The common *CARD14* gene missense polymorphism rs11652075 (c.C2458T/p. Arg820Trp) is associated with psoriasis: a meta-analysis

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ABSTRACT. Recent genetic evidence suggests a robust association of the *CARD14* single nucleotide polymorphism rs11652075 (c.C2458T/p. Arg820Trp) and other rare mutations in this gene with psoriasis. To assess whether combined data support the relationship between *CARD14* rs11652075 and susceptibility to this disease, we conducted a meta-analysis. PubMed (MEDLINE), EMBASE, Web of Science, and the Cochrane Library were searched for relevant papers published in English. Pooled odds ratios (ORs) and 95% confidence intervals (CIs) were calculated using random-effect models. Heterogeneity between studies was assessed using the Cochran's Q and I^2 statistics. A total of

five published studies, including 32,807 psoriasis patients and 45,458 controls, met our inclusion criteria and were included in the meta-analysis. The pooled OR of the association between the minor allele of this polymorphism and psoriasis was 0.877 (95%CI = 0.834-0.922; P < 0.001). In a stratified analysis, pooled ORs relating to European and Asian ancestry were 0.883 (95%CI = 0.822-0.948) and 0.872 (95%CI = 0.812-0.936), respectively. Those calculated for studies with case sample sizes above and below 1000 were 0.912 (95%CI = 0.870-0.956) and 0.824 (95%CI = 0.734-0.924), respectively. No publication bias was present, and the exclusion of any single dataset did not substantially alter the corresponding pooled ORs. Due to the limited data available regarding clinical classification of cases and genotypes, subgroup stratification by clinical type was not performed. Our results demonstrate a significant association between the *CARD14* rs11652075 polymorphism and psoriasis.

Key words: Psoriasis; *CARD14* gene; Polymorphism (rs11652075); Meta-analysis

INTRODUCTION

Psoriasis is a complex cutaneous disease affecting 2-5% of individuals of Western European descent and 0.1-0.3% of those of Asian ancestry (Parisi et al., 2013). Mounting evidence has demonstrated that this skin disease has a strong genetic component (Garber, 2012). Moreover, several sequence variants related to psoriasis have been identified by genome-wide association (GWAS) and deep sequencing studies (Chandran, 2013). Recently, the single nucleotide polymorphism (SNP) rs11652075 (c.C2458T/p.Arg820Trp) in caspase recruitment domain family member 14 (*CARD14*) and several other rare mutations of this gene have been reported to be associated with psoriasis in Han Chinese and Caucasian populations (Jordan et al., 2012; Tsoi et al., 2012; Tang et al., 2014).

The *CARD14* gene encodes a member of the caspase recruitment domain-containing membrane-associated guanylate kinase protein (CARMA) family that mediates activation of the nuclear factor kappa B (NF-κB) pathway (Jiang and Lin, 2012). NF-κB is a crucial factor involved in the pathophysiology of psoriasis (Goldminz et al., 2013). rs11652075 is a functional polymorphism affecting the amino acid at position 820 (p.Arg820Trp) in the C-terminal guanylate kinase domain of the CARMA2 protein (Jordan et al., 2012). As this domain plays an important role in the transmission of external signals to the intracellular milieu, this SNP may exert an important effect on protein function (Bertin et al., 2001; Jordan et al., 2012). CARMA2 proteins carrying a Trp residue at position 820 may activate the NF-κB pathway and promote the development of psoriasis (Jordan et al., 2012).

Although studies performed in different ethnic groups have suggested that certain *CARD14* rs11652075 alleles may confer susceptibility to psoriasis, not all investigations have supported this association (González-Lara et al., 2013; Qin et al., 2014), leading to inconsistent conclusions. Hence, we performed a meta-analysis of all eligible studies to derive a more precise estimation of the association between the rs11652075 SNP and risk of psoriasis.

MATERIAL AND METHODS

Literature search

Following standard guidelines for the performance of meta-analyses (Stroup et al., 2000), we conducted a systematic literature search for studies having examined the association between the *CARD14* rs11652075 polymorphism and psoriasis. Two individual investigators performed the search using PubMed (MEDLINE), EMBASE, Cochrane Library, and Web of Science databases to identify available articles published in English (up to August 10, 2014). The search strategy comprised the use of various combinations of the terms "psoriasis" and "rs11652075", "CARMA2", "*CARD14*", "caspase recruitment domain family member 14", "caspase recruitment domain-containing membrane-associated guanylate kinase protein". To be included, studies had to: A) include available allele frequency or genotype distribution data, B) use a case-control design with unrelated case-control, and C) provide sufficient published data for estimation of an odds ratio (OR) and a 95% confidence interval (CI). In addition, reference lists included in retrieved articles were manually searched to identify any other potentially relevant articles. We also contacted the authors of primary studies to retrieve missing data.

Data collection

We extracted the following data from original publications: first author, year of publication, ethnicity, total sample size, and allele and genotype frequencies in case and control groups, if available. Two investigators independently extracted data from all eligible studies according to the criteria listed above. Disagreements were resolved by discussion. When the same samples were included in several publications, only the most recent or complete study was used.

Meta-analysis methods

The strength of the association between psoriasis and the *CARD14* rs11652075 polymorphism was estimated by OR and 95%CI. Minor allele frequencies in each study were determined using the allele-counting method. Stratified analysis was conducted based on study population (Asian or European ancestry) and sample size (number of psoriasis cases > or <1000). Between-study heterogeneity was tested using Cochran's Q statistic (Egger et al., 1997), for which P values <0.10 were considered significant. A random-effect model was used when heterogeneity was detected.

Evaluation of publication bias and sensitivity analysis

Potential publication bias was evaluated by funnel plot and the Egger linear regression test. This latter was used to measure funnel plot asymmetry using a logarithmic scale of ORs. Intercept significance was determined by the *t*-test, with a P value less than 0.05 being considered to represent significant publication bias (Egger et al., 1997). We conducted sensitivity analysis to evaluate the stability of our meta-analysis results. When any single dataset was excluded,

the corresponding pooled ORs were not substantially altered, suggesting that our results are robust. All P values are two-sided, and all statistical analyses were conducted using Stata version 11.0 (StataCorp., College Station, TX, USA).

RESULTS

Study characteristics

Thirty-two papers concerning the *CARD14* gene and psoriasis were retrieved. After selection and data extraction according to our inclusion criteria, five studies were included in our analysis, comprising 32,807 cases and 45,458 controls (Jordan et al., 2012; Tsoi et al., 2012; González-Lara et al., 2013; Qin et al., 2014; Tang et al., 2014; Table 1). Data from each subgroup were treated as independent. From the five studies, a total of 17 datasets were used in the final analysis. Five subgroups from three studies involved Asian subjects (Jordan et al., 2012; Qin et al., 2014; Tang et al., 2014), and 12 subgroups from three studies concerned those of European ancestry (Jordan et al., 2012; Tsoi et al., 2012; González-Lara et al., 2013). As most studies did not categorize samples based on clinical subtype (psoriasis vulgaris or psoriasis arthritis), our analysis did not take this factor into account. Eight of the 17 datasets suggested that the rs11652075 SNP had no significant relationship with psoriasis ($P > 0.05$), while nine revealed the major C allele to be associated with psoriasis risk ($P < 0.05$). Only two studies provided genotype data, therefore it was not possible to assess the association between rs11652075 genotypes and psoriasis.

Table 1. Characteristics of studies included in the meta-analysis.

No.	First author	Year	Subgroups	MAF (%)	Cases (N)	MAF (%)	Controls (N)	P value	OR	95%CI
1	Qin	2014	Asians-Chinese	44.3	236	48.2	365	0.870	0.85	0.68-1.08
2	Tang 1	2014	Asians-Chinese	43.7	781	49.7	676	2.32E-03	0.79	0.68-0.91
3	Tang 2	2014	Asians-Chinese	46.2	9946	48.2	9906	1.10E-04	0.92	0.89-0.96
4	Tang 3	2014	Asians-Chinese	47.2	4480	49.6	6521	1.47E-04	0.91	0.86-0.96
5	González-Lara	2013	European ancestry-Spain	43.8	400	52.1	420	6.73E-04	0.71	0.59-0.87
6	Tsoi 1	2012	European ancestry-Kiel	48.0	474	49.0	1146	0.580	0.96	0.83-1.11
7	Tsoi 2	2012	European ancestry-CASP	47.0	1359	48.0	1400	0.049	0.85	0.71-1.00
8	Tsoi 3	2012	European ancestry-WTCCC2	48.0	2178	51.0	5175	7.50E-03	0.90	0.84-0.97
9	Tsoi 4	2012	European ancestry-PAGE	48.0	3580	48.0	5902	0.460	0.98	0.93-1.04
10	Tsoi 5	2012	European ancestry-GAPC	45.0	2997	50.0	9183	2.20E-08	0.84	0.79-0.90
11	Jordan 1	2012	European ancestry-Saint Louis/Dallas/UCSF	38.1	676	48.2	570	1.28E-06	0.66	0.56-0.78
12	Jordan 2	2012	European ancestry-National Psoriasis Foundation	46.4	486	44.3	154	0.524	1.09	0.84-1.42
13	Jordan 3	2012	European ancestry-Utah	45.3	931	51.5	236	0.017	0.78	0.63-0.96
14	Jordan 4	2012	European ancestry-Michigan	47.0	2768	49.2	2749	0.023	0.92	0.85-0.99
15	Jordan 5	2012	European ancestry-Newfoundland	50.3	981	48.7	483	0.546	1.07	0.87-1.31
16	Jordan 6	2012	European ancestry-Toronto	45.9	340	51.7	379	3.64E-03	0.80	0.68-0.93
17	Jordan 7	2012	Asians	32.2	194	43.1	193	2.87E-03	0.64	0.48-0.86

MAF = minor allele frequency, OR = odds ratio, CI = confidence interval.

Meta-analysis of the association between the rs11652075 polymorphism and psoriasis

A total of five studies including 17 datasets were considered in our meta-analysis. Table 2 summarizes ORs and levels of heterogeneity and publication bias calculated using the total data and subgroup analyses. We found a highly significant association between the SNP allele and psoriasis ($P < 0.001$), and the T allele has a protective significance of the disease

(OR = 0.877, 95%CI = 0.834-0.922), as shown in Table 2 and Figure 1. Subgroup analysis was conducted based on ethnicity (Asian or European ancestry) and sample size (number of psoriasis cases > or <1000). The minor T allele was significantly associated with psoriasis in the total data. Moreover, the difference in minor allele frequency between patients and controls was not substantially affected by the study population or sample size. Overall, strong evidence of an association between rs11652075 and psoriasis risk was observed in the total and subgroup analyses. As heterogeneity was detected, all of these were conducted using a random-effect model. Due to limited data concerning clinical classification of cases (psoriasis vulgaris or psoriasis arthritis) and gender, subgroup stratification based on these factors was not performed.

Table 2. Main meta-analysis results concerning the relationship between rs11652075 and psoriasis risk.

Subgroup	No. of datasets	Pooled OR (95%CI)	Test of OR = 1		Heterogeneity test			Publication bias (Egger test)	
			z	P	I^2	Chi-square	P	*t*	P
Total	17	0.877 (0.834-0.922)	5.12	0.000	76%	65.97	0.000	-1.46	0.165
Population									
Asian	5	0.872 (0.812-0.936)	3.77	0.000	63%	10.75	0.029	-1.71	0.186
European ancestry	12	0.883 (0.822-0.948)	3.43	0.001	80%	55.05	0.000	-0.57	0.580
Sample size (cases)									
≤1000	10	0.824 (0.734-0.924)	3.30	0.001	73%	32.88	0.000	-0.47	0.652
>1000	7	0.912 (0.870-0.956)	3.82	0.000	76%	24.64	0.000	0.04	0.971

OR = odds ratio, CI = confidence interval.

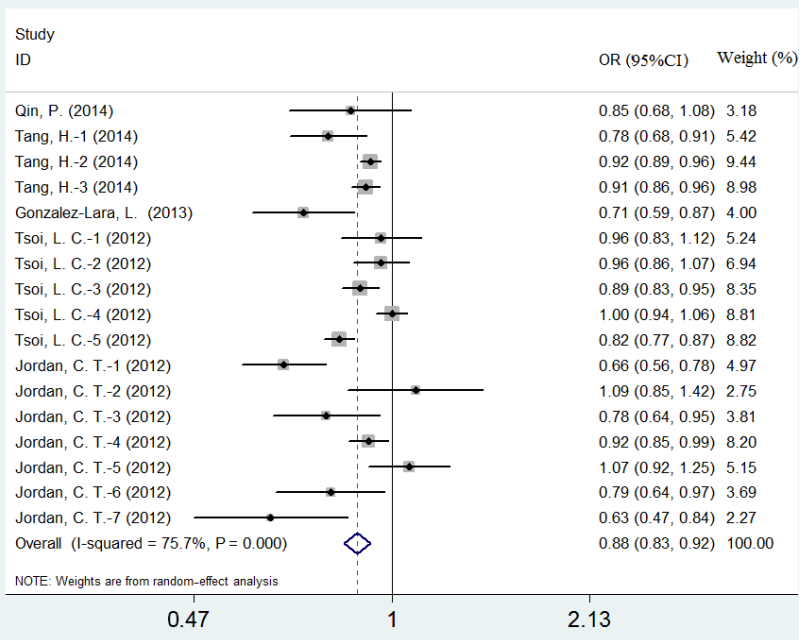

Figure 1. Forest plots of individual and pooled odds ratios with 95% confidence intervals relating to the association between rs11652075 and psoriasis risk. OR = odds ratio, CI = confidence interval.

Publication bias and sensitivity analysis

No obvious publication bias was found using a funnel plot and the Egger test (Figure 2). Moreover, we conducted sensitivity analysis to evaluate the stability of our results. When any single dataset was excluded, the corresponding pooled ORs were not substantially altered (Figure 3).

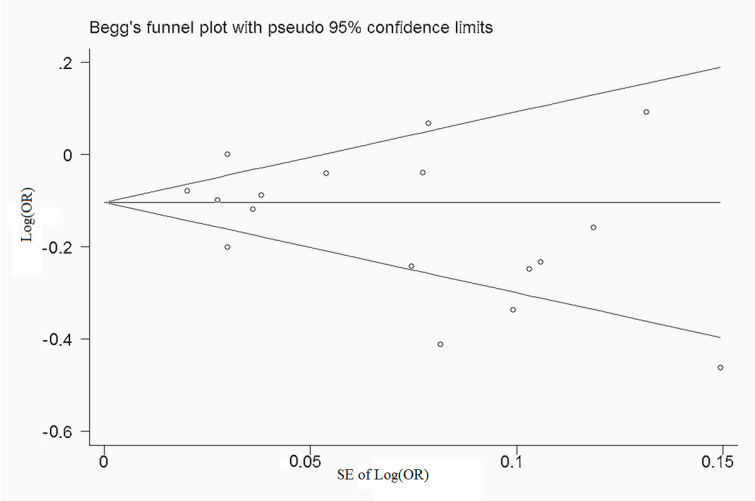

Figure 2. Begg's funnel plot testing publication bias in our meta-analysis. Each circle represents a separate study of the association between rs11652075 and psoriasis risk. SE = standard error, OR = odds ratio.

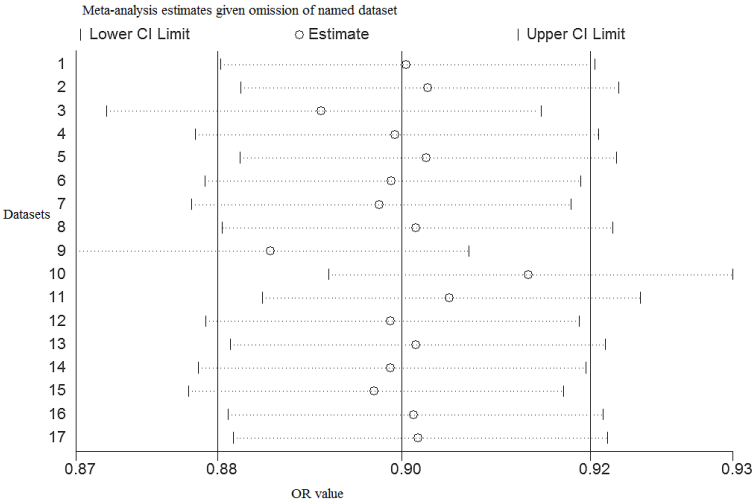

Figure 3. Influence of removing individual datasets on adjusted effect estimates. Circles represent effect estimates and horizontal dotted lines represent 95% confidence intervals from meta-analyses of the remaining datasets. The vertical line in the center shows the pooled effect estimate from all datasets. CI = confidence interval; OR = odds ratio.

DISCUSSION

In this study, we examined previously published data to evaluate the association between the *CARD14* polymorphism rs11652075 and psoriasis susceptibility by meta-analysis. Thirty-two papers concerning the *CARD14* gene and psoriasis were retrieved. By the data extraction stage, five studies (comprising 32,807 cases and 45,458 controls) were included in the analysis according to our inclusion criteria. Our results demonstrated strong evidence of an association between rs11652075 and psoriasis based on total data and subgroup analyses, without obvious publication bias. All of these were conducted using a random-effect model, as heterogeneity was detected. Sensitivity analysis revealed that pooled ORs were not substantially altered by the exclusion of individual datasets.

The *CARD14* gene encodes CARMA2, a protein primarily expressed in epithelial tissues (Scudiero et al., 2014), and a member of the CARD-containing scaffold protein family. The CARMA family is conserved across species and has three members, CARMA1, CARMA2, and CARMA3, encoded by *CARD11*, *CARD14*, and *CARD10*, respectively (Scudiero et al., 2014). CARMA2 mediates activation and recruitment of components of the NF-κB pathway (Blonska and Lin, 2011; Jiang and Lin, 2012). The NF-κB family of transcription factors plays a crucial role in cell activation, survival, and proliferation, and its aberrant activity results in immunodeficiency and autoimmune disorders such as psoriasis (Lippens et al., 2011; Jiang and Lin, 2012). *CARD14* gain-of-function mutations lead to unopposed NF-κB activation and induction of keratinocyte-derived inflammatory mediators (Jordan et al., 2012). Hence, *CARD14* mutation may result in an amplified inflammatory response upon epidermal activation, and lead more readily to lesion development (Gaffen, 2009). Recent linkage and mutation analyses have identified several missense mutations in *CARD14* in psoriasis patients. For instance, Jordan et al. (2012) have described 15 additional rare missense variants in *CARD14* and their effects on NF-κB activation and the keratinocyte transcriptome. These studies illustrate how pathogenic sequence variants are involved in pathways leading to common psoriasis.

GWAS have revealed an association between psoriasis and rs11652075 in Han Chinese individuals and people of European ancestry (Jordan et al., 2012; Tsoi et al., 2012; Tang et al., 2014). However, some investigations with small sample sizes have obtained contrasting results (González-Lara et al., 2013; Qin et al., 2014). Functional studies of this SNP have not provided further verification. However, previous study has suggested that it may exert an important effect on protein function, as rs11652075 is a functional polymorphism located in the C-terminal guanylate kinase domain of the CARMA2 protein (Jordan et al., 2012), and this region plays an important role in signal transduction (Bertin et al., 2001; Jordan et al., 2012).

Two caveats to the present investigation should be considered. First, most of the articles included in the analysis lacked information relating to clinical subtypes. Second, the included studies did not provide rs11652075 genotype frequencies. Other studies addressing the association between *CARD14* and psoriasis, whether examining the same SNP in individuals of other ethnicities (such as African populations), or the effect of other *CARD14* polymorphisms on psoriasis risk in different populations, are required. Nevertheless, our results may provide useful information for follow-up studies.

In conclusion, our meta-analysis incorporating data from five published articles suggests that rs11652075 is associated with susceptibility to psoriasis. However, due to a

current lack of data concerning clinical subtypes and genotype frequencies, more careful stratification and interaction analyses taking into account disease subgroups are needed. The pathogenicity of the mutation examined here needs to be validated further in independent cohorts and subsequent pathophysiologic and therapeutic studies.

Conflicts of interest

The authors declare no conflict of interest.

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