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Polymorphisms in *IRF5* and *TYK2* Genes Are Associated with Rheumatoid Arthritis in a Chinese Han Population

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Background: The *IRF5* and *TYK2* gene polymorphisms are associated with autoimmune diseases. However, the relationship between the *IRF5* and *TYK2* gene polymorphisms and RA risk in the Chinese Han population was inconsistent.

Material/Methods: A total of 578 RA patients (case group) and 578 healthy controls (control group) were assessed in a case-control study. Genotyping of *IRF5* (Exon 6 insertion/deletion (in/de), rs2004640, rs2070197, rs10954213) and *TYK2* (rs280500, rs280519, rs280521, rs8108236, rs12720253) was performed by direct sequencing method. Data analysis was performed by SHEsis.

Results: The rs2004640T allele (P=0.0003) and the dominant (P=0.001) and recessive (P=0.01) models of rs2004640 were associated with RA risk after stringent Bonferroni correction (0.05/4). The *IRF5* exon 6 (in), rs2070197 and rs10954213 were not associated with RA (P>0.05). Two haplotypes of *IRF5* (DTAT and DTGG) were associated with RA susceptibility (P<0.05). In addition, the frequencies of *TYK2* rs280500A, rs280521A, and rs8108236A were significantly higher in the RA group compared with the control group (P<0.05). *TYK2* rs280500, rs280521, and rs8108236 were associated with RA susceptibility in the dominant model, but the same was not observed for rs280519 and rs12720253 (P<0.05). Furthermore, 3 risk haplotypes (AAAGT, AGGAT, and GAAAT) and a protective haplotype (GAGGT) of *TYK2* gene were associated with RA susceptibility (P<0.05).

Conclusions: Our results suggest that *IRF5* rs2004640, *TYK2* rs280500, rs280521, rs8108236, and haplotypes *IRF5* (DTAT and DTGG) and *TYK2* (AAAGT, AGGAT, GAAAT, and GAGGT) are susceptible factors for RA in a Chinese Han population.

Keywords: Arthritis, Juvenile • Genetic Association Studies • Polymorphism, Genetic

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Background

Rheumatoid arthritis (RA) is a kind of multisystem inflammatory disease that mainly affects synovium and surrounding tissues [1,2]. The prevalence rate of RA is 0.8-1.0% in Europeans and Americans, and around 0.5% in Chinese [3]. The incidence is highest in adults aged 20-50 years, and the incidence rate in women is 2-3 times higher than that in men [4]. The etiology of RA is unknown and is thought to be related to genetic factors and environmental factors such as infection, smoking, and pregnancy [5-7].

Recent studies have confirmed that the interferon family and its immunomodulatory pathways, especially type I interferon (IFNs) pathway, play an important role in the pathogenesis of RA [8]. IFNs are cytokines produced, among others, by plasmacytoid dendritic cells during infection [9]. Studies have found that IFNs gene expression is abnormal in whole blood of patients with autoimmune diseases, especially systemic lupus erythematosus (SLE) [10] and RA [11]. Genetic association analysis has confirmed that interferon regulator 5 (*IRF5*) and tyrosine kinase 2 (*TYK2*) in the type I interferon pathway are risk genes for SLE [12]. RA and SLE have similar autoimmune abnormalities, and the polymorphisms of *IRF5* and *TYK2* genes may also affect susceptibility to RA.

IRF-5 is a member of the IRF family of transcription factors and plays an important role in inflammation and autoimmune response [13]. The gene encoding IRF5 maps to chromosome 7q32 [14]. Overexpression of *IRF5* gene leads to increased levels of IFN and IL6 proteins and promotes the occurrence and development of inflammation in autoimmune diseases [15]. rs2004640 is located in the cleavage region of exon 1 at the 5' end of the *IRF5* gene, and different bases can form a variety of *IRF5* mRNA splices, affecting the stability of the *IRF5* gene [16]. In addition, rs10954213 was previously reported to change the polyadenylate site of *IRF5* and is associated with *IRF5* mRNA overexpression [17]. Multiple case-control association studies have reported the genetic association of *IRF5* gene polymorphisms with RA in white and Asian populations [18-20]. However, the susceptibility loci of the *IRF5* gene were inconsistent in different races, and there was racial heterogeneity. Moreover, the genetic association between the *IRF5* gene polymorphisms and RA risk in Chinese Han populations were controversial.

TYK2 is a member of the non-receptor tyrosine kinase-linked Janus kinase (JAK) family and is part of the Janus kinase/signal transduction and transcriptional activator 4(JAK-STAT) pathway [21]. Several studies have confirmed the important role of TYK2 in the type I interferon signaling pathway [22,23], and in SLE [12], systemic sclerosis (SS) [24], multiple sclerosis [25], and other immunological diseases. Zheng et al reported that expression of the *TYK2* gene is closely related to RA disease progression and may be involved in the development of RA by regulating the levels of IL-2, IL-17, and IL-21 [26]. The association between variants in the *TYK2* gene and increased expression of type 1 IFN gene has been identified in a previous study [27]. Graham et al indicated that *TYK2* rs280500 was a susceptibility locus for SLE [28]. Subsequently, Tang et al revealed that *TYK2* rs280500, rs8108236, and rs280519 polymorphisms were significantly associated with SLE risk [12]. However, no study has investigated the polymorphisms of the *TYK2* gene and RA risk in a Chinese Han population.

Therefore, the present study sought to investigate the genetic association between *IRF5* and *TYK2* gene polymorphisms and RA susceptibility in a Chinese Han population.

Material and Methods

Samples

The Ethics Committee of Changshu Hospital Affiliated to Soochow University (CSHEC-190922) approved the study protocol. Written informed consent for genetics analysis was obtained from all subjects. The case group was composed of 578 unrelated patients (51 men and 527 women) from Jiangsu province who fulfilled the American College of Rheumatology 1982 criteria for RA [29]. The control group was composed of 578 healthy controls (55 men and 523 women) matched for sex, ethnicity, and age. Clinical features of RA patients and controls are shown in **Table 1**. All the individuals were of Chinese Han ethnicity.

Single-Nucleotide Polymorphisms (SNPs) Selection and Genotyping

Genomic DNA was extracted from peripheral leukocytes using the standard phenol-chloroform method. The SNP selection of the *IRF5* gene was performed using the methods described by Tang et al [12]. Four SNPs in the *IRF5* gene were selected: exon 6 (de/in), rs2004640 (G/T), rs2070197 (C/T), and rs10954213 (G/A). The SNP selection of the *TYK2* gene was performed using Haploview Software with minor allele frequency (MAF) higher than 0.05 and r2 ≥0.8 based on the HapMap database (CHB, Chinese Han population) (*http://www.hapmap.org/index.-html.ja*). After screening, 5 tag SNPs (rs280500, rs280519, rs280521, rs8108236, and rs12720253) were selected. All the SNPs were genotyped by direct sequencing with the ABI 3730XL DNA Sequencer.

Statistical analysis

SHEsis software was used in statistical analysis (*http://analysis.bio-x.cn/myAnalysis.php*). The χ^2 test was

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Table 1. Clinical Characteristics of patients with rheumatoid arthritis and healthy controls (n=578).

Clinical features	Case	Control	p
Male/Female	51/527	55/523	>0.05
Age (years)	44.7±12.3	45.6±12.8	>0.05
ESR (mm/h)	35.1±14.2	12.1±8.5	<0.05
CRP (mg/l)	26.5±11.2	4.7±12.5	<0.05
HLA-B27+, %	1/578 (0.17%)	0/578 (0%)	>0.05
ANA+, %	102/578 (17.6%)	0/578 (0%)	<0.05
RF+, %	224/578 (387%)	0/578 (0%)	<0.05

ANA – antinuclear antibodies; ESR – erythrocyte sedimentation rate; CRP – C-reactive protein; RF – rheumatoid factor. '+' – positive.

Table 2. Distributions of the genotypes of IRF5 and TYK2 polymorphisms in cases and controls.

Genes	SNP (A/B)	Genotype (AA/AB/BB)	Genotype frequencies (%)		[a] P, OR (95% CI) (AB+BB vs AA)	[b] P, OR (95% CI) (BB vs AA+AB)	Power
			Cases	Controls			
IRF5	Exon 6 (D/I)	DD/DI/II	138/278/162	157/288/133	0.20, 1.19 [0.91, 1.55]	0.05, 1.30 [1.00, 1.70]	95.6
	rs2070197(T/C)	TT/CT/CC	571/7/0	575/3/0	0.22, 2.35 [0.60, 9.13]	NA	
	rs10954213(A/G)	AA/GA/GG	277/231/70	265/256/57	0.48, 0.92 [0.73, 1.16]	0.22, 1.26 [0.87, 1.82]	
	rs2004640(G/T)	GG/GT/TT	317/232/29	371/193/14	0.001, 1.49 [1.17, 1.88]	0.01, 2.30 [1.18, 4.46]	
TYK2	rs280500(G/A)	GG/AG/AA	277/260/41	347/202/29	0.0001, 1.63 [1.29, 2.06]	0.14, 1.45 [0.89, 2.36]	
	rs280519(A/G)	AA/GA/GG	157/269/152	182/270/126	0.11, 1.23 [0.96, 1.59]	0.07, 1.28 [0.98, 1.68]	
	rs280521(G/A)	GG/AG/AA	324/229/25	423/140/15	1.35e-008, 2.14 [1.67, 2.74]	0.11, 1.70 [0.89, 3.25]	
	rs8108236(G/A)	GG/AG/AA	311/221/46	372/175/31	0.0003, 1.55 [1.22, 1.96]	0.08, 1.53 [0.95, 2.44]	
	rs12720253(T/G)	TT/GT/GG	499/77/2	475/101/2	0.05, 0.73 [0.53, 1.00]	1.00, 1.00 [0.14, 7.12]	

[a] P _dominant model; [b] P –recessive model; SNP – single nucleotide polymorphism; OR – odds ratio, 95% CI – 95% confidence intervals, NA – not available; de(D)/in(I) – deletion/insertion; A – allele with major frequency; B – allele with minor frequency.

used to analyze the significance of Hardy-Weinberg equilibrium (HWE), genotype, and allele frequency of single-nucleotide polymorphisms. Stringent Bonferroni correction was applied to correct the *P* values obtained by logistic regression, in multiple comparisons, for associations with RA (*P*=0.05/N). Haploview 4.2 software was used for linkage disequilibrium (LD) analysis and haplotype analysis. *P*<0.05 was considered statistically significant. Calculation power was obtained at the 0.05 level of significance, assuming an OR of 1.5 (small effect size) by using G*Power software (*www.gpower.hhu.de*).

Results

The genotype distributions of all the included polymorphisms were in HWE (P>0.05) (**Supplementary Table 1**). Moreover, the statistical power was 95.6%, which indicated that the results were not be influenced by the sample size in this study (**Table 2**). LD analysis showed that IRF5 rs2070197 and rs10954213, as well as rs2070197 and rs2004640, were in strong LD (D'>0.85) (**Figure 1A**), while TYK2 rs8108236 and rs12720253 were in weak LD (D'=0.56) (**Figure 1B**).

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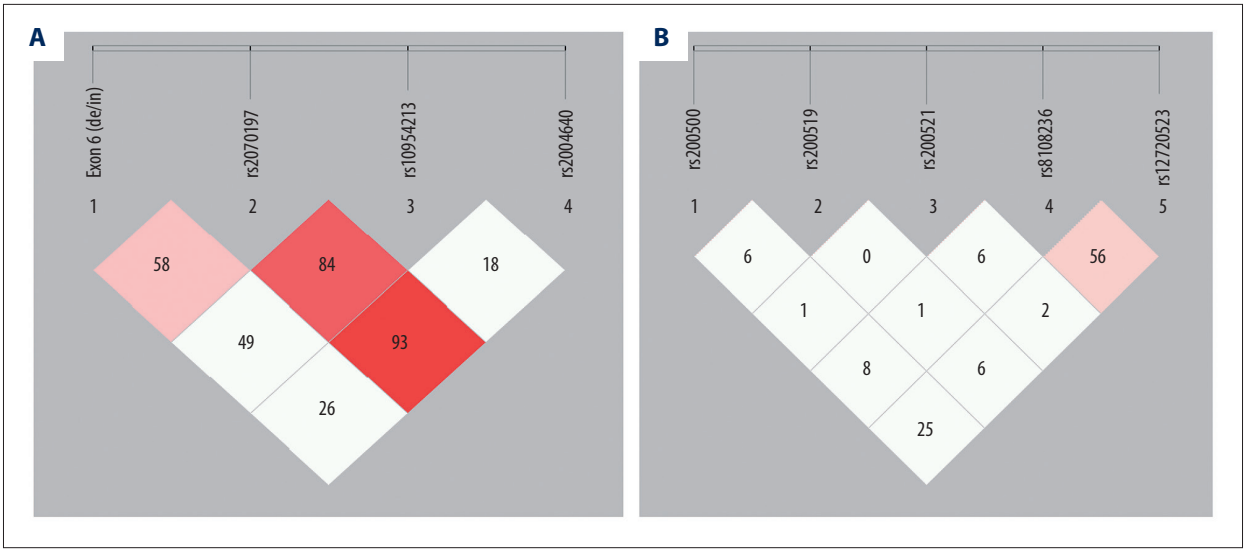

Figure 1. Pairwise linkage disequilibrium of IRF5 and TYK2 single-nucleotide polymorphisms in relation to rheumatoid arthritis. The numbers in the squares refer to the pairwise linkage disequilibrium measured as D'. Haplotype blocks were defined using a setting pairwise D'. (**A**) Linkage disequilibrium of IRF5 gene. (**B**) Linkage disequilibrium of TYK2 gene.

Table 3. The association between the alleles of IRF5 and TYK2 gene polymorphisms and RA risk.

	SNP (A/B)	Position	Case (N=578)	Control (N=578)	P (B vs A+B)	OR (95% CI)
IRF5	Exon 6 (D/I)	Exon 6	602	554	0.05	0.85 [0.72-0.99]
	rs2070197(T/C)	3'-UTR	7	3	0.21	2.34 [0.60-9.07]
	rs10954213(A/G)	3'-UTR	371	370	0.96	1.00 [0.84-1.20]
	rs2004640(G/T)	Exon 1	290	221	0.0003	1.43 [1.17-1.74]
TYK2	rs280500(G/A)	5' UTR	342	260	0.0001	1.45 [1.20, 1.75]
	rs280519(A/G)	Intron	573	522	0.03	0.84 [0.71-0.98]
	rs280521(G/A)	Intron	279	170	1.06e-008	1.85 [1.49-2.28]
	rs8108236(G/A)	Intron	313	237	0.0002	1.44 [1.19-1.75]
	rs12720253(T/G)	Intron	81	105	0.07	0.75 [0.56-1.02]

SNP – single nucleotide polymorphism; OR – odds ratio, 95% CI – 95% confidence intervals; de(D)/in(I) – deletion/insertion; UTR – untranslated region; A – allele with major frequency; B – allele with minor frequency.

Association of IRF5 Polymorphisms and RA

The frequency of rs2004640T was significantly higher in cases than in controls (P=0.0003) after stringent Bonferroni correction (0.05/4) (**Table 3**). Genotype analysis ascertained that rs2004640 was associated with RA according to the dominant and recessive models (dominant model: *P*=0.001; recessive model: *P*=0.01) after stringent Bonferroni correction (0.05/4) (**Table 3**). No significant difference in the other 3 variants – exon 6 (de/in), rs2070197, and rs10954213 – were detected between the cases and healthy controls (*P*>0.05) (**Tables 2, 3**).

As shown in **Table 4**, 8 major haplotypes were identified by 3 SNPs and the exon 6 insertion/deletion with the lowest frequency threshold (LFT)>0.01). Significant associations were observed between the haplotype (I_{H1}) involving exon 6 (de), rs2070197T, rs10954213A, and rs2004640T and RA (*P*=0.0009, OR (95%CI): 1.54 [1.191~1.976]) after stringent Bonferroni correction (0.05/8) (**Table 2**). Additionally, a protective haplotype (I_{H4}) carrying the exon 6 deletion, rs2070197T, rs10954213G, and rs2004640G, was identified (*P*=4.45e-005, OR (95%CI): 0.48 [0.337~0.688]) after stringent Bonferroni correction (0.05/8) (**Table 2**).

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Table 4. Haplotypes structure and frequencies of IRF5 and TYK2 gene polymorphism.

Genes	Number	Haplotypes	Case		Control		P	OR [95%CI]
IRF5	I_{H1}	DTAT	167	(0.144)	115	(0.099)	0.0009	1.54 [1.191~1.976]
	I_{H2}	DTAG	317	(0.274)	370	(0.320)	0.02	0.81 [0.673~0.962]
	I_{H3}	DTGT	18	(0.016)	22	(0.019)	0.61	0.85 [0.455~1.591]
	I_{H4}	DTGG	48	(0.041)	95	(0.082)	4.45e-005	0.48 [0.337~0.688]
	I_{H5}	ITAT	61	(0.052)	37	(0.032)	0.01	1.69 [1.114~2.572]
	I_{H6}	ITAG	259	(0.224)	261	(0.226)	0.95	0.99 [0.817~1.208]
	I_{H7}	ITGT	44	(0.038)	46	(0.039)	0.87	0.97 [0.632~1.472]
	I_{H8}	ITGG	236	(0.204)	207	(0.179)	0.12	1.18 [0.957~1.449]
TYK2	T_{H1}	AAAGT	43	(0.037)	14	(0.012)	9.88e-005	3.18 [1.723~5.878]
	T_{H2}	AAGAT	35	(0.030)	24	(0.021)	0.15	1.46 [0.866~2.472]
	T_{H3}	AAGGG	12	(0.011)	10	(0.009)	0.66	1.20 [0.524~2.753]
	T_{H4}	AAGGT	83	(0.072)	80	(0.069)	0.79	1.04 [0.758~1.434]
	T_{H5}	AGAGT	26	(0.022)	13	(0.011)	0.05	1.96 [1.005~3.838]
	T_{H6}	AGGAT	67	(0.058)	29	(0.025)	9.82e-005	2.35 [1.513~3.667]
	T_{H7}	AGGGT	73	(0.063)	80	(0.069)	0.55	0.90 [0.651~1.255]
	T_{H8}	GAAAT	42	(0.036)	17	(0.015)	0.001	2.52 [1.427~4.460]
	T_{H9}	GAAGT	58	(0.050)	48	(0.042)	0.34	1.21 [0.819~1.789]
	T_{H10}	GAGAT	71	(0.061)	87	(0.075)	0.18	0.80 [0.578~1.108]
	T_{H11}	GAGGT	220	(0.190)	304	(0.263)	2.60e-005	0.65 [0.538~0.799]
	T_{H12}	GGAAT	12	(0.011)	9	(0.008)	0.55	1.29 [0.552~3.032]
	T_{H13}	GGAGT	56	(0.049)	54	(0.046)	0.82	1.04 [0.713~1.535]
	T_{H14}	GGGAT	58	(0.050)	59	(0.051)	0.87	0.97 [0.668~1.408]
	T_{H15}	GGGGG	23	(0.020)	33	(0.029)	0.41	0.92 [0.745~1.126]

D – deletion; I – insertion; OR – odds ratio; 95% CI – 95% confidence intervals, '–' – not calculated. a The program, Plink, was used to estimate common (frequency[0.01) haplotypes constructed by the SNPs of IRF5 (exon 6 (in/de), rs2070197, rs10954213, rs2004640) and five SNPs of TYK2 genotyped (rs280500, rs280519, rs280521, rs8108236, rs12720253). b Each haplotype was compared with the other haplotypes combined. c The Bonfferoni correction was applied to correct the p value.

Association of TYK2 Polymorphisms and RA

Significant association between rs280500A, rs280521A, and rs8108236A and RA were observed after the stringent Bonferroni correction (0.05/5) (rs280500: *P*=0.0001; rs280521: *P*=1.06e-008; rs8108236: *P*=0.0002) (**Table 3**). Similar results were obtained for the association between the dominant model of rs280500, rs280521, and rs8108236 and RA (rs280500: *P*=0.0001; rs280521: *P*=1.35e-008; rs8108236: P=0.0003) after stringent Bonferroni correction (0.05/5) (**Table 2**). No correlation was detected between the *TYK2* rs280519 and rs12720253 polymorphisms and RA risk (*P*>0.05) (**Tables 2, 3**).

As shown in **Table 4**, 15 major haplotypes containing rs280500, rs280519, rs280521, rs8108236, and rs12720253 alleles with the lowest frequency threshold (LFT)>0.01) were identified. Haplotypes containing rs280500A- rs280519A- rs280521A- rs8108236G- rs12720253T(T_{H1}) (*P*=9.88e-005, OR (95% CI): 3.18 [1.723~5.878]), rs280500A- rs280519G- rs280521G- rs8108236A- rs12720253T(T_{H6}) (*P*=9.82e-005, OR (95% CI): 2.35 [1.513~3.667]), and rs280500G- rs280519A- rs280521A- rs8108236A- rs12720253T (T_{H8}) (*P*=0.001, OR (95% CI): 2.52 [1.427~4.460]) were shown to be significantly associated with RA after stringent Bonferroni correction (0.05/15). A protective haplotype – rs280500G- rs280519A-rs280521G- rs8108236G- rs12720253T (T_{H11}) – was also identified in our study (*P*=2.60e-005, OR (95% CI): 0.65 [0.538~0.799]) after stringent Bonferroni correction (0.05/15).

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Discussion

Our results confirmed significant associations between rs2004640 and RA in a Chinese Han population according to allele, dominant, and recessive models. The frequencies of rs2004640 T allele and TT genotype were lower in controls than in the RA cases, which is consistent with previous studies conducted in Chinese Han populations with RA [30,31]. Kozyrev et al found that *IRF5* gene rs2004640 had significant ethnic heterogeneity [32]. Maalej et al reported a close association between rs2004640 and RA in Tunisians, but not in Spanish, Swiss, or Argentine populations [19]. In the present study, we verified that rs2004640 was associated with RA in a Han population in Jiangsu, China. Although the frequency of rs2004640 T allele in Chinese Han RA patients in this study was much lower than that in white RA patients, the correlation of rs2004640T allele with RA risk remained. Therefore, the *IRF5* rs2004640T allele is a risk factor of RA in both the white and Asian populations.

No0, no association was found between the *IRF 5* exon 6 (de/in), rs2070197 (C/T), and rs10954213 (G/A) and RA susceptibility in the Chinese Han population. No association between rs10954213 and RA was identified in the present study, which agrees with the results reported by Li et al [30]. Moreover, rs2070197 was not found to be polymorphic in our sample. This polymorphism has been shown to be strongly correlated with SLE in Argentina, Spain, and Germany [32], although no genetic association was identified for *IRF 5* exon 6 (de/in), rs2070197, and rs10954213 and RA risk in this study. The difference between white and Chinese cohorts may explain this inconsistency. In addition, there could be other variants in linkage disequilibrium in this gene conferring RA susceptibility.

Significant associations were detected between the rs280500A, rs280521A, and rs8108236A alleles in *TYK2*, as well as the dominant model of *TYK2* rs280500, rs280521, and rs8108236 and RA risk. As for the variants in non-coding regions of *TYK2*, most of the variants were reported in SLE. The *TYK2* polymorphisms were reported to be significantly associated with SLE risk in Scandinavian and Finnish populations [33]. The rs280500 polymorphism, located upstream of the 5' UTR of the *TYK2* gene, has been investigated in patients with SLE in white and Asian populations. Only 1 study has reported a significant association between *TYK2* rs280500 polymorphism and SLE risk in a Chinese Han population [12]. The *TYK2* rs280500 polymorphism was not related to SLE risk in other populations in Chinese Han [34] and whites [33]. Although we found that rs280500 polymorphism was significantly associated with RA risk in a Chinese Han population, the function of this polymorphism in the pathogenesis of RA needs further investigation.

Few studies have been conducted on the association between *TYK2* rs280521 and rs8108236 polymorphisms and autoimmune diseases susceptibility. Our study has shown a significant association between *TYK2* rs280521 and RA risk for the first time, which was different from the results reported by Sigurdsson et al [33] in patients with SLE. We also found that *TYK2* rs8108236 polymorphism was significantly associated with RA risk, which was different from that reported by Tang et al [12] and Li et al [30] in patients with SLE in Chinese Han populations. These conflicting results suggest there is a difference in the pathogenesis of SLE and RA. Although the function of *TYK2* rs280521 and rs8108236 polymorphisms in RA is unclear, we may hypothesize that these variants are in moderate to high LD with other functional variants. Thus, further investigation with a larger sample size and multiple ethnicities is necessary.

Furthermore, research in a white population confirmed a significant association between *TYK2* rs280519 and SLE [35]. However, no association was found between *TYK2* rs280519 and SLE in a Chinese Han population [12] or in a Japanese population [36]. In the present study, we found that *TYK2* rs280519 was not associated with RA risk in a Chinese Han population in Jiangsu province, which may indicate *TYK2* rs280519 is a susceptible factor for autoimmune diseases including SLE and RA in whites but not in Asians. To confirm this result, larger studies with multiple ethnicities are necessary in the future.

Haplotype analysis has shown that the haplotype (I_{H1}) involving exon 6 (de), rs2070197T, rs10954213A, and rs2004640T and haplotype (I_{H4}) carrying the exon 6 deletion, rs2070197T, rs10954213G, and rs2004640G were associated with the RA susceptibility. This is the first time that haplotypes defined by exon 6, rs2070197, rs10954213, and rs2004640 polymorphisms were found to be associated with RA in a Chinese Han population. However, comparison of *IRF5* haplotypes among Japanese [37], whites [28], and Han Chinese [12] indicated differences between Whites and Han Chinese. The white risk haplotype was not present in Japanese and Han Chinese for the absence of rs2070197C allele. Instead, a risk haplotype [(exon 6 (de)- rs2070197T- rs10954213A- rs2004640T)] was identified in Han Chinese. Furthermore, neither of these 2 susceptible haplotypes [exon 6 (in)- rs10954213A- rs2004640T and exon 6 (de)- rs10954213A- rs2004640G] that were identified in patients with SLE [12] have been found in patients with RA in a Chinese Han population, which may indicate the different pathogenesis RA and SLE. Regarding the haplotypes of *TYK2* gene polymorphisms in RA, we found 2 risk haplotypes and 1 protective haplotype defined by rs280500, rs280519, rs280521, rs8108236, and rs12720253 polymorphisms in patients with RA in a Chinese Han population for the first time. Notably, the SNPs included in the haplotypes in our research were a little different from those in Tang et al [12]. Therefore,

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to confirm these results, larger case-control studies are necessary in the future.

There are limitations in the present study. First, although the calculation power indicated that the sample size is large enough to detect an association at an odds ratio of 1.5, the sample size included in the study was relatively small. Our results need to be confirmed in studies with larger sample sizes. Second, both genetic and environmental factors were determined to play a role in the development of RA, but we did not assess the influence of environmental factors and RA risk due to insufficient data.

Conclusions

Our results suggest that *IRF5* rs2004640, *TYK2* rs280500, rs280521, and rs8108236 are risk factors for RA in a Chinese Han population, and haplotypes of *IRF5* DTAT and DTGG, and *TYK2* AAAGT, AGGAT, GAAAT, and GAGGT may be risk factors for RA susceptibility in a Chinese Han population.

Conflict of Interest

None.

Supplementary Data

Supplementary Table 1. The Hardy-Weinberg equilibrium of *IRF5* and *TYK2* gene polymorphisms in case and control groups.

Genes	SNP ID	Position	Case (p)	Control (p)
IRF5	Exon 6	Exon 6	0.38	0.96
	rs2070197	7: 128948946	0.88	0.95
	rs10954213	7: 128949373	0.05	0.67
	rs2004640	7: 128938247	0.10	0.06
TYK2	rs280500	19: 10351402	0.06	0.95
	rs280519	19: 10333933	0.09	0.71
	rs280521	19: 10334392	0.05	0.40
	rs8108236	19: 10326832	0.44	0.08
	rs12720253	19: 10339683	0.59	0.16

References:

1. Wells PM, Adebayo AS, Bowyer RCE, et al. Associations between gut microbiota and genetic risk for rheumatoid arthritis in the absence of disease: A cross-sectional study. Lancet Rheumatology, 2020;2(7):e418-27
2. Mehta N, Schneider LK, McCardell E. Rheumatoid arthritis: Selecting monotherapy versus combination therapy. J Clin Rheumatol, 2017 [Online ahead of print]
3. Gulati M, Farah Z, Mouyis M. Clinical features of rheumatoid arthritis. Medicine, 2018;211-15
4. Kawahito Y. [Guidelines for the management of rheumatoid arthritis.] Nippon Rinsho, 2016;74(6):939-43 [in Japanese]
5. Kim H, Sookgyung Cho, Lee J, et al. Increased risk of opportunistic infection in early rheumatoid arthritis. Int J Rheum Dis, 2019;22(1):355-60
6. Ummarino D. Smoking influences autoimmunity to vimentin. Nat Rev Rheumatol, 2016;12:624
7. Smith WD, West HF. Pregnancy and rheumatoid arthritis. Acta Rheumatologica Scandinavica, 2006;6(1-4):189-201
8. Huang JJ, Feng G, Li Z, et al. Clinical implications of type I interferon signature gene expression in patients with rheumatoid arthritis. Acta Academiae Medicinae Wannan, 2018;3
9. Uddin S, Platanias LC. Mechanisms of type-I interferon signal transduction. J Biochem Mol Biol, 2004;37(6):635-41
10. Rönnblom L, Eloranta ML, Alm GV. The type I interferon system in systemic lupus erythematosus. Arthritis Rheum, 2010;54(2):408-20
11. Higgs BW, Liu Z, Zhu W, et al. Type I interferon in rheumatoid arthritis, systemic lupus erythematosus, myositis and systemic scleroderma – a review of transcript profiling studies in the blood. European Musculoskeletal Review, 2012;7(1):22-28
12. Tang L, Wan PC, Wang Y, et al. Genetic association and interaction between the IRF5 and TYK2 genes and systemic lupus erythematosus in the Han Chinese population. Inflamm Res, 2015;64(10):817-24
13. Hammami A, Charpentier T, Smans M, et al. IRF-5-mediated inflammation limits CD8+ T cell expansion by inducing HIF-1α and impairing dendritic cell functions during leishmania infection. PLoS Pathog, 2015;11(6):e1004938
14. Reykande SE, Rezaei A, Sadr M, et al. Association of interferon regulatory factor 5 (IRF5) gene polymorphisms with juvenile idiopathic arthritis. Clin Rheumatol, 2018;37(10):2661-65
15. Weiss M, Byrne AJ, Blazek K, et al. IRF5 controls both acute and chronic inflammation. Proc Natl Acad Sci, 2015;112(35):11001-6
16. van der Helm-van Mil, Toes REM, Huizinga TWJ. Genetic variants in the prediction of rheumatoid arthritis. Ann Rheum Dis, 2010;69(9):1694-96
17. Graham RR, Kozyrev SV, Baechler EC, et al. A common haplotype of interferon regulatory factor 5 (IRF5) regulates splicing and expression and is associated with increased risk of systemic lupus erythematosus. Nat Genet, 2006;38(5):550-55
18. Rueda B, Reddy MVPL, González-Gay MA, et al. Analysis of IRF5 gene functional polymorphisms in rheumatoid arthritis. Arthritis Rheum, 2014;54(12):3815-19
19. Maalej A, Hamad MB, Reba A, et al. Association of IRF5 gene polymorphisms with rheumatoid arthritis in a Tunisian population. Scand J Rheumatol, 2008;37(6):414-18
20. Shimane K, Kochi Y, Yamada R, et al. A single nucleotide polymorphism in the IRF5 promoter region is associated with susceptibility to rheumatoid arthritis in the Japanese patients. Ann Rheum Dis, 2008;68(3):377-83
21. Yin Q, Wu LC, Zheng L, et al. Comprehensive assessment of the association between genes on JAK-STAT pathway (IFIH1, TYK2, IL-10) and systemic lupus erythematosus. A meta-analysis. Arch Dermatol Res, 2018;310:711-28
22. Yokota SI, Yokosawa N, Kubota T, et al. Herpes simplex virus type 1 suppresses the interferon signaling pathway by inhibiting phosphorylation of STATs and Janus kinases during an early infection stage. Virology, 2001;286(1):119-24
23. Gongora C, Mechti N. [Interferon signaling pathways.] Bull Cancer, 1999;86(11):911-19 [in French]

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24. Johnson BA, Wang J, Taylor EM, et al. Multiple sclerosis susceptibility alleles in African Americans. Genes Immun, 2010;11(4):343-50
25. Dyment DA, Cader MZ, Chao MJ, et al. Exome sequencing identifies a novel multiple sclerosis susceptibility variant in the TYK2 gene. Neurology, 2012;79(5):406-11
26. Zheng XL, Yao ZJ. [Study on expression and role of TYK2 gene in rheumatoid arthritis.] Chinese Journal of Immunology, 2020;36:1868-73 [in Chinese]
27. Mori Y, Hirose K, Suzuki K, et al. Tyk2 is essential for IFN-alpha-induced gene expression in mast cells. Int Arch Allergy Immunol, 2004;134(Suppl. 1):25-29
28. Graham RR, Kyogoku C, Sigurdsson S, et al. Three functional variants of IFN regulatory factor 5 (IRF5) define risk and protective haplotypes for human lupus. Proc Natl Acad Sci USA, 2007;104:6758-63
29. Arnett FC, Edworthy SM, Bloch DA, et al. The American Rheumatism Association 1987 revised criteria for the Classification of rheumatoid arthritis, Arthritis Rheum, 1988;31:315-24
30. Li XB, Li T, Yan MF, et al. [Association of interferon regulatory factor 5 gene polymorphisms with rheumatoid arthritis in Shanxi Han Chinese population.] Chin J Rheumatol, 2015;19(7):440-46 [in Chinese]
31. Shen GY, Shu R, Cui LF, et al. [Association of interferon regulatory factor 5 gene polymorphism with rheumatoid arthritis.] Suzhou University Journal of Medical Science, 2010;30(6):1246-48 [in Chinese]
32. Kozyrev SV, Lewe'n S, Reddy PM, et al. Structural insertion/deletion variation in IRF5 is associated with a risk haplotype and defines the precise isoforms expressed in SLE. Arthritis Rheum, 2007;56:1234-41
33. Sigurdsson S, Nordmark G, Göring HH, et al. Polymorphisms in the tyrosine kinase 2 and interferon regulatory factor 5 genes are associated with systemic lupus erythematosus. Am J Hum Genet, 2005;76:528-37
34. Li P, Chang YK, Shek KW, Lau YL. Lack of association of TYK2 gene polymorphisms in Chinese patients with systemic lupus erythematosus. J Rheumatol, 2011;38:177-78
35. Cunninghame Graham DS, Morris DL, et al. Association of NCF2, IKZF1, IRF8, IFIH1, and TYK2 with systemic lupus erythematosus. PLoS Genet, 2011;7:e1002341
36. Kyogoku C, Morinobu A, Nishimura K, et al. Lack of association between tyrosine kinase 2 (TYK2) gene polymorphisms and susceptibility to SLE in a Japanese population. Mod Rheumatol, 2009;19:401-6
37. Kawasaki A, Kyogoku C, Ohashi J, et al. Association of IRF5 polymorphisms with systemic lupus erythematosus in a Japanese population: Support for a crucial role of intron 1 polymorphisms. Arthritis Rheum, 2008;58:826-34

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