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ORIGINAL ARTICLE

Association of *ITGAM* polymorphism with systemic lupus erythematosus: a meta-analysis

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Abstract

Background *ITGAM* is one of the major non-human leucocyte antigen that has been implicated in the pathogenesis of systemic lupus erythematosus (SLE). The association of *ITGAM* polymorphism with SLE has been reported in several studies, but with inconclusive results.

Objectives The aim of this study was to assess whether combined evidence shows the association between *ITGAM* polymorphism and SLE.

Methods *A meta-analysis was performed to survey* studies on the *ITGAM* polymorphism and SLE using comprehensive Medline search and review of the references. *A total of five published studies including 12 123 patients with SLE and 17 016 controls were involved.* Meta-odds ratios (ORs) and 95% confidence intervals (CIs) based on fixed effects models or random effects models *were* depended on Cochran's Q-statistic and I^2 values.

Results The overall ORs for the minor A-allele (OR 1.795; 95%CI 1.676–1.921), AA vs. GG (OR 3.540; 95%CI 2.771–4.522), AG vs. GG (OR 1.750; 95%CI 1.617–1.895), dominant model (OR 1.857;95%CI 1.719–2.005), recessive model (OR 3.041; 95%CI 2.384–3.878) of *ITGAM* rs1143679 were significantly increased in SLE and fixed effects models were conducted. All controls were in Hardy–Weinberg (HW) proportion. Although this meta-analysis showed significant association of rs1143683 (A vs. G, OR 1.534;95%CI 1.312–1.792), rs9888739 (T vs. C,OR 1.627;95%CI 1.380–1.918), rs1143678 (T vs. C, OR 1.543; 95%CI 1.330–1.790), rs9937837 (A vs. G, OR 0.466; 95%CI 0.227–0.957) with SLE; all of these were conducted in random effects model as heterogeneity was detected. *No significant association was detected in the analysis between rs11574637 and SLE.* No publication bias was presented.

Conclusions This meta-analysis demonstrates a significant association between *ITGAM* gene polymorphism and SLE in multiple ethnic populations.

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Keywords

ITGAM, meta-analysis, polymorphism, systemic lupus erythematosus

Conflicts of interest

None declared.

Introduction

Systemic lupus erythematosus (SLE) is a clinically heterogeneous disease in which the risk of disease is influenced by complex genetic and environmental contributions.[1] *ITGAM*, also known as CD11b or complement receptor 3 (CR3), is the newest member of this pathway to be convincingly associated with SLE.[2] The product of *ITGAM*, integrin-α_M(CD11b+), is a molecule that combines with integrin-β_2 to form a leucocyte-specific integrin. The $\alpha_M\beta_2$ -integrin is important in the adherence of neutrophils and monocytes *to stimulate* endothelium and in the phagocytosis of complement-coated particles.[3] *ITGAM*, *a CD18 subfamily* of integrin receptors that is highly expressed on requires antigen-presenting cells (APCs), *ITGAM* deficiency leads to enhanced interleukin 6 (IL-6)

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production by APCs, which subsequently promotes preferential differentiation of native T cells to T helper 17 (Th17) cells, which are a T cell lineage characterized by their production of IL-17.[4]

Although studies have suggested that *ITGAM* allele may confer susceptibility to SLE, investigations performed in different ethnic groups yielded apparently inconclusive results regarding the disease susceptibility conferred by specific polymorphism. *Han et al.*[5] *demonstrated that* rs1143679, which converts Arginine 77 to Histidine explains the association of rs9937837 or rs11574637 with SLE and best explains the *ITGAM*-SLE association. The study by Yang *et al.*[3] found that logistic regression analysis on Thai samples indicates that rs1143679 remains significant when controlling the effect of rs9888739 or rs1143678. However, both rs1143679 and rs1143683 lose significant association when analysed with each other using the logistic regression analysis. Therefore, we performed a meta-analysis of published studies to clarify whether the ITGAM rs1143679, rs1143683, rs9888739, rs1143678, rs9937837, rs11574637 single-nucleotide polymorphisms (SNPs) *confer risk for development of SLE.*

Materials and methods

Literature search

We performed an exhaustive search on studies that examined the association of the *ITGAM* polymorphism with SLE. A search of the literature was made using PubMed citation to identify available articles in which the *ITGAM* SNPs were determined in patients with SLE and control. All references cited in the retrieved articles were also reviewed to identify additional published work not indexed by PubMed. We used the following Medical Subject Heading (MeSH) terms: '*ITGAM*' and 'systemic lupus erythematosus'. All patients of these studies fulfilled the American College of Rheumatology (ACR) Criteria[6] or the American College of Rheumatology 1997 Revised Criteria for SLE.[7]

Data collection

According to the meta-analysis of observational studies in epidemiology (MOOSE) guidelines for reporting meta-analysis of observational studies,[8] the following data were extracted from included studies: first author, year of publication, country where the trial was conducted, sample origin, numbers of cases and controls and distribution of allele A/T or G/C in both case and control groups.

Statistical analysis

Evaluation of statistical associations. Allele frequencies at the *ITGAM* rs1143679, rs1143683, rs9888739, rs1143678, rs9937837, rs11574637 from each respective study were determined using the allele-counting method. A chi-square test was used to determine if observed frequencies of genotypes in controls conformed to Hardy–Weinberg (HW) expectations. We assessed the within- and between-study variation or heterogeneity by testing Cochran's Q-statistic.[9] This heterogeneity test assessed the null hypothesis that all studies were evaluating the same effect. A significant Q-statistic ($P < 0.10$) indicated heterogeneity across studies, and then the random effect model was used for meta-analysis as well as to take into account the possibility of heterogeneity between studies. Otherwise, the fixed effect model was used. Fixed effect model assumes that all of the studies are estimating the same underlying effect and considers only within-study variation. We also quantified the effect of heterogeneity using $I^2 = 100\% \times (Q-df)/Q$.[10] The I^2-statistic measures the degree of inconsistency in the studies by calculating what percentage of the total variation across studies is resulting from the heterogeneity rather than by chance.

The overall or pooled estimate of risk odd ratio (OR) was obtained using Mantel–Haenszel method in the fixed effect model[11] and using DerSimonian and Laid method in the random effect model.[12] Pooled OR in the meta-analysis was performed weighting individual OR by the inverse of their variance.

Evaluation of publication bias. Funnel plots are used to detect publication bias, but they require a range of studies of varying sizes and subjective judgment, and thus, we evaluated publication bias using Egger's linear regression test,[13] which measures funnel plot asymmetry on the natural logarithm scale of the OR.

Statistical analyses were carried out using the STATA software package v.8.2 (Stata Corporation, College Station, TX, USA).

Results

Studies included in the meta-analysis

A total of nine relevant studies concerning the *ITGAM* polymorphism and SLE were identified using PubMed.[1,3,5,14–19] Two studies were excluded as the number of allele could not be extracted.[18,19] Thus, seven studies met the inclusion criteria.[1,3,5,14–17] Of these, one study contained data on three different groups,[14] one study contained data on five different groups,[5] one study contained data on two different groups,[3] the study of Marian Suarez-Gestal *et al.*[17] contained data on 16 collections from nine European countries; we combined the data according to *countries*. In this study, we analysed these groups independently. Therefore, a total of 22 separate comparisons, consisting of 12 123 SLE patients and 17 016 controls which includes ten European, three Asian, seven North American and two Latin American, were considered in this meta-analysis.

Meta-analysis of the association between the *ITGAM* rs1143679 polymorphism and SLE

Crude ORs with 95% CIs were used to assess the strength of association between ITGAM rs1143679 polymorphism and SLE risk. The pooled ORs were performed for the allele-contrast, dominant model, recessive model and additive model, respectively. We assessed the within- and between-study variation or heterogeneity

Table 1 Selected characteristics of included studies of rs1143679 (*ITGAM*) and SLE, HWE test

Study	Country (ethnicity)	Numbers		A alleles (%)		HWE		Association	
		SLE	Control	SLE	Control	χ^2	*P*	OR	95% CI
Swapan K Nath 2008[14]	USA (EA)	1916	1902	0.172	0.104	0.416	0.5191	1.78	1.56–2.03
Swapan K Nath 2008[14]	USA (AA)	588	701	0.154	0.105	0.766	0.3814	1.55	1.23–1.96
Swapan K Nath 2008[14]	USA (Gullah)	137	134	0.201	0.108	0.149	0.6996	2.07	1.27–3.36
Shizhong Han 2009[5]	USA (EA)	738	1053	0.18	0.11	0.119	0.7297	1.73	1.43–2.10
Shizhong Han 2009[5]	USA (HA)	731	229	0.16	0.08	2.097	0.1476	2.09	1.47–2.98
Shizhong Han 2009[5]	UK (E)	445	528	0.16	0.09	0.109	0.7410	2.10	1.60–2.76
Shizhong Han 2009[5]	Mexican (LA)	389	284	0.15	0.09	0.276	0.5994	1.68	1.20–2.35
Shizhong Han 2009[5]	Colombian (SA)	205	381	0.22	0.10	0.013	0.9091	2.28	1.65–3.16
Wanling Yang 2009[3]	HongKong Chinese (A)	918	1440	0.0113	0.0051	0.039	0.8425	2.21	1.11–4.42
Wanling Yang 2009[3]	Thai (A)	278	383	0.0622	0.021	0.174	0.6763	3.10	1.61–5.98
Marian Suarez-Gestal 2009[17]	Portugal (E)	94	95	0.255	0.195	0.155	0.6934	1.418	0.872–2.305
Marian Suarez-Gestal 2009[17]	Spain (E)	525	570	0.2629	0.154	0.240	0.6244	1.959	1.585–2.421
Marian Suarez-Gestal 2009[17]	Italy (E)	293	316	0.2517	0.1724	0.056	0.8129	1.614	1.222–2.132
Marian Suarez-Gestal 2009[17]	The Netherlands (E)	104	180	0.212	0.144	1.121	0.2897	1.589	1.019–2.477
Marian Suarez-Gestal 2009[17]	Hungary (E)	95	95	0.168	0.100	0.003	0.9545	1.823	0.993–3.346
Marian Suarez-Gestal 2009[17]	Greece (E)	191	186	0.2644	0.1882	1.541	0.2145	1.551	1.098–2.190
Marian Suarez-Gestal 2009[17]	Slovakia (E)	94	93	0.117	0.108	0.998	0.3177	1.100	0.579–2.092
Marian Suarez-Gestal 2009[17]	Czech Republic (E)	101	99	0.134	0.111	1.547	0.2136	1.234	0.677–2.250
Marian Suarez-Gestal 2009[17]	Germany (E)	82	92	0.207	0.092	0.953	0.3289	2.569	1.374–4.803

CI, confidence interval; HWE, Hardy-Weinberg equilibrium; OR, odds ratios; SLE, systemic lupus erythematosus.

by testing Cochran's Q-statistic. Heterogeneity was not detected in these comparisons and therefore the ORs were obtained using the fixed-effect model. The chi-square goodness of fit is used to test if observed frequencies of genotypes in controls conformed to Hardy–Weinberg expectations (HWE). A total of four studies and 19 comparisons were considered in the *ITGAM* rs1143679 polymorphism and SLE meta-analysis (Table 1).

The overall ORs for A-allele (OR 1.795; 95%CI 1.676–1.921), AA vs. GG (OR 3.540; 95%CI 2.771–4.522), AG vs. GG (OR 1.750; 95%CI 1.617–1.895), dominant (OR 1.857; 95%CI 1.719–2.005), recessive (OR 3.041; 95%CI 2.384–3.878) of *ITGAM* rs1143679 were all significantly increased in SLE and fixed effects models were conducted. These summary estimates suggested the presence of a dose effect. In the AA vs. GG and recessive model, Wanling Yang 2009[3] HongKong Chinese was excluded (Table 2, Fig. 1). The funnel plot (not showed) and Egger's test provided no evidence of publication bias for all these Comparisons (Table 2).

Meta-analysis of the association between the other *ITGAM* SNPs and SLE

We examined the contrast of the allelic effect of A vs. G, the meta-analysis produced overall odds ratios of 1.534 (95% CI: 1.312, 1.792), 1.627 (95% CI: 1.380, 1.918), 1.543 (95% CI: 1.330, 1.790), 0.466 (95% CI: 0.227, 0.957) and 0.640 (95% CI: 0.396, 1.034) for rs1143683, rs9888739, rs1143678, rs9937837, rs11574637, respectively. Although overall ORs for these SNPs were significantly increased in SLE, except rs11574637, the Q-tests of heterogeneity were all significant and we conducted analyses using random effect models. Egger's test provided no evidence of publication bias for these comparisons (Table 2).

Discussion

Aetiology of systemic lupus erythematosus *is largely unknown, as it involves multiple* genetic and environmental factors. Evidence suggests that many genetic loci contribute to the occurrence and clinical presentation of lupus. In this study, we subjected previously published data to meta-analysis to evaluate the association between the *ITGAM* rs1143679, rs1143683, rs9888739, rs1143678, rs9937837, rs11574637 polymorphism and SLE susceptibility.

Pathogenic *ITGAM encodes* integrin alpha M (also known as CD11b, Mac-1, and complement receptor type 3). *Variants* that alter binding to ICAM-1 (intercellular adhesion molecule 1), activated by antibodies or cytokines, increase the adherence of leucocytes to endothelial cells, which promotes vascular *disease. Recent* studies have reported an allelic association between *ITGAM* and SLE, *and rs1143679 SNP has the strongest evidence of association with SLE.* This variant encodes an amino-acid change from arginine to histidine at position 77, an alteration that may modify the conformation of the protein's α1 domain, the region responsible for binding ICAM-1 (Complement C3 fragment iC3b binds to a distinct site.).[20] The result of our meta-analysis demonstrated *the association* of the minor A allele vs. G allele of *ITGAM* rs1143679 SNP with SLE (OR, 1.795; 95% CI, 1.676–1.921; z = 16.81, *P* = 0.000). Egger's test suggested the absence of publication bias

Table 2 Fixed/random-Effects OR for SLE and Heterogeneity test results for the risk Allele of ITGAM Gene Polymorphisms in relation to SLE

Polymorphism and model	Sample size		Test of association				Test of heterogeneity			Egger's test for publication bias	
	Case	Control	OR	95% CI	Z	*P*-value	Q	*P*-value	I^2	*t*	*P*
rs1143679											
A vs. G	15 834	17 502	1.795	1.676–1.921	16.81	< 0.001	18.10	0.449	0.6%	0.24	0.813
AA vs. GG	4974	5871	3.540	2.771–4.522	10.12	< 0.001	11.97	0.802	0.0%	−0.36	0.724
AG vs. GG	7664	8660	1.750	1.617–1.895	13.87	< 0.001	12.81	0.802	0.0%	0.83	0.415
Recessive model	6999	7311	3.041	2.384–3.878	8.96	< 0.001	11.77	0.814	0.0%	−0.31	0.759
Dominant model	7917	8751	1.857	1.719–2.005	15.79	< 0.001	14.80	0.676	0.0%	0.49	0.628
rs1143683 A vs. G	11 236	13 522	1.534	1.312–1.792	5.37	0.000	18.20	0.006	67.0%	0.28	0.788
rs9888739 T vs. C	24 744	38 364	1.627	1.380–1.918	5.80	0.000	44.98	0.000	82.2%	0.54	0.607
rs1143678 T vs. C	24 756	38 348	1.543	1.330–1.790	5.73	0.000	36.41	0.000	78.0%	1.24	0.253
rs9937837 A vs. G	11 648	25 030	0.466	0.227–0.957	2.08	0.037	30.93	0.000	93.5%	−1.98	0.298
rs11574637 T vs. C	11 648	25 030	0.640	0.396–1.034	1.82	0.068	9.41	0.009	78.7%	−0.57	0.669

CI, confidence interval; OR, odds ratios; SLE, systemic lupus erythematosus.

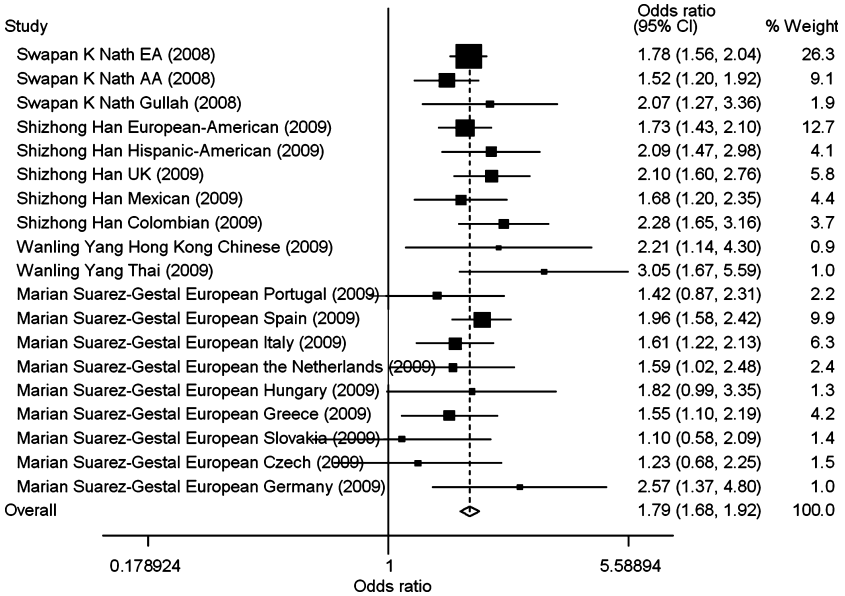

Figure 1 Meta-analysis of the association between systemic lupus erythematosus (SLE) and rs1143679 A vs. G among overall populations.

in the overall studies ($P = 0.813$). And there was no heterogeneity detected ($Q = 18.10$, $P = 0.449$). Meta-analysis was also performed for dominant model, recessive model and additive model, and the overall ORs for these genetic models were all significant. These summary estimates suggested the presence of a dose effect. *There* was no heterogeneity and publication bias in these comparisons.

Four studies[1,3,5,18] reported that SNPs that showed significant association with SLE in *ITGAM* have high linkage disequilibrium (LD) with each other. Linkage disequilibrium between these SNPs has prevented dissection of their relationship to SLE susceptibility. Han *et al.*[5] performed two-SNP haplotype analysis including rs1143679 paired with any other SNP. To exclude the possibility that multiple observed effects are caused by LD with a single true effect, pairs of SNPs were conditioned on each other, one at a time. All of the two-SNP combinations with rs1143679 showed highly significant global association. Conditional analyses demonstrated that the two-SNP global association disappeared for all sets

if *conditioned* on rs1143679, but remained significant when conditioned on the other SNP. They demonstrated that all significant associations surrounding rs1143679 arise from the high correlation between themselves and rs1143679. But Yang *et al.*[3] analysed conditioning on the effect of rs1143679, there was also a residual effect of the haplotypes, indicated association besides rs1143679. In the studies of Hom, SLEGEN[15] and Han[16] the SNPs in *ITGAM* that showed significant association with SLE were reported, *but rs1143679 was not reported. Considering* these *differences*, we also performed Meta-analysis of rs1143683, rs9888739, rs1143678, rs9937837, rs11574637 and SLE susceptibility besides rs1143679. The Q-tests of heterogeneity were all significant; random effect models were conducted and overall ORs for SNPs were significantly increased in SLE, except rs11574637 (OR, 0.640; 95%CI: 0.396–1.034).

The present study may have two caveats. First, the number of studies *was* small, which limited the ability to conduct a more detailed subgroup *analysis*. Second, significant between-study heterogeneity was detected in some comparisons and linkage disequilibrium between these SNPs were *high. The conclusions* of our *study* for the association between *ITGAM* rs1143683, rs9888739, rs1143678, rs9937837, rs11574637 polymorphism and SLE susceptibility should be cautious.

Despite of these limitations, our meta-analysis strongly demonstrated that *ITGAM* rs1143679 polymorphism was significantly associated with an increased risk of SLE in the overall population samples. This meta-analysis provides further evidence that the *ITGAM* gene plays a significant role in the aetiology of SLE. More work worldwide is needed to clarify the actual role of other SNPs of *ITGAM* in affecting the development of SLE.

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