosum (EMN), drug-induced urticaria (DU), hyperergic vasculitis (HV), Stevens-Johnsson syndrome and toxic epidermal necrolysis (SJS/TEN). The activity of both proteins were measured using immunoenzymatic ELISA method: a) in the acute stage of disease, before treatment was administered, and b) after clearing of skin symptoms, after treatment. In the acute stage of disease highly elevated mean concentrations of IL-2 and sIL-2R in all 6 groups of patients were found ($p<0.001$) in comparison with the control. After clearing of skin lesions IL-2 mean concentrations were lowered to the level not different significantly from the control ($p>0.05$), but sIL-2R mean plasma concentrations, despite the deep decrease, were still highly significantly elevated in comparison with control values ($p<0.001$).

Interleukina-2 i jej rozpuszczalny receptor w skórnych reakcjach polekowych

Badano stężenia interleukiny-2 (IL-2) i jej rozpuszczalnego receptora (sIL-2R) w osoczu 126 chorych z skórnymi reakcjami polekowymi: osutkami plamistogrudkowymi (ME), rumieniem wielopostaciowym (EM), rumieniem wielopostaciowym współistniejącym z rumieniem guzowatym (EMN), ostrą pokrzywka polekową (DU), hyperergicznym zapaleniem naczyń (HV) oraz zespołem Stevens-Johnsona i toksyczną nekrolizą naskórka (SJS/TEN). Aktywność badanych białek oznaczano metodą immunoenzymatyczną ELISA: a) w ostrym okresie choroby przed rozpoczęciem leczenia oraz b) po ustąpieniu zmian chorobowych i zakończeniu leczenia. Stwierdzono znaczne podwyższenie średnich stężeń IL-2 i jej receptora we wszystkich badanych grupach przed leczeniem ($p<0,001$) w porównaniu z grupą kontrolną. Po ustąpieniu zmian chorobowych średnie stężenia IL-2 obniżyły się do wartości nieróżniących się statystycznie od kontroli ($p>0,05$), natomiast średnie stężenia sIL-2R, pomimo znacznego obniżenia, pozostały nadal wysoce istotnie podwyższone w porównaniu z wartościami kontrolnymi ($p<0,001$).