

Association of IL-10 level and IL-10 promoter SNPs with specific antibodies in penicillin-allergic patients

Hai-Ling Qiao · Qiang Wen · Na Gao · Xin Tian ·
Lin-Jing Jia

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Abstract

Objective Our aim was to investigate the hypothesis that the sera interleukin-10 (IL-10) level and polymorphic nucleotides within the IL-10 gene promoters would link to specific IgE and IgG production and the expression of penicillin allergy.

Methods One hundred and two patients and 86 healthy subjects were chosen for assay of serum IL-10 level by enzyme-linked immunosorbent assay (ELISA) and type -1082 G/A and -819 C/T alleles by sequence-specific primer polymerase chain reaction (SSP-PCR). Radioallergosorbent test (RAST) and ELISA were used to examine eight types of specific immunoglobulin-E (IgE) and IgG antibodies, respectively, which included four types of antibodies to major and minor antigenic determinants.

Results Compared with control subjects and patients with negative-specific IgE, there were significantly lower levels of IL-10 in patients with positive-specific IgE ($P<0.05$). Similarly, there were significantly lower levels of IL-10 in patients with positive-specific IgG compared with normal controls and allergic patients with negative-specific IgG ($P<0.05$). The visible negative correlations existed between IL-10 and four types of specific IgE [benzylpenicilloyl (BPO), phenoxomethylpenicilloyl (PVO), benzylpenicillanyl (BPA), amoxicillanyl (AXA)], and patients with three or more positive-specific IgE had significantly lower IL-10 levels than normal controls ($P<0.01$). There was a declining trend for IL-10 level in serum with the increase in types of positive-specific IgE. But there was no significant difference in serum IL-10 level between the

positive skin-test group and the allergic-history group. Compared with controls and patients with negative antibodies, a significantly decreased frequency of the -1082 G allele was present in patients with positive antibodies ($P<0.01$). The allele T and TT genotype at -819 C/T position had lower frequency in the negative-specific IgG group than that in the positive group and controls ($P<0.01$).

Conclusions Positive specific IgE and IgG are associated with decreased IL-10 level in allergic reaction to penicillins. The distributions of genotype and frequency of allele at the -1082 G/A position may be associated with the production of both specific IgE and IgG antibodies.

Keywords Penicillin · Allergy · IgE · IgG · IL-10 · Genetic polymorphisms

Introduction

Penicillin antibiotics are widely used in many clinical situations, but allergic reactions to penicillin antibiotics are common, and the symptoms vary from mild skin rashes to, at worst, anaphylactic shock and even death. The mechanisms involved are not yet completely understood. However, clinical and immunologic studies suggest that immunoglobulin-E (IgE)-mediated and cell-mediated pathogenic mechanisms are involved in most immediate and nonimmediate reactions, respectively. However, despite a large number of studies on the production of specific IgE, IgG and the Th1/Th2 balance during immune response to protein antigens, little is known concerning the early events, which regulate secretion of specific antibodies and direction of Th1/Th2 differentiation [1, 2].

Interleukin-10 (IL-10), an essential Th2 cytokine, is reported to play a crucial role in allergic diseases such as

H.-L. Qiao (✉) · Q. Wen · N. Gao · X. Tian · L.-J. Jia
Department of Clinical Pharmacology, School of Medicine,
Zhengzhou University,
Zhengzhou 450052, People's Republic of China
e-mail: qiaohl@zzu.edu.cn

asthma by down-regulating cytokine production of Th1 and Th2 [3]. IL-10 also inhibits accessory cell function, and in the presence of IL-10, not only do T lymphocytes not proliferate and produce cytokines in response to allergen, those cells are rendered irreversibly nonresponsive (tolerant). The effects of IL-10 on survival, proliferation, and differentiation of human B cells are much more evident [4]. Support for a modulating role for IL-10 in human allergic diseases is further derived from observations that IL-10 inhibits eosinophil survival and Ig synthesis [5]. The inhibitory effects of IL-10 are in contrast to its effect on B-lymphocytes, where it stimulates proliferation and Ig secretion [6]. Furthermore, in its direct effect on T cells, IL-10 shows the same dual role as an active factor or a suppressor [7–9]. These results suggest that the role of IL-10 in the pathogenesis of allergic diseases is sophisticated and still inconclusive.

As knowledge about IL-10 progresses, its anti-inflammation effect in allergic diseases has been accepted and interest has developed in the gene on which IL-10 is loaded [10–13]. Variability in IL-10 production has a hereditary component of approximately 75% [14]. Human IL-10 is encoded by five exons on chromosome 1(q31–q32). The open reading frames encode secreted proteins of 178 amino acids, their sequence being consistent with an α -helical bundle structure similar to that of interferon (IFN) [15]. This variation is mainly due to polymorphisms in the promoter region of the IL-10 gene. Some single nucleotide polymorphisms (SNPs) and CA-repeat microsatellites have been described in the IL-10 promoter. Three of them, at positions –1082 G/A, –819 T/C, and –592 C/A, are reported to have correlation with IL-10 production. The –819 T/C polymorphism is in complete linkage disequilibrium with the –592 C/A polymorphism, –819 C is inherited together with –592 C, and –819 T is inherited together with –592 A [16, 17]. The functional significance of the three SNPs on serum IL-10 levels has been investigated, even though recent contrasting result has been reported on haplotype [17]. Due to the importance of IL-10 in allergic diseases, we conducted an association study between the level of serum IL-10, SNPs on its promoter, and specific antibodies in patients with penicillin antibiotic allergy.

Materials and methods

Subjects

The study involved 102 penicillin-allergic patients (47 with clinical penicillin-allergic history and the remainder with positive skin test) and 86 controls (Table 1). Allergic patients were mainly recruited from the first affiliated hospital of Zhengzhou University. There were 92 patients

Table 1 Clinical data of 102 patients with penicillin allergy and 86 healthy volunteers

Group	Control	Patients
Age ($\bar{x} \pm s$)	20.91 $\pm$ 5.78	21.32 $\pm$ 8.42
<18	8	10
18–59	76	89
≥ 60	2	3
Gender		
Male	46	68
Female	40	34
All patients		
Allergic history		47
Positive skin test		55
Total	86	102

allergic to penicillin G (PG), six allergic to amoxicillin (AX), two allergic to ampicillin (AMP), and two allergic to other β -lactam antibiotics. Skin testing was performed with PG on the inner side of the forearm by intradermal testing. PG sodium salt was diluted in saline solution until reaching a concentration of 200 U/ml as reagent. Approximately 0.1 ml (20 U) of reagent was injected for intradermal testing. Wheal diameter was measured after 20 min, and the reaction was considered positive when the diameter was larger than 5 mm with a surrounding area of erythema. All 86 controls had no history of allergic diseases, urticaria, or other atopic diseases. Additionally, fetal cord sera used as negative controls in the radioallergosorbent test (RAST) had been collected from the third affiliated hospital of Zhengzhou University.

The purpose of the protocol was explained clearly to the patients before inclusion in the study, and their written informed consent was obtained. The local independent ethics committees approved the study.

RAST for detection specific IgE

Major and minor antigenic determinant paper disks were made, including major antigenic determinants: benzylpenicilloyl-polylysine (BPO-PLL), ampicilloyl-polylysine (APO-PLL), amoxicilloyl-polylysine (AXO-PLL), and phenoxomethylpenicilloyl-polylysine (PVO-PLL), as well as minor antigenic determinants benzylpenicillanyl-polylysine (BPA-PLL), 6-aminopenicillanic-polylysine (APA-PLL), amoxicillanyl-polylysine (AXA-PLL), and phenoxomethylpenicillanyl-polylysine (PVA-PLL). Cyanogen-bromide-activated cellulose disks (filter paper 542, Whatman International, Maidstone, UK) were coupled to PLL and then treated with PG, phenoxymethylpenicillin (PV), AMP, and AX, as described. The tests were performed as previously reported [18]. RAST results reflected the radio-labeled substance uptake, expressed as a percentage of the

maximum counts per minute (cpm) added. Values over the mean+2.33 standard deviation (SD) of the results in the control group were considered positive against the corresponding allergen. Meanwhile, the fetal cord sera were used as negative control.

ELISA for detection specific IgG

Major and minor antigenic determinants included BPO, BPA, APO, APA, AXO, AXA, PVO, and PVA. Penicilloylation of human serum albumin (HSA) was carried out following Zhao et al.'s method [19]. The ELISA was performed according to published procedure [20], with modification. Next, 100 µg/ml antigen coated on microtiter plates with 100 µl/well were stored at 4°C overnight. Plates were washed three times with phosphate-buffer saline (PBS) 0.1 mol·L⁻¹ containing 0.05% (vol/vol) Tween-20. Prediluted sera (1:20) 100 µl were added in triplicate then incubated at 37°C for 2 h. Plates were washed three times with PBS-T, and 100 µl horseradish peroxidase-labeled sheep anti-human IgG (1:2,000) were added and incubated at 37°C for 2 h. After washing, 100 µl substrata solution containing O-phenylenediamine 400 mg·L⁻¹ and 0.15% H₂O₂ (30% wt/vol) in citrate phosphate buffer (0.1 mol·L⁻¹ pH 5.0) were added and incubated at 37°C for 0.5 h. Enzyme-substrate reaction was stopped by addition of 25 µl of H₂SO₄ (4 mol·L⁻¹). Absorbance (A) was read at 490 nm using enzyme immunoassay (EIA) reading (Bio-Rad, USA). Values over the mean+2.33 SD of the results in the control group were considered positive against the corresponding allergen.

ELISA for detection IL-10

Blood samples were drawn from each subject. After clotting for 2 h at room temperature, the sera were separated and stored at -30°C until in vitro tests were performed. Concentrations of IL-10 in sera were determined using a human IL-10-specific ELISA kit from Jingmei Biotech Co. Ltd., which had a lower limit of detection of 30 pg/ml.

Genotyping of the IL-10 polymorphisms

Genomic DNA was extracted from whole-blood samples using a standard proteinase-K digestion and phenol/chloroform extraction. To determine the correct haplotypes deriving from complete linkage disequilibrium, we analyzed two of the three polymorphisms of the IL-10 promoter region, -1082 G/A and -819 C/T, using sequence-specific primer polymerase chain reaction (SSP-PCR) restriction. Genotyping of these polymorphisms was

performed following the methods previously described [21]. Primer sequences and conditions were as follows:

PCR product size = 258 bp:

- IL-10 (-1082) generic primer (antisense): 5'-cagtgc caactgagaatttg-3'
- IL-10 (-1082) primer G (sense): 5'-ctactaaggcttcttgg gag-3'
- IL-10 (-1082) primer A (sense): 5'-actactaaggcttcttgg gaa-3'

PCR product size = 233 bp:

- IL-10 (-819)* generic primer (antisense): 5'-aggatgtgttccaggctcct-3'
- IL-10 (-819)* primer C (sense): 5'-ccctgtacaggtgatg taac-3'
- IL-10 (-819)* primer T (sense): 5'-accctgtacaggtgatg taat-3'

(*IL-10 -592 polymorphism is in linkage disequilibrium with -819 polymorphism: allele C at -592 is always present when at position -819 is allele C and allele A at -592 is always present when at position -819 is allele T).

DNA was amplified in a 20-µl reaction. Final reagent concentrations were: 1× reaction buffer, 250 nmol/l primers, 1 µmol/l dNTPs, 1.5 mmol/l MgCl₂, and 0.25 U TaqDNA polymerase. The program was predenaturation 95°C for 3 min, denaturation 95°C for 45 s, annealing 59°C for 45 s, and polymerization 72°C for 1 min for 30 cycles, and polymerization 72°C for 5 min. The amplified products could be monitored by electrophoresis on a 2% agarose gel containing ethidium bromide (0.5 mg/ml).

Statistical analysis

Comparisons for statistical significance were performed using the Mann-Whitney *U* test (two-tailed *P* value) and Kruskal-Wallis *H* test. Correlation coefficients for statistical significance were determined using Spearman's test. For genetic comparisons, differences in allele, genotype, and haplotype frequencies were evaluated using the chi-square (χ^2) test. The Statistical Package for the Social Sciences (SPSS10.0) was used for all analyses, and statistical significance was defined as *P*<0.05.

Results

Clinical and functional data

One hundred and two patients and 86 healthy subjects were chosen for the study aimed at determining the serum IL-10 level and examining -1082 and -819 alleles in the IL-10

promoter region by PCR-SSP. Of the 102 patients, 55 had positive skin test, and 47 had allergic history, including 14 with anaphylactic shock, 25 with urticaria, and eight with other symptoms. Among them, 52 (50.98%) patients were positive to IgE and 63 (61.67%) patients were positive to positive-specific IgG.

The serum levels of IL-10

There were no significant differences in levels of IL-10 between penicillin-allergic patients and control subjects (Table 2). However, compared with control subjects, there were significantly lower levels of IL-10 in patients with positive-specific IgE ($P<0.01$), and patients negative to IgE had significantly higher levels of IL-10 than patients positive to IgE ($P<0.05$). Similarly, there were significantly lower IL-10 levels in allergic patients with positive-specific IgG antibodies compared with normal controls ($P<0.05$), and a same result was observed in allergic patients with positive-specific IgG antibodies compared with allergic patients with negative-specific IgG ($P<0.05$).

Correlation between IL-10 and the specific antibody

Among patients, there were significant correlations between IL-10 and specific IgE. As shown in Table 3, visible inverse correlations existed between IL-10 and four types of

Table 3 Correlation between interleukin-10 (IL-10) and specific antibodies in serum of allergic patients

		IL-10 vs. IgE	IL-10 vs. IgG
BPO	Correlation coefficient	−0.210*	−0.093
	Significance (2 tailed)	0.027	0.351
	<i>n</i>	102	102
PVO	Correlation coefficient	−0.210*	0.087
	Significance (2 tailed)	0.034	0.383
	<i>n</i>	102	102
APO	Correlation coefficient	−0.160	−0.073
	Significance (2 tailed)	0.107	0.463
	<i>n</i>	102	102
AXO	Correlation coefficient	−0.127	−0.064
	Significance (2 tailed)	0.202	0.521
	<i>n</i>	102	102
BPA	Correlation coefficient	−0.205*	−0.004
	Significance (2 tailed)	0.038	0.967
	<i>n</i>	102	102
PVA	Correlation coefficient	−0.132	−0.070
	Significance (2 tailed)	0.186	0.487
	<i>n</i>	102	102
APA	Correlation coefficient	−0.134	0.097
	Significance (2 tailed)	0.179	0.332
	<i>n</i>	102	102
AXA	Correlation coefficient	−0.252*	−0.099
	Significance (2 tailed)	0.011	0.322
	<i>n</i>	102	102

IgE immunoglobulin-E, BPO, PVO, AXO, APO specific antibody (IgE or IgG) produced by stimulating major antigenic determinants of benzylpenicilloyl-polylysine (BPO-PLL), ampicilloyl-polylysine (APO-PLL), amoxicilloyl-polylysine (AXO-PLL), and phenoxomethylpenicilloyl-polylysine (PVO-PLL), BPA, PVA, AXA, APA specific antibody (IgE or IgG) produced by stimulating minor antigenic determinants of benzylpenicillanyl-polylysine (BPA-PLL), phenoxomethylpenicillanyl-polylysine (PVA-PLL), 6-aminopenicillanic-polylysine (APA-PLL), and amoxicillanyl-polylysine (AXA-PLL).

* $P<0.05$ inverse correlations existed between IL-10 and four types of specific IgE

specific IgE (BPO, PVO, BPA, AXA). But compared with IgG, we could not obtain the same result (Table 3).

IL-10 level and types of positive-specific IgG and IgE antibodies in sera of subjects

According to the different number of positive-specific IgE, allergic patients with positive-specific IgE were divided into groups with 1, 2, 3, or more positive-specific IgE. We observed (Table 4) a declining trend with the increasing number of positive-specific IgE types, but only patients with three or more positive-specific IgE had significantly lower IL-10 levels than normal controls ($P<0.01$). However, we could not observe significant differences in every group divided according to types of positive-specific IgG antibodies (Table 4).

Table 2 Interleukin-10 (IL-10) level in patient serum

Group	n	IL-10 level (ng·L ^{−1})		
		Median	Minimum	Maximum
Control	86	106.0	22.7	363.5
Patients				
According to specific immunoglobulin (Ig) E				
Positive IgE	52	75.7** ^a	22.7	189.3
Negative IgE	50	106.5	45.4	263.5
According to specific IgG				
Positive IgG	63	98.4*** ^a	22.7	181.7
Negative IgG	39	106.9	45.4	263.5
According to symptom ^b				
Positive skin test	55	98.4	30.3	189.3
Allergic history	47	98.6	22.7	263.5
Anaphylactic shock	14	106.9	37.9	227.2
Urticaria	25	98.6	22.7	263.5
Other symptoms	8	90.85	45.4	151.4
Total	102	98.6	22.7	263.5

^a $P<0.05$ vs. negative IgE

^b Forty-seven of 102 subjects had clinical penicillin-allergic history, while the others only had positive reactions in skin test.

* $P<0.01$ vs. control

** $P<0.05$ vs. control

Table 4 Interleukin-10 (IL-10) level in serum of allergic patients with one, two, or three or more positive-specific antibodies

Group		IL-10 level	
		<i>n</i>	Median (ng·L ⁻¹)
Control		86	106.0
IgE	A	16	106.0
	B	17	75.7
	C	19	75.7*
IgG	A	47	98.4
	B	12	98.4
	C	4	90.8

Ig immunoglobulin, A allergic patients with only one positive-specific antibody, B allergic patients with two positive-specific antibodies, C allergic patients with three or more positive-specific antibodies

* $P<0.01$ vs. control

Distribution of IL-10 -1082 G/A and -819 C/T polymorphisms

Distribution of IL-10 -1082 G/A and -819 C/T polymorphisms among patients and control subjects are shown in Table 5. Allele G at the -1082 position in the IL-10 promoter gene had significantly lower frequency in patients with positive-specific antibodies, whether IgE or IgG, than in controls ($P<0.01$) and negative patients ($P<0.01$), and

the same result were observed in examination of the GG genotype. Except for a few patients, we did not find the TT genotype in patients with negative-specific IgG, so the results show that the allele T and TT genotype at the -819 C/T position had lower frequency in the negative-specific IgG group than that in the positive group and controls ($P<0.01$).

Discussion

This study mainly showed the relationship between serum-specific antibodies and IL-10 in patients with penicillin allergy. The study involving IgE, IgG, and IL-10 was explored the most based on our last result [2, 18, 22, 23] in this field.

IL-10 is reported to play a crucial role in allergic diseases by regulating cytokine and antibody production [24, 25]. In this study, we observed that the IL-10 level in patients with positive-specific IgE or IgG was significantly lower than that in control subjects and patients with negative-specific IgE or IgG. This finding suggests that IL-10 might be a down-regulating factor of specific antibodies in serum. This function ought to be based on the differentiation and isotype switching of B cells elicited by IL-10 [26]. The relationship of IL-10 and IgE was same

Table 5 Allele and genotype distributions of interleukin-10 (IL-10) polymorphisms in five groups

	A (<i>n</i> =52)	B (<i>n</i> =50)	C (<i>n</i> =63)	D (<i>n</i> =39)	E (<i>n</i> =86)
IL-10 (-1082)					
Allele A	81 (0.78)	60 (0.6)	96 (0.76)	45 (0.58)	104 (0.6)
Allele G	23 (0.22)* ^a	40 (0.4)	30 (0.24)* ^b	33 (0.42)	68 (0.4)
$\chi^2=10.260$ $P=0.006$					
AA	30 (0.58)	21 (0.42)	35 (0.56)	16 (0.41)	35 (0.4)
AG	21 (0.4)	18 (0.36)	26 (0.41)	13 (0.33)	34 (0.4)
GG	1 (0.02)* ^a	11 (0.22)	2 (0.03)* ^b	10 (0.26)	17 (0.2)
$\chi^2=10.999$					
IL-10-819					
Allele C	83 (0.8)	74 (0.74)	88 (0.7)	69 (0.88)	125 (0.73)
Allele T	21 (0.2)	26 (0.26)	38 (0.3)* ^b	9 (0.12)*	47 (0.27)
$\chi^2=1.832$					
CC	33 (0.63)	28 (0.56)	31 (0.49)	30 (0.77)	48 (0.56)
CT	17 (0.33)	18 (0.36)	26 (0.41)	9 (0.23)	29 (0.34)
TT	2 (0.04)	4 (0.08)	6 (0.1)* ^b	0 (0)*	9 (0.1)
$\chi^2=2.239$					

Numbers in the parentheses are allele frequency or genotype rates in the samples

Group A allergic patients with positive penicillin-specific IgE Group B allergic patients with negative penicillin-specific IgE, Group C allergic patients with positive penicillin-specific IgG, Group D allergic patients with negative penicillin-specific IgG, Group E control subjects, Allele A Allele G allele of -1082 according to single nucleotide polymorphism, AA GA GG genotype of -1082 according to single nucleotide polymorphism, Allele C Allele T allele of -819 according to single nucleotide polymorphism, CC CT TT genotype of -819 according to single nucleotide polymorphism

^a $P<0.01$ vs. B

^b $P<0.01$ vs. D

* $P<0.01$ vs. E

as some reports about other allergic diseases, whereas the association of IL-10 with IgG contrasted with the report on multiple sclerosis published in 2000 [6]. This condition should be associated with the fact that different IgG subtypes had a discrepant proportion in serum, and IL-10 had a different function to each IgG subtype. Therefore, IL-10 showed a disparate level in different diseases.

At the same time, our results showed significant negative correlation between IL-10 and specific IgE. The decrease of IL-10 level might be associated with the increase of positive-specific IgE. In addition, the results showed the level of IL-10 declined with the rise in positive-specific IgE types. These results were the same as those reported by Makela et al. [27].

From the above, we properly draw a conclusion that lower levels of IL-10 may be connected with IgE- or IgG-mediated penicillin allergy but are not cell mediated. It seems counterintuitive that IL-10, produced mainly by Th2-type lymphocytes and monocytes, has a role in the down-regulation of Th1 cytokines and should induce allergic reaction by promoting secretion of immune globulin. As we know, the balance of Th1/Th2 plays a major role in orchestrating allergic reaction. Th1 cytokines, just like IFN- γ and IL-2, inhibit secretion of Th2 cytokines, whereas Th2 cytokines, IL-4, IL-5, IL-10, and IL-13 inhibit secretion of Th1 cytokines. However, IL-10 has an inhibitory function to both Th1 and Th2 cytokines [3]. It alternates the direct of allergic reaction through adjusting the balance of Th1/Th2. IL-4, IL-5, and IL-13 secreted by Th2 cells induced and amplified the allergic response by enhancing recruitment and activation of eosinophils, basophils, and mast cells and by increasing the production of IgE. IL-10 could suppress these cytokines and induced Th2 cell anergy by acting directly on the above-mentioned inflammatory cells through its activity on other immune cells, such as dendritic (DC) and T-regulatory (Tr1) cells [28]. In a previous study, Qiao et al. [2] confirmed that levels of cytokine-like IL-4, IL-13, and IFN- γ were correlated with specific IgE in allergic patients. In Sehra et al.'s research [33], IgG-induced function was dependent on IFN- γ . In this study, results showed that patients with positive-specific IgE or IgG had lower levels of IL-10 than did the control group and negative antibody group, and converse correlation was seen in the relationship between IL-10 and IgE. Based on these studies, we might hypothesize that the lower-level IL-10 reduced control to the production of these cytokines and then these cytokines up-regulated the specific antibodies. These findings indirectly support the hypothesis that IL-10 had suppressive activity in these diseases, because the production of IgE and IgG was the key elements in the determinism of penicillin allergy and other atopic diseases [18]. All of the above demonstrates that IL-10 might play a crucial role in

penicillin allergy and Th1/Th2 balance. The relationships among IL-10, IgG isotypes, and penicillin allergy are complex, and more research is needed in this field.

We grouped 102 patients into a positive skin-test group and a patient group with allergic history. Among the patient group, there were 14 cases of anaphylactic shock, 25 of urticaria, and eight with other symptoms. According to allergic reaction types, there were 25 patients with immediate hypersensitivity reaction (IHR) and 22 with delayed hypersensitivity reaction (DHR). Serum IL-10 level was different in each group and the control group, but the difference was too small to show significant association between allergic symptoms and IL-10 production, though there was one case in which IL-10 level affected onset of the antibody-dependent autoimmune disease systemic lupus erythematosus (SLE) [29]. The same result with cytokines such as IL-4, IL-13, and IFN- γ was seen in a previous study by our group [2]. This result suggests that the clinical manifestation in penicillin allergy might not be directly attributed to cytokine level.

Another major goal in this study was to understand the role of the IL-10 gene promoter region -1082 G/A and -819 C/T genetic polymorphisms in penicillin allergy. Several polymorphisms described in the human IL-10 gene 5'-flanking sequence were believed to affect the promoter region. A possible linkage of IL-10 promoter haplotype to disease susceptibility was reported [30]. Although the strongest association was for SLE, genetic predisposition to high IL-10 expression was also associated with a higher mortality rate in meningococcal disease and a lower efficacy of IFN- α therapy in patients with chronic hepatitis C infection [4, 31]. In our study, the significant difference in genotype distribution and allele frequencies at -1082 G/A were observed between patients with positive-specific IgE and normal controls or patients with negative-specific IgE. At the same time, we observed a difference in genotype distribution at the -819 C/T position. These observations showed that the SNP at -1082 G/A position in the IL-10 gene promoter region was associated with production of specific IgE in patients with penicillin allergy, whereas the polymorphism -819 C/T might associate with specific IgG in penicillin allergy. SNPs determined IL-10 production in the promoter region, and -1082 GG and GA showed greater expression compared with the AA genotype at the same position [32]. In our study, the -1082 G/G genotype was infrequently found in patients with positive-specific IgE compared with control subjects or patients with negative-specific IgE. This might explain the lower level in the positive IgE group.

In all, this study suggests that IL-10 is an important cytokine in immune balance and has a close relationship with the production of specific antibodies in penicillin allergy but not with allergic symptoms of allergy. Poly-

morphisms in the IL-10 promoter region -1082 G/A are associated with the level of specific IgE, and -819 C/T is associated with specific IgG in serum by adjusting IL-10 secretion. IL-10 must play an important role in penicillin allergy, though the allergic mechanism is as yet unclear. Many clinical data show that there exists a close association between IL-10 and specific antibodies, and all reports, including ours, suggest IL-10 will be an important cytokine in prevention, diagnosis, and immunotherapy of penicillin allergy and other atopic diseases.

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