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LETTER TO THE EDITOR

TNF-238A is associated with juvenile onset psoriasis in patients of Han population in Southwest China

Psoriasis vulgaris is a recalcitrant inflammatory and hyperproliferative dermatosis, and has an exceedingly complex genetic basis. It can be divided into type I (onset before the age of 40 years old) and type II (onset beyond 40 years old). It has been reported that TNF-238A, one of the two common polymorphisms (another is TNF-308A) in promoter of TNF-α gene, is associated with a higher risk of developing type I psoriasis in Caucasian [1,2]. However, such association is rejected by subsequent researches in both Caucasian [3] and Japanese populations [4,5]. More data from other populations are necessary to test this association.

In this study, we used a case-control design to investigate whether these polymorphisms in promoter of TNF-α gene, especially at the positions -238 and -308, influence the susceptibility to PsV in Chinese population.

Seventy-seven patients with characterized PsV and eighty-two healthy subjects as controls were involved in our study (see Table 1), all of which belonged to Han nationality in Southwest China. All patients did not suffer from any other diseases and were divided into two subgroups: types I and II according to the onset age.

Genomic DNA was extracted using phenol—chloroform method from whole peripheral blood leukocytes. PCR was performed in 50 μl reaction mixture, consisting of genomic DNA (250 ng) 2 μl, 5 μl 10$\times$ PCR buffer (20 mM Tris—HCl pH 8.3, 500 mM KCl, 15 mM $MgCl_2$), 2 μl dNTP mixture (2.5 mM of each deoxynucleotide), 2 μl sense primer (5′-AACACAGGCCTCAGGACTCAACAC-3′) and anti-sense primers (5′-CTCTCCCTCTTAGCTGGTCCTCTG-3′, 10 pmol/μl each), 0.25 μl Taq DNA polymerase (5U/μl, TaKaRa, China), and 36 μl pure H_2O, to acquire a 552-bp fragment spanning position from -499 to +53 of the TNF-α gene promoter. Reaction conditions executed in the thermal cycler (Eppendorf, German) were as follows: 95°C for 4 min, 30 cycles consisting of 94°C for 1 min, 66°C for 1 min, and 72°C for 1 min, and finally 72°C for 10 min. The PCR products were separated by electrophoresis in 1.5% agarose gel and then sequenced directly using BigDye™ Terminator Cycle Sequencing kit and ABI PRISM 3700 sequencer (Applied Biosystems Inc., USA).

The frequencies of TNF-238A and TNF-308A were compared separately between either the type I or II group and the control group. Statistics analysis was performed by SPSS software (SPSS Inc.). Relative risk associated with a particular genotype was estimated by the odds ratio (OR) [6]. OR was tested using a Chi-Square distribution and the null hypothesis being tested is OR = 1. The level of significance was detected by Pearson Chi-Square test with Yates correction for small numbers. Probability (P) value <0.05 was regarded as significant.

The distribution of TNF-α gene promoter polymorphisms is shown in Table 2. No other polymorphisms were observed in this region besides TNF-238A and TNF-308A. A significantly higher genotype frequency of the TNF-238A was observed in patients (17% versus 4%, $P = 0.006$, OR = 5.35 [95% CI 1.5, 19.6]), particularly in type I (25% versus 4%, $P = 0.003$, OR = 8.17 [95% CI 1.7, 38.8]). The observed difference was further underlined by the presence of three TNF-238.2 homozygote persons in the type I PsV and the absence in the controls and the type II PsV. Our results strongly suggested an association between TNF-238A and type I PsV and thereby, support Höhler et al's viewpoint [2].

The frequency of TNF-308A genotype was lower in type I psoriasis (4.2% versus 17.6%, $P = 0.033$). Further studies with more samples are necessary to verify if there is a negative association between TNF-308A and type I psoriasis since the difference is marginal.

The frequency of the TNF-238A allele in type I patients in this study (12 of 48, 25%) was evidently lower than that of Arias et al. (29 of 64, 45%) [1] and Höhler et al. (23 of 60, 38%) [2] in Caucasian patients. In addition, such frequency in control group in our study (3 of 82, 4%) was also lower than that in Caucasian (7 of 99, 7%) [2], which revealed the similar trend as in type I patients. Such disaccord in genotype frequency may reflect the difference on genetic background between Caucasian and Mongolian.

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Table 1 Detailed list of patients and controls[a]

	Controls						Psoriasis vulgaris					
	<40 years old		≥40 years old		Total		Type I		Type II		Total	
	n[b]	%	*n*	%	*n*	%	*n*	%	*n*	%	*n*	%
Male	29	35.4	20	24.4	49	59.8	26	33.8	17	22.0	43	55.8
Female	22	26.8	11	13.4	33	40.2	22	15.6	12	15.6	34	44.2
Total	51	62.2	31	37.8	82	100	48	62.4	29	37.6	77	100

[a] The mean age of controls and patients were 37.2 and 39.3, respectively, and the average age of onset of patients was 38.4 years old.
[b] Number of investigated subjects.

Table 2 TNF-α promoter genotypes frequencies in the investigated groups

Locus/genotype	Controls (*n* = 82)						PsV (*n* = 77)					
	<40 years old		≥40 years old		Total		Type I		Type II		Total	
	n	%	*n*	%	*n*	%	*n*	%	*n*	%	*n*	%
TNF-α-238												
G/G	49	96	30	97	79	96	36[a]	75	28	97	64[b]	83
G/A	2	4	1	3	3	4	9	19	1	3	10	13
A/A	0	0	0	0	0	0	3	6	0	0	3	4
TNF-α-308												
G/G	42	82	29	94	71	87	46[c]	96	27	93	73[d]	95
G/A	9	18	2	6	11	13	2	4	2	7	4	5
A/A	0	0	0	0	0	0	0	0	0	0	0	0

[a] $P = 0.003$, G/A and A/A vs. G/G in type I PsV patients compared with controls whose age were before 40 years.
[b] $P = 0.006$, G/A and A/A vs. G/G in PsV patients compared with controls.
[c] $P = 0.033$, G/A and A/A vs. G/G in type I PsV patients compared with controls whose age before 40 years.
[d] $P = 0.076$, G/A and A/A vs. G/G in PsV patients compared with controls.

Our findings, however, were contrary to that in Japanese. The frequencies of TNF-238A and TNF-308A in Japanese population were reported as 2.3 and 2.9%, 2.0 and 1.7%, 4.2 and 2.1% in previous reports [4,5], while the frequencies in our study were 4 and 13%. One possible explanation is that such difference may reflect the genetic differentiation between South and North East Asia populations, which has been observed with other genetic markers. Our results suggest the importance of sampling from the same population in the case-control studies.

We reported the association between TNF-238A and type I psoriasis in Mongolian for the first time. In addition, polymorphisms in the promoter region of the TNFA may influence the level of TNF-α secretion [7], and the increased amount of TNF-α has been reported in both psoriatic skin [8] and synovial fluid [9]. Some recent trials have shown a clinical benefit by anti-TNF treatment in severe psoriasis [10]. All these indicated that studying the polymorphisms in the promoter region of TNF-α has great prospect of application.

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References

[1] Arias AI, Giles B, Eiermann TH, Sterry W, Pandcy JP. Tumor necrosis factor—alpha gene polymorphism in psoriasis. Exp Clin Immunogenet 1997;14:118—22.
[2] Höhler T, Kruger A, Schneider PM, et al. A TNF-α promoter polymorphism is associated with juvenile onset psoriasis and psoriatic arthritis. J Invest Dermatol 1997;109:562—5.
[3] Jacob N, Franz R, Marcus S-E, et al. Promoter polymorphism at −238 of the tumor necrosis factor-a gene is not associated with early onset psoriasis when tested by the transmission disequilibrium test. J Invest Dermatol 1999;112:514—5.
[4] Nishibu A, Oyama N, Nakamura K, Kaneko F. Lack of association of TNF-238A and TNF-308A in Japanese

patients with psoriasis vulgaris, psoriatic arthritis and generalized pustular psoriasis. J Dermatol Sci 2002;29(3): 181—4.
[5] Tsunemi Y, Nishibu A, Saeki H, Oyama N, Nakamura K, Kishimoto M, et al. Lack of association between the promoter polymorphisms at positions -308 and -238 of the tumor necrosis factor alpha gene and psoriasis vulgaris in Japanese patients. Dermatology 2003;207(4): 371—374.
[6] Dyer P, Warrens A. Design and interpretation of studies of the major histocompatibility complex in disease. In: Lechler R, editor. HLA and disease. London: Academic Press; 1994. p. 93—121.
[7] Kaluza W, Reuss E, Grossmann S, et al. Different transcriptional activity and in vitro TNF-α production in psoriasis patients carrying the TNF-α 238A promoter polymorphism. J Invest Dermatol 2000;114(6):1180—3.
[8] Ettehadi P, Greaves MW, Wallach D, Aderka D, Camp RDR. Elevated tumor necrosis factor-α (TNF-α) biological activity in psoriatic skin lesions. Clin Exp Immunol 1994;96: 146—51.
[9] Partsch G, Steiner G, Leeb BF, Dunky A, Broll H, Smolen JS. Highly increased levels of tumor necrosis factor-α and other proinflammatory cytokines in psoriatic arthritis fluid. Br J Rheumatol 1997;24:518—23.
[10] Oh CJ, Das KM, Gottlieb AB. Treatment with anti-tumor necrosis factor alpha (TNF-α) monoclonal antibody dramatically decreases the clinical activity of psoriasis lesions. J Am Acad Dermatol 2000;42(5):829—30.

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