POSTERS only

Genomics, genetic basis of disease and functional genomics

POS0344

RISK OF ACPA POSITIVITY BY MOTIF-BASED CLASSIFICATION OF HLA-DRB1 SHARED EPITOPE ALLELES IN RA

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Background: The shared epitope (SE) is the strongest known genetic risk factor for rheumatoid arthritis (RA) and is associated with an amino acid motif at positions 70-74 in HLA-DRB1. It is linked to anti-citrullinated protein antibody (ACPA) positivity, a specific serological marker for RA. In 2005, a new classification of HLA-DRB1 alleles was proposed, in which each allele is sorted into one of five classes (S1, S2, S3P, S3D, or X).¹ The method was subsequently validated for predicting risk of developing RA² and was described as “a major advance and a template for future studies.”³ However, there have been relatively few studies utilizing this classification.

Objectives: To characterize the prevalence of each SE genotype from the 2005 classification and their association with ACPA positivity in a cohort of individuals with RA.

Methods: Participants in FORWARD, The National Databank for Rheumatic Diseases, with RA and donated serum (collected 2010-2019) were analyzed to obtain ACPA status and high-resolution HLA-DRB1 type. Alleles were classified based on the presence (S2, S3P) or absence (S1, S3D, X; collectively L) of the SE motif, and individuals were classified by allele pair. Logistic regression models (adjusted for age, sex, and race/ethnicity) to determine risk of ACPA positivity by class were generated using L/L as the reference.

Results: Characteristics of the 855 participants at the time of sample collection are presented in Table 1. Overall, 67% of participants were SE positive and 51% were ACPA positive. Of the 1,710 total alleles, X was the most common (29%) followed by S3P (24%), S1 (18%), S2 (18%), and S3D (9%). Adjusted models showed that the risk of ACPA positivity was highest among those with the genotype S2/S3P, followed

Table 1. Characteristics of study population by SE status and allele count (mean [SD] or n [%]). Significance was assessed by Student's t-test or χ^2 test, as appropriate.

	SE negative		SE positive		P (SE- vs SE+)
	0 alleles n=286	All n=569	1 allele n=407	2 alleles n=162	
Age, years	57.4 (12.3)	58.1 (11.8)	58.0 (11.9)	58.5 (11.5)	0.42
RA duration, years	15.2 (12.8)	17.0 (13.9)	16.8 (14.1)	17.6 (13.4)	0.07
Female	259 (90.6)	500 (87.9)	359 (88.2)	141 (87.0)	0.24
White	260 (90.9)	525 (92.3)	372 (91.4)	153 (94.4)	0.49
History of smoking	117 (40.9)	223 (39.2)	166 (40.8)	57 (35.2)	0.63
RDCI, 0-9	2.4 (1.8)	2.0 (1.7)	2.1 (1.7)	1.9 (1.7)	<0.01
ACPA positive	74 (25.9)	365 (64.1)	241 (59.2)	124 (76.5)	<0.001
RF positive	84 (29.4)	333 (58.5)	219 (53.8)	114 (70.4)	<0.001
Pain VAS, 0-10	4.2 (2.7)	3.6 (2.7)	3.7 (2.6)	3.5 (2.9)	<0.01
Patient global severity, 0-10	4.0 (2.5)	3.3 (2.5)	3.4 (2.4)	3.1 (2.6)	<0.001
HAQ-II, 0-3	1.0 (0.7)	0.8 (0.6)	0.8 (0.6)	0.8 (0.6)	<0.001
csDMARD use	204 (71.3)	433 (76.1)	302 (74.2)	131 (80.9)	0.13
bDMARD use	165 (57.7)	351 (61.7)	244 (59.95)	107 (66.0)	0.26
Corticosteroid use	98 (34.3)	165 (29.0)	121 (29.7)	44 (27.2)	0.12

by S2/S2, S2/S3D, S2/X, S3P/S3D, S1/S2, S3P/S3P, S3P/X, and S1/S3P (Figure 1).

Figure 1. Risk of ACPA positivity by the motif-based classification and prevalence of SE by binary status, allele count, and class. The area within each circle is proportionate to the sample size of that group.

Conclusion: The ranking by risk of ACPA positivity among this RA cohort is similar to the order of risk for developing RA, as determined in the 2006 validation paper for this classification method. A notable trend among the classes with one SE positive allele is the consistent ranking of S3D as higher risk, X in the middle, and S1 as the lowest, suggesting a potential protective effect from S1 alleles. Future work should examine clinical associations with these allele classes, including disease progression and treatment effects.

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POS0345

CLINICOGENETIC STUDY OF FIVE NOVEL SUSCEPTIBILITY LOCI FOR TAKAYASU ARTERITIS: SUSCEPTIBILITY LOCI IN THE IL12B AND PTK2B REGION, BUT NOT THE LILRA3, DUSP22, KLHL33 REGIONS, ARE ASSOCIATED WITH VASCULAR DAMAGE IN TAKAYASU ARTERITIS

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Background: We have previously identified single nucleotide polymorphism (SNP) rs6871626 in *IL12B*, rs103294 in *LILRA3*, rs17133698 in *DUSP22*, rs2322599 in *PTK2B*, and rs1713450 in *KLHL33* as non-HLA susceptibility loci in Takayasu arteritis (TAK) [1, 2]. However, the association of these SNPs with clinical features has scarcely investigated.

Objectives: In this study, we aimed to examine how these SNPs contribute to clinical features and vascular damage in TAK.

Methods: We enrolled 99 TAK patients who were enrolled in our previous genome-wide association study (GWAS) [2]. To assess vascular damage, Takayasu Arteritis Damage Score (TADS) and Vasculitis Damage Index (VDI) were measured at the last visit before November 2020. As for organ damages, the presence or absence of aortic regurgitation (AR), hypertension, ischemic heart disease, cerebrovascular event, visual loss, end-stage renal failure, and inflammatory bowel disease were evaluated. Treatment profiles including immunosuppressive drugs and vascular interventions were also reviewed.

Results: The incidence of AR was positively associated with the risk allele of *IL12B* rs6871626 ($p=0.0052$; odds ratio (OR) 2.45, 95% confidence interval (CI) 1.27-4.73), and so was the proportion of patients who underwent aortic valve replacement ($p=0.023$; OR 3.64, 95% CI 1.08-12.24) (table 1). The incidence of hypertension was associated with the risk allele of *IL12B* rs6871626 ($p=0.049$; OR 1.82, 95% CI 0.99-3.36) and *PTK2B* rs2322599 ($p=0.044$; OR 2.52, 95% CI 0.97-6.54) (table 1). The proportion of biologic users tended to be higher in the risk genotypes of *IL12B* rs6871626 ($p=0.15$; OR 1.80, 95% CI 0.79-3.99). Regarding vascular damage, there was positive correlation between TADS and the risk allele of *IL12B* rs6871626 ($p=0.0035$; $\beta=1.35$) (Figure 1). Moreover, VDI was also positively correlated with the allele ($p=0.0054$; $\beta=0.96$) (Figure 1). No other clinicogenetic associations were observed between five SNPs and vasculitis-associated damages.

Table 1. The association of the five SNPs with aortic regurgitation and hypertension

	Aortic regurgitation		Hypertension	
	OR (95% CI)	p value	OR (95% CI)	p value
<i>IL12B</i> rs6871626	2.45 (1.27-4.73)	0.0052*	1.82 (0.99-3.36)	0.049*
<i>PTK2B</i> rs2322599	1.21 (0.51-2.86)	0.67	2.51 (0.97-6.54)	0.044*
<i>LILRA3</i> rs103294	1.16 (0.52-2.61)	0.71	1.20 (0.55-1.64)	0.64
<i>DUSP22</i> rs17133698	0.56 (0.28-1.13)	0.090	0.87 (0.46-1.63)	0.66
<i>KLHL33</i> rs1713450	0.89 (0.42-1.91)	0.77	1.48 (0.68-3.22)	0.31

SNP, single nucleotide polymorphism; OR, odds ratio; CI, confidence interval

Figure. The association of the five SNPs with VDI

Conclusion: In the present study, *IL12B* rs6871626 was closely correlated with vascular damage. We also found association between *PTK2B* rs2322599 and hypertension. There was no significant relevance between vascular damage and *LILRA3* rs103294, *DUSP22* rs17133698, or *KLHL33* rs1713450.

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POS0346

THE THERAPEUTIC EFFECTS OF OMEGA-6 PUFA (PINOLENIC ACID) ON THE TRANSCRIPTOMIC PROFILE OF ACTIVATED PERIPHERAL BLOOD MONONUCLEAR CELLS ISOLATED FROM HEALTHY CONTROLS AND RHEUMATOID ARTHRITIS PATIENTS

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Background: Globally, Rheumatoid arthritis (RA) is the most common chronic inflammatory arthritis. EULAR recommends patient education and disease modifying anti-rheumatic drugs including advanced targeted therapies for management^{1,2}. Many patients seek advice on a dietary intervention that may improve symptoms. Foods high in omega-3 polyunsaturated fatty acids (PUFAs) is one of the most common recommended based on their anti-inflammatory properties³.

⁴. Previously we showed pinolenic acid (PNLA), an omega-6 (PUFAs) found in pine nuts, reduced lipid uptake, endocytosis and migration of monocyte *in vitro*. PNLA also reduced IL-6, TNF- α , IL-1 β and PGE2 produced by lipopolysaccharide (LPS) activated PBMCs from patients with RA and healthy controls (HCs).

Objectives: We hypothesised that PNLA inhibits key inflammatory processes in synovitis as in diagram.1. We investigated the transcriptomic profile of PNLA treatment on LPS activated PBMCs isolated from HCs and RA patients.

Methods: Adult RA patients (n=6) were recruited from the Rheumatology Department of University Hospital of Wales together with 6 HCs. Blood was collected after taking signed informed consent. PBMCs were isolated by Ficoll gradient centrifugation and pre-treated with 25 μ M PNLA or vehicle and then activated with 100ng/ml LPS. RNA was extracted followed by library construction and sequencing for whole genomic transcriptome. DEGs were analysed using DESeq2 and pathway analysis was performed using Ingenuity pathway analysis. The study was approved by the Research Ethics Committee for Wales (reference no. 12/WA/0045).

Results: DEGs were selected with at least differential genes using at least 1.2* fold changes and adjusted p-value of <0.05. PNLA significantly upregulates the expression of PDK4, PAI-1 (SERPINE1), FBP1, and NDRG2. These genes have important roles in metabolic pathways. Pathway analysis predicted upstream activation of nuclear receptors peroxisome proliferator-activated receptor (PPARs) involved in the anti-inflammatory process, and inhibition of NF- κ B and STAT1, the major transcription factors of the pro-inflammatory cytokines TNF- α , IL-1, IL-6 and IFN- γ .

Conclusion: PNLA has significant effects on the regulation of metabolic and inflammatory pathways in PBMCs from RA patients and HCs. Based on these results we demonstrate that PNLA may have beneficial anti-inflammatory effects in patients with RA

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