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Activity of Tumor Necrosis Factor-alfa (TNF-α) and selected acute phase proteins in plasma of psoriatic patients receiving local treatment

Tumor Necrosis Factor-α (TNF-α) is a pleiotropic cytokine believed to play an essential role as mediator of inflammatory and immunological reactions. The pivotal role of TNF- α in numerous physiologic and pathologic phenomena is stressed by the fact that its high-affinity membrane receptors are expressed on all of cell types, except erythrocyte (8,11,12). TNF-α is a protein that can be released from almost all common resident skin cells, and also from infiltrating leukocytes (3,5,6,10,12). Among various acknowledged sources of this cytokine there are skin macrophages, activated T lymphocytes and keratinocytes which are known as active participants of pathogenetic events in psoriasis (3-5). TNF-α exerts many activities relevant to the pathogenesis of psoriasis. Due to its biological properties, this cytokine can strongly influence the recruitment of inflammatory cells into the skin, neutrophil and monocyte chemotaxis and is involved in the control of keratinocyte growth (3,6). It can also stimulate release of proinflammatory mediators from mast cells in skin (3,6). In synergy with IFN-γ, TNF-α can induce HLA-DR, ICAM-1 and VCAM-1 expression on the surface of keratinocytes and endothelial cells (3,4,5,11) and is capable of initiating apoptosis (6,11).TNF-α can activate a number of genes for enzymes and cytokines, potentiate granulocyte and macrophage phagocytosis and together with IL-1 and IL-6, can stimulate the systemic symptoms of inflammation, such as the acute phase response (2, 3, 5, 7, 12, 13). Acute phase reaction is of special interest in psoriasis because is induced and enhanced by proinflammatory cytokines taking part in skin inflammation.

OBJECTIVE

The aim of the present study was to study the plasma activity of Tumor Necrosis Factor-α (TNF-α), C-reactive protein (CRP) and α-2 macroglobulin (α-2 MG) in patients with medium-severe and severe psoriasis treated locally.

MATERIAL AND METHODS

100 male patients with medium-severe and severe plaque type psoriasis were included into the study. Mean age of the group was 37 years, range 18-60. They have suffered from psoriasis for 1 to 42 years, mean duration of disease was 16 years. The clinical activity of disease was evaluated by the same investigator according to the PASI score. It ranged from 18 do 64, mean PASI value was 25.8. Remission was achieved after 4 to 11 weeks of topical treatment with 0.125-2% dithranol ointment. Control group consisted of 30 healthy male volunteers in appropriate age. Blood samples were taken from all the patients: a) during the active phase of disease, before the treatment was administered; b) after clearing of psoriatic lesions, following the treatment.

Measurements of protein concentrations. An enzyme-linked immunosorbent assay (ELISA) was used to detect and quantify the presence of selected proteins in plasma. The kits for ELISA were provided by Endogen Inc. USA (TNF-α), Eucardio Laboratory Inc. USA (CRP); Immunodiagnostik GmbH, Germany (α-2 MG). The measurements were done in duplicates according to the instructions included in the assays. Microplate ELISA Reader Model 960 Metertech Inc. Austria was used for the assay. 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 averages was tested by the Student's t-test or Cochran's and Cox's test.

RESULTS AND DISCUSSION

In 100 patients mean plasma levels of TNF-α, CRP and α-2 MG examined in the active stage of psoriasis were highly significantly elevated ($p<0.001$) when compared with healthy control (Tab.1, Fig.1). In remission the plasma concentrations of these proteins lowered towards the control values and both TNF-α and α-2 MG mean levels did not differ from control values ($p>0.90$ and $p>0.15$; respectively). The CRP mean level, however, despite its deep decrease following local treatment was still significantly higher in comparison with the control (Fig.1).

Concerning the changes in plasma activities of the measured proteins due to treatment administered, the influence of the local treatment with dithranol ointment on the decrease in protein values after treatment was found highly significant.

Induced by TNF-α C-reactive protein is the most sensitive indicator of inflammation, whose concentration can increase 100 and more times even in first twenty-four hours and can lower to almost normal level during several days after effective treatment or decrease spontaneously with clearing of the disease (9). In the examined group of psoriatic patients the 19-fold increase in plasma in comparison with the control values was found, but the complete reduction to normal values was not observed. In remission the CRP mean concentration in plasma was still 2.5 times elevated above the control values. Also α-2 MG proved to be a useful indicator of disease activity, decreasing to the control values when the clinical remission was achieved. α-2 MG apart from being a broad-spectrum proteinase inhibitor, plays the role of carrier protein for many mediators of immune response, including cytokines and growth factors (1) and due to these features has also immunomodulating properties (1). Observed changes in activity of TNF-α, C-reactive protein and α-2 macroglobulin from the active stage to remission confirm the belief that in the course of severe psoriasis the acute phase response can be initiated, and what is more, that despite the lack of clinical symptoms of psoriasis, there are persistent inflammatory process and some symptoms of immune preactivation anticipating future relapse of psoriasis can be found.

Parameter	Group	Statistical characteristics							Comparison with control	
		Min	Max	M	Me	SD	SE	V%	p	lg%
	P	18	60	37.58	37	11.79	1.18	31.4	> 0.80	-
Age	C	18	57	37.07	38	11.45	2.09	30.9		
PASI	P	18	64	25.87	24.3	7.04	0.70	27.2	-	-
Duration of psoriasis	P	1	42	15.92	13.5	10.65	1.07	66.9	-	-
TNF-α (pg/mL)	P_1	16	68	31.98	32	9.60	0.96	30.0	< 0.001	2.388
	P_2	2	30	13.14	12	6.14	0.61	46.7	> 0.90	2.001
	C	0	28	13.1	14	9.3	1.70	70.9		
CRP (mg/L)	P_1	0	26,34	5.54	4.27	5.20	0.52	93.7	< 0.001	3.281
	P_2	0	2,61	0.74	0.76	0.53	0.05	71.3	< 0/001	2.407
	C	0	0,86	0.29	0.30	0.30	0.05	102.5		
α-2MG (mg%)	P_1	126	1000	307.8	263	167.9	16.79	54.5	< 0.001	2.376
	P_2	30	196	127.1	128	34.5	3.45	27.2	> 0.70	1.992
	C	30	190	129.5	133	42.9	7.83	33.1		

P – patients, P_1 – patients before treatment, P_2 – patients after treatment, C – control.

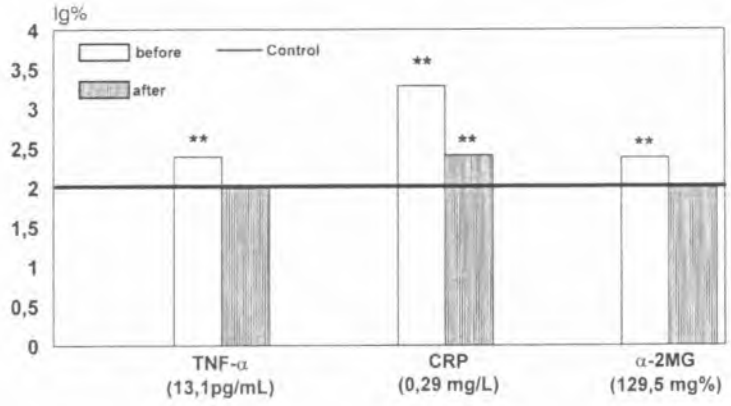

Fig. 1. Plasma concentrations of TNF-α, CRP and α–2 MG before and affer the local treatment expressed as Lg % of control

CONCLUSIONS

1. In the examined group of psoriatic patients highly elevated expression of TNF-α, CRP and α-2 MG in plasma of 100 psoriatic patients was found in the active stage of disease. Remission achieved as a result of the local treatment was connected with considerable decrease in the examined proteins' level in plasma.

2. Acute phase response initiated in psoriasis is not extinguished completely

with the achievement of remission, elevated concentrations of CRP still remain in the peripheral blood.

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SUMMARY

Plasma concentrations of Tumor Necrosis Factor-α (TNF-α), C-reactive protein (CRP) and α-2 macroglobulin (α-2 MG) were examined in 100 patients with medium-severe and severe psoriasis. Activity of selected proteins were measured using the ELISA method in the active stage of psoriasis and in remission achieved due to the local treatment with 0.125-2% dithranol treatment. In the active stage of disease highly increased plasma levels of TNF-α, CRP and α-2 MG were found ($p<0.001$) and remission was connected with a considerable decrease of the examined proteins towards the control values. But despite its lowering, CRP mean plasma level remained in remission significantly

elevated in comparison with healthy control. Results of this study indicate that in severe psoriasis the acute phase response can be initiated and is not completely extinguished with achievement of clinical remission.

Stężenia osoczowe Czynnika Martwicy Nowotworów-alfa (TNF-α) i wybranych białek ostrej fazy u chorych na łuszczycę leczonych miejscowo

Badano stężenia osoczowe Czynnika Martwicy Nowotworów-α (TNF-α), białka C-reaktywnego (CRP) i α-2 makroglobuliny (α-2 MG) u 100 chorych z łuszczycą średniociężką i ciężką. Stężenia wybranych białek mierzono w osoczu metodą immunoenzymatyczną ELISA w okresie objawowym łuszczycy oraz po skutecznym leczeniu miejscowym maścią cygnolinową 0,125-2%. Stwierdzono wysoce istotne ($p<0,001$) podwyższenie stężenia TMF-α, CRP i α-2 MG w aktywnym okresie choroby oraz obniżenie poziomu badanych białek w kierunku wartości kontrolnych wraz z uzyskaniem remisji pod wpływem leczenia. Jednak pomimo znacznego obniżenia stężenia osoczowe CRP pozostały w stanie klinicznej remisji nadal znacznie podwyższone w porównaniu z kontrolą. Uzyskane wyniki wskazują na to, że uruchomiona w łuszczycy odpowiedź ostrej fazy nie wygasa całkowicie wraz z osiągnięciem remisji zmian skórnych.