Send Orders for Reprints to reprints@benthamscience.ae

The Open Rheumatology Journal, 2016, *10*, 1-4

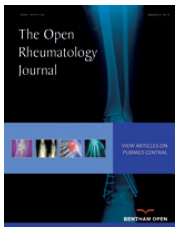
BENTHAM OPEN

The Open Rheumatology Journal

Supplementary Material

Content list available at: www.benthamopen.com/TORJ/

DOI: 10.2174/1874312901610010001



Association Study of Single Nucleotide Polymorphisms Rs4552569/Rs17095830 With Ankylosing Spondylitis In A Chinese Population

Qingwen Wang[1], Yuanyuan Yang[1], Jiyang Lv[1], Qi Lin[1], Luo Wang[2] and Zhengyu Fanga[1,2,*]

[1] *Department of rheumatism and immunology, Peking University Shenzhen Hospital, Shenzhen, Guangdong, PR China*

[2] *Biomedical Research Institute, Shenzhen Peking University- The Hong Kong University of Science and Technology Medical Center, Guangdong Province, China*

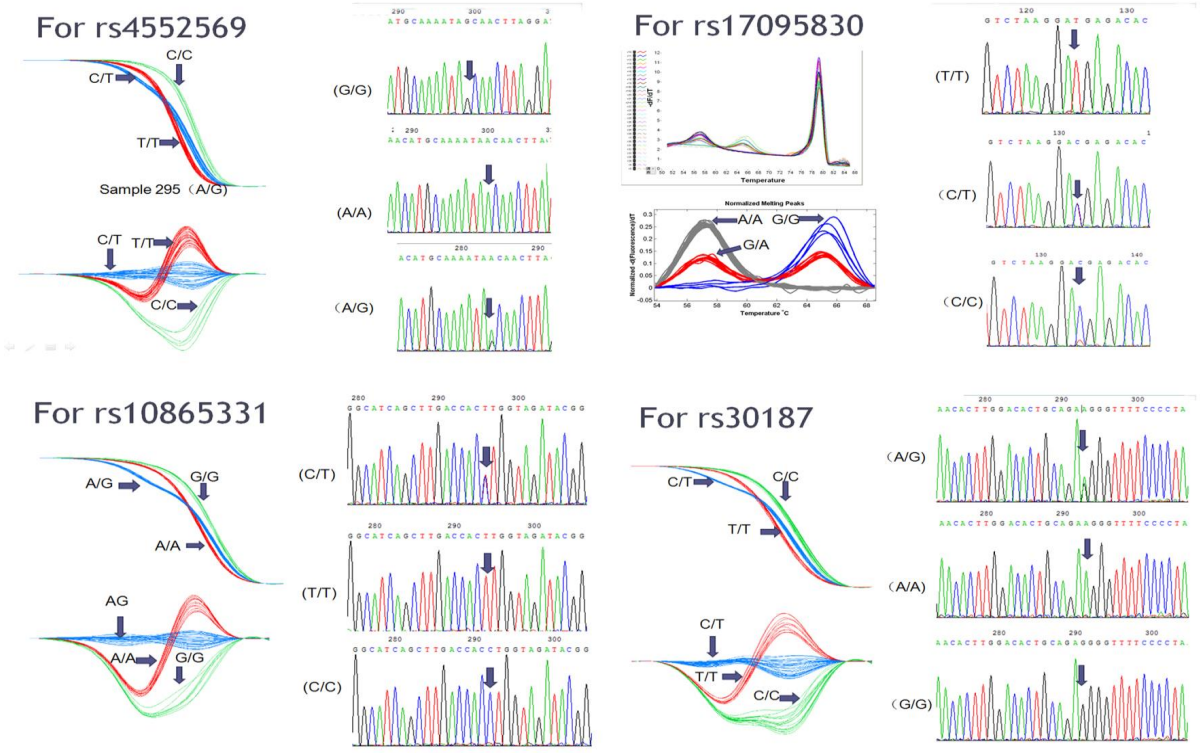


Fig (S1). Auto- grouping graphics of the indicated SNPs using LightCyclersoftware (version 1.5.0) and the results of verification by DNA sequencing.

Supplementary Table S1: Extraspinal manifestations of the AS patients enrolled.

Extraspinal manifestations		No. of cases	Percentage (%)
Enthesitis	With Symptom	125	17.01
	Symtom + evidence	26	3.54
Mucocutaneous manifestations	Skin lesion	5	0.68
	Dental ulcer	15	2.04
	Conjunctivitis	1	0.14
	Balanitis	2	0.27
Eye	Uveitis	64	8.71
Intestinal tract	Inflammatory bowel disease	49	6.67
Cardiovascular manifestations	Macroangiopathy & valve disease	17	2.31
	Abnormal electrocardiogram	296	40.27
Lung	Interstitial pneumonia	68	9.25
Nerve	Atlanto-axial Joint Dislocation	4	0.54
Kidney & urinary system	Abnormal urinalysis	124	16.87
	Kidney or urinary system disease	13	1.77

Supplementary Table S2: Sequences of primer sets used for SNP genotyping.

SNP	Forward	Reverse	unlabeled C3-blocked probe
rs4552569	CCCACAGACTGTCTCTTCGT	TGAAAGGCCTCCATTTGTAGC	N/A
rs17095830	CAGCAACCAGAACATTCCCA	AGGTTGTCTGGAACTTGGGG	TTGTCTAAGGACGAGACACACTCATA
rs10865331	GCAAGGTCTAAGTGACTGACT	GCAATGGCATCAGCTTGAC	N/A
rs30187	CCTCACTGTGATGGTTATTAG	TGATGAACACTTGGACACT	N/A

Supplementary Table S3: Genotype and allele frequencies of indicated polymorphisms in male or female AS patients.

		male (%)	**female (%)**		**male (%)**	**female(%)**	**Genot ype**	**Allelic**
	Genotype	(n =576)	(n =159)	Allele	(n =576)	(n =159)	p value	p value
rs4552569	CC	52 (9.0)	16 (10.1)	C	376 (32.6)	95 (29.9)	0.2332	0.3496
	CT	272 (47.2)	63 (39.6)	T	776 (67.4)	223 (70.1)		
	TT	252 (43.8)	80 (50.3)					
rs17095830	GG	6 (1.0)	3 (1.9)	G	136 (11.8)	45 (14.2)	0.4787	0.2598
	AG	124 (21.5)	39 (24.5)	A	1016 (88.2)	273 (85.8)		
	AA	446 (77.5)	117 (73.6)					
rs10865331	AA	174 (30.2)	39 (24.5)	A	634 (55.0)	156 (49.1)	0.05839	0.1588
	AG	286 (49.7)	78 (49.1)	G	518 (45.0)	162 (50.9)		
	GG	116 (20.1)	42 (26.4)					
rs30187	TT	166 (28.8)	43 (27.0)	C	619 (53.7)	169 (53.1)	0.8523	0.8612
	CT	287 (49.8)	83 (52.2)	T	533 (46.3)	149 (46.9)		
	CC	123 (21.4)	33 (20.8)					

Supplementary Table S4: Genotype and allele frequencies of indicated polymorphisms in juvenile (<18) or adult ≧ 18) on set AS patients

		Adult (%)	**Juvenile (%)**		**Adult (%)**	**Juvenile (%)**	**Genotype**	**Allelic**
	Genotype	**(n =573)**	**(n =162)**	**Allele**	**(n =573)**	**(n =162)**	**p value**	**p value**
rs4552569	CC	57 (9.9)	11 (6.8)	C	373 (32.5)	98 (30.2)	0.4722	0.4332
	CT	259 (45.2)	76(46.9)	T	773 (67.5)	226 (69.8)		
	TT	257 (44.9)	75(46.3)					
rs17095830	GG	6 (1.1)	3 (1.9)	G	137 (12.0)	44 (13.6)	0.4317	0.6312
	AG	125 (21.8)	38 (23.5)	A	1009 (88.0)	280 (86.4)		
	AA	442 (77.1)	121 (74.6)					
rs10865331	AA	168 (29.3)	48 (29.6)	A	624 (54.5)	172 (53.1)	0.6562	0.6636
	AG	288 (50.3)	76 (46.9)	G	522 (45.5)	152 (46.9)		
	GG	117 (20.4)	38 (23.5)					
rs30187	TT	153 (26.7)	56 (34.6)	C	600 (52.4)	188 (58.0)	0.0708	0.1388
	CT	294 (51.3)	76 (46.9)	T	546 (47.6)	136 (42.0)		
	CC	126 (22.0)	30 (18.5)					

Supplementary Table S5: Genotype and allele frequencies of indicated polymorphisms in HLA-B27 positive AS patients.

		HLA-B27(+)(%)	Control (%)		HLA-B27(+)(%)	Control (%)	Genotypic	Allelic
	Genotype	(n=671)	(n =1204)	Allele	(n =671)	(n =1204)	p value	p value
rs4552569	CC	64 (9.5)	103 (8.6)	C	436 (32.5)	735 (30.5)	0.4390	0.2131
	CT	308 (45.9)	529 (43.9)	T	906 (67.5)	1673 (69.5)		
	TT	299 (44.6)	572 (47.5)					
rs17095830	GG	9 (1.3)	11 (0.9)	G	170 (12.7)	263 (10.9)	0.2611	0.1088
	AG	152 (22.7)	241 (20.0)	A	1172 (87.3)	2145 (89.1)		
	AA	510 (76.0)	952 (79.1)					
rs10865331	AA	186 (27.7)	292 (24.3)	A	710 (52.9)	1185 (49.2)	**0.0429**	**0.0300**
	AG	338 (50.4)	601 (49.9)	G	632 (47.1)	1223 (50.8)		
	GG	147 (21.1)	311 (25.8)					
rs30187	TT	194 (28.9)	296 (24.6)	T	729 (54.3)	1184 (49.2)	**0.0078**	**0.0025**
	CT	341 (50.8)	592 (49.2)	C	613 (45.7)	1224 (50.8)		
	CC	136 (20.3)	316 (26.2)					

AS: ankylosing spondylitis; HNC: healthy normal control

Supplementary Table S6: Genotype and allele frequencies of rs4552569 and rs17095830 in AS patients with or without uveitis.

		uveitis(%)	Without (%)		uveitis (%)	Without (%)	Genotype	Allelic
	Genotype	(n =64)	(n =671)	Allele	(n =64)	(n =671)	p value	p value
	CC	3 (4.7)	65 (9.7)	C	33 (25.8)	438 (32.6)	0.2554	0.1122
rs4552569	CT	27 (42.2)	308 (45.9)	T	95 (74.2)	904 (67.4)		
	TT	34 (53.1)	298 (44.4)					
	GG	2 (3.1)	7 (1.0)	G	19 (14.8)	162 (12.1)	0.3312	0.3617
rs17095830	AG	15 (23.4)	148 (22.1)	A	109 (85.2)	1180 (87.9)		
	AA	47 (73.5)	516 (76.9)					

Supplementary Table S7: Comparison of allele frequencies of indicated polymorphismin different studies.

SNP	rs4552569	rs17095830	rs10865331	rs30187
Ref	C	G	A	T
	case/conrol	case/conrol	case/conrol	case/conrol
Previous GWAS in Caucasians	N/A	N/A	0.44/0.37[a]	0.40/0.33[a]
Dr. Gu's GWAS in Han Chinese	0.315/0.273[b]	0.117/0.095 [b]	0.54/0.48 [b]	0.55/0.53 [b]
The current study	0.320/0.305	0.123/0.109	0.541/0.492	0.536/0.492
Study in Taiwanese	0.274/0.279[c]	0.102/0.087[c]	N/A	0.580/0.529[d]
Dr Cortes *et al.* GWAS in East Asians (Chinese, Taiwanese and Koreans)	0.302/0.319[e]	0.120/0.119[e]	N/A	0.542/0.486[e]

a.Evans DM *et al*. Interaction between ERAP1 and HLA-B27 in ankylosing spondylitis implicates peptide handling in the mechanism for HLA-B27 in disease susceptibility. Nature genetics 2011, 43(8): 761-7. PubMed PMID: 21743469. Pubmed Central PMCID: 3640413.
b.Lin Z *et al.* A genome-wide association study in Han Chinese identifies new susceptibility loci for ankylosing spondylitis. Nature genetics 2012 Jan; 44(1): 73-7. PubMed PMID: 22138694.
c.Wei JC *et al*. Association study of polymorphisms rs4552569 and rs17095830 and the risk of ankylosing spondylitis in a Taiwanese population. PLoS One 2013; 8(1): e52801. PubMed PMID: 23308121. Pubmed Central PMCID: 3537770.
d.Wang CM *et al.* ERAP1 genetic variations associated with HLA-B27 interaction and disease severity of syndesmophytes formation in Taiwanese ankylosing spondylitis. Arthritis Res Ther 2012; 14(3): R125. PubMed PMID: 22632381. Pubmed Central PMCID: 3446506.
e. Cortes A *et al*. Identification of multiple risk variants for ankylosing spondylitis through high-density genotyping of immune-related loci. Nature genetics 2013 Jul; 45(7): 730-8.