Four Polymorphisms of *VEGF* (+405C>G, –460T>C, –2578C>A, and –1154G>A) in Susceptibility to Psoriasis: A Meta-Analysis

Min Qi,¹ Xiaoyuan Huang,¹ Lei Zhou,² and Jianglin Zhang²

The contribution of genetic polymorphisms in the vascular endothelial growth factor (*VEGF*) gene to psoriasis risk is a controversial topic. The aim of this meta-analysis was to investigate large-scale evidence to determine the degree to which four common *VEGF* polymorphisms (+405C>G [dbSNP: rs2010963], –460T>C [dbSNP: rs833061], –2578C>A [dbSNP: rs699947], and –1154G>A [dbSNP: rs1570360]) are associated with susceptibility to psoriasis. A literature search of PubMed, Embase, Web of Science, Chinese National Knowledge Infrastructure, and Chinese Biomedical Literature Database was conducted to identify all eligible studies published before September 15, 2013. The principal outcome measure for evaluating the strength of the association was crude odds ratios (ORs) along with their corresponding 95% confidence intervals (95% CIs). Two thousand five hundred thirty-one patients and 2670 controls from nine case–control studies detailing a possible association between the *VEGF* genotypes and psoriasis risk were selected. Our meta-analysis provides evidence that two independent alleles +405G and –460C may be a protective factor for psoriasis in Asians, whereas the –1154A allele had a slight but statistically significant preventive effect on the development of psoriasis in Caucasians. The –2578C>A polymorphism, however, did not correlate with any significant difference between patients and healthy controls, even when the groups were stratified by ethnicity. Results from the meta-analysis do support the hypothesis that single-nucleotide polymorphism markers at +405C>G, –460C>T, and –1154G>A of the *VEGF* gene may serve as biological markers of psoriasis. Future studies should investigate interactions among multiple genotypes and environmental exposures to identify the role of proangiogenic markers in psoriasis and to delineate the underlying mechanisms of psoriasis.

Introduction

PSORIASIS IS A COMMON chronic inflammatory skin disorder that results from genetic predetermination in conjunction with environmental triggers; it affects ~1–4% of the general population (Henseler and Christophers, 1985; Elder, 2005). The pathology of psoriasis is characterized by an extensive inflammatory response, subsequent proliferation and differentiation of cells from the skin, and expansion and dilation of superficial dermal microvasculature (Schon *et al.*, 2005; Costa *et al.*, 2007). Angiogenesis, as a common and important component of several pathogenic mechanisms, was found to be associated with tumor growth and metastasis (Ferrara *et al.*, 1992; Barbera-Guillem *et al.*, 2002; Evensen *et al.*, 2009), inflammatory arthritis (Brenchley, 2000), atheroma formation (Ho-Tin-Noe and Michel, 2011), and psoriasis (Detmar *et al.*, 1994). Interestingly, morphological and functional microvascular changes appear to be the first-order event in the development of psoriatic lesions, suggesting that angiogenesis may be one of the key features of psoriasis pathogenesis (Ragaz and

Ackerman, 1979; Braverman and Sibley, 1982; Creamer and Barker, 1995; Creamer *et al.*, 2002). In particular, it has long been reported that the cytokines vascular endothelial growth factor (VEGF), epidermal growth factor, and fibroblast growth factors 1 and 2 (FGF1 and FGF2) are powerful mitogens and play a central role in the initiation of angiogenesis among angiogenic factors (Xu *et al.*, 2008). VEGF, also known as vascular permeability factor, has been identified as a major epidermis-derived vessel-specific growth factor that is strongly upregulated in psoriatic skin lesions (Detmar *et al.*, 1994). An increasing body of evidence for the role of VEGF in the pathogenesis of psoriasis has led to interest in the *VEGF* gene as a candidate gene for psoriasis.

The *VEGF* gene, which contains a 14-kb coding region with eight exons and seven introns, is located at chromosome 6p21.3, close to a major psoriasis susceptibility locus (Trembath *et al.*, 1997). The *VEGF* gene is reported to be highly polymorphic and more than 15 single-nucleotide polymorphisms (SNPs) have been described (Ruggiero *et al.*, 2011). Among these polymorphisms, the –634G>C SNP,

Departments of ¹Cosmetic and Plastic Surgery and ²Dermatology, Xiangya Hospital, Central South University, Changsha, China.

located within the *VEGF* promoter region, was reported to be associated with immune-mediated diseases, such as giant cell arteritis (Rueda *et al.*, 2005) and Henoch–Schönlein purpura (Rueda *et al.*, 2006). Moreover, the other four common SNPs within the promoter and 5′-untranslated regions, including +405C>G, −460T>C, −2578C>A, and −1154G>A, have even been shown to reduce or increase the levels of circulating VEGF that may be relevant to the risk of psoriasis (Watson *et al.*, 2000; Young *et al.*, 2006). Young *et al.* (2004, 2006) provided the first genetic evidence for a genetic link between −460T>C and +405C>G polymorphisms and decreased psoriasis risk in the British population. Conversely, another study by Barile *et al.* (2006) demonstrated that −460CC homozygous genotype but not +405C>G were associated with an increased risk in developing psoriasis for Italians. Yet, two more recent studies in China reported that +405C>G was significantly associated with psoriasis risk (Wang *et al.*, 2008; Wu *et al.*, 2010). Previous studies in this field have limited sample sizes and their lack of standardization makes their results difficult to generalize. Thus, despite many studies, the effects of these SNPs on psoriasis are still unclear and controversial. To the best of our knowledge, there were no published meta-analysis investigating the association between *VEGF* gene polymorphisms and psoriasis risk. Therefore, to address this issue, we carried out a quantitative meta-analysis of all studies relating VEGF gene polymorphisms and the risk of psoriasis. This meta-analysis could be useful for improving early identification of psoriasis and developing new anti-VEGF therapies for it.

Materials and Methods

Literature search

Relevant studies were searched in the PubMed, Embase, Web of Science, Chinese National Knowledge Infrastructure, and Chinese Biomedical Literature Database (last update on September 15, 2013) relating to *VEGF* gene polymorphisms and psoriasis risk. Literature searches were performed using the following MeSH terms: (“psoriasis” or “arthritis, psoriatic” or “psoriasis”) and (“polymorphism, genetic” or “single nucleotide polymorphism” or “SNP” or “mutation” or “variation”) and (“Vascular endothelial growth factor A” or “VEGF” or “VEGF-A”). We also perused the bibliographies of retrieved articles to find those that may have been missed using the computer-assisted search.

Inclusion and exclusion criteria

Studies of *VEGF* +405C>G, −460T>C, −2578C>A, and −1154G>A that qualified for inclusion should meet the following criteria: (a) case–control design, (b) presence of a quantitative assessment of the relationship between *VEGF* polymorphisms and psoriasis susceptibility, (c) studies with a 95% confidence interval (CI) for odds ratio (OR) or with sufficient data to calculate these measures, and (d) the genotype distribution of the controls conforms to Hardy–Weinberg equilibrium (HWE). Studies were excluded when they were meta-analyses, case reports, or reviews. In addition, studies with less than 30 subjects were also excluded from analyses. Furthermore, family-based studies were also excluded because of their different design settings. When data were in-

cluded in multiple studies by using the same case series in several publications, we only retained the most recently published study or the one with a largest sample size.

Data extraction

For each study, we coded information on the first author’s last name, year of publication, country of subject recruitment, subjects’ ethnicities, total number of subjects recruited or genotyped, mean age (or range whenever possible) of cases and controls, matching criteria of control groups, SNP detection method, the genetic polymorphism(s) assessed, allele/genotype frequency in case and controls if available, and evidence of HWE in controls. Ethnicity was categorized as Asian, Caucasian, African, and mixed for those studies that included subjects of more than one ethnicity. In addition, we also compared key study characteristics, such as location, study time, and authorship, to determine the existence of multiple publications from the same study. All data from eligible studies or contributed by investigators were coded independently by two investigators using a piloted data standardized form and compared afterward. In cases of conflicting evaluations, minor discrepancies were resolved by consensus with a third investigator’s careful reexamination of the full texts.

Quality assessment of included studies

The quality of eligible studies was assessed independently by two investigators based on the modified STROBE quality score system (da Costa *et al.*, 2011). Forty assessment items related to quality appraisal with scores ranging from 0 to 40. The eligible studies were classified into three levels based on their scores: low quality (0–19), moderate quality (20–29), and high quality (30–40), respectively.

Statistical analysis

Crude ORs with corresponding 95% CIs were calculated to assess the strength of the association between *VEGF* gene polymorphisms and psoriasis risk under five genetic models: the allele model, the dominant model, the recessive model, the homozygous model, and the heterozygous model. Genotype distributions in the controls were tested for HWE using the Chi-square goodness of fit test. Between-study heterogeneity was checked by the Cochran *Q*-statistic (Jackson *et al.*, 2012). The *I*² test was also used to quantify heterogeneity (ranging from 0 to 100%) (Peters *et al.*, 2006). When *p* < 0.005 for *Q*-test or *I*² > 50% indicated the existence of heterogeneity across the studies, a fixed-effect model (Mantel–Haenszel method) was used; otherwise, a random-effect model (DerSimonian–Laird method) was applied in our analysis. To explore potential sources of heterogeneity, we also performed stratification analyses by ethnicity wherever applicable. To evaluate the stability of the results, a one-way sensitivity analysis was conducted by omitting each study in turn. Funnel plot and Egger’s linear regression test were used to investigate whether publication bias might affect the validity of the estimates (Zintzaras and Ioannidis, 2005). The significance of the pooled data was determined using the *Z*-test. All *p*-values are two-tailed and *p* < 0.05 was considered as statistically significant. All the statistical tests for our meta-analysis were conducted using STATA version 12.0 (Stata Corp., College Station, TX).

Results

The characteristics of included studies

Ninety-six potentially eligible studies were retrieved through our bibliographic and manual searches. In accordance with the inclusion criteria, nine case-control studies (Young *et al.*, 2004; Barile *et al.*, 2006; Butt *et al.*, 2007; Wang *et al.*, 2008; Wongpiyabovorn *et al.*, 2008; Lu *et al.*, 2009; Wu *et al.*, 2010; Zablotna *et al.*, 2013; Xu *et al.*, 2013) with 2531 psoriasis cases and 2670 healthy controls were included in this meta-analysis and 87 were excluded. The flow diagram of the selection of studies and specific reasons for exclusion from the meta-analysis are shown in Figure 1. Overall, there were eight case-control studies on *VEGF* +405C>G polymorphism, five case-control studies on *VEGF* -460T>C polymorphism, four case-control studies on *VEGF* -2578C>A polymorphism, and three case-control studies on *VEGF* -1154G>A polymorphism. Five of the nine case-control studies were conducted in Asian populations and the remaining four studies were conducted in Caucasian populations. For genotyping of *VEGF* gene polymorphisms, seven studies used the classic polymerase chain reaction-restriction fragment length polymorphism (PCR-RELP) method, two studies used the MassArray method, and one study used the amplification refractory mutation system-PCR method. A HWE test was conducted on the genotype distributions of the controls in all included studies, and each study was coherent with the assumption of HWE (all $p > 0.05$). Characteristics of the included studies are summarized in Table 1. Table 2 summarizes the results of the combined data, depicting a plot of ORs (95% CI) for the risk of developing psoriasis associated with *VEGF*

+405C>G, -460T>C, -2578C>A, and -1154G>A polymorphisms in the nine case-control studies.

VEGF +405C>G polymorphism effect on psoriasis

For *VEGF* +405C>G polymorphism, a total of eight studies involving 2279 cases and 2335 controls were included in the pooled analysis. Since pronounced between-study heterogeneity was found in every comparison, the random-effect model was conducted. Overall, our data did not show any marked association between +405C>G polymorphism and psoriasis risk regardless of the genetic contrast (allele model: OR=0.96, 95% CI: 0.81–1.15, $p=0.664$; dominant model: OR=0.91, 95% CI: 0.68–1.21, $p=0.510$; recessive model: OR=0.91, 95% CI: 0.61–1.36, $p=0.695$; homozygous model: OR=0.80, 95% CI: 0.62–1.05, $p=0.110$; heterozygous model: OR=0.96, 95% CI: 0.65–1.43, $p=0.849$). However, when an ethnicity-stratified analysis was performed, a significantly lower prevalence of the +405G allele was observed among Asians under dominant and homozygous models (dominant model: OR=0.64, 95% CI: 0.50–0.82, $p < 0.001$; homozygous model: OR=0.58, 95% CI: 0.40–0.84, $p=0.005$) (Fig. 2), whereas similar findings were not observed in Caucasians ($p > 0.05$ for all comparisons).

VEGF -460T>C polymorphism effect on psoriasis

Five studies investigated the association between -460T>C polymorphism and psoriasis risk with a total of 698 cases and 966 controls. We fitted a random-effect model since between-study heterogeneity was anticipated. The meta-analysis results revealed that a highly significant protection was conferred by the -460C allele against overall psoriasis risk, with an

FIG. 1. Flow diagram of the selection of studies and specific reasons for exclusion from the present meta-analysis.

TABLE 1. CHARACTERISTICS OF THE STUDIES THAT EXAMINED RISK OF PSORIASIS IN RELATION TO VEGF POLYMORPHISMS

<i>Study: first author, year</i>	<i>Country</i>	<i>Ethnicity</i>	<i>Cases [n, age (years)]</i>	<i>Controls [n, age (years)]</i>	<i>Matching variables</i>	<i>Genotyping</i>	<i>SNPs</i>	<i>Mutation site</i>	<i>Quality scores</i>
Young, 2004	UK	Caucasian	137, mean 38	102, NA	Healthy	PCR-RFLP	rs2010963 rs833061	+405G>C -460T>C	30
Barile, 2006	Italy	Caucasian	117, mean 55.1 (SD 10.1)	215, mean 41.3 (SD 12.1)	Gender and ethnicity	PCR-RFLP	rs2010963 rs833061	+405G>C -460T>C	27
Butt, 2007	Canada	Caucasian	225, mean 49.7 (SD 10.9)	154, NA	Ethnicity	MassARRAY	rs2010963 rs1570360 rs699947	+405G>C -1154G>A -2578C>A	26
Wang, 2008	China	Asian	103, mean 33.2 (SD 15)	117, NA	Ethnicity	PCR-RFLP	rs2010963 rs833061 rs699947	+405G>C -460T>C -2578C>A	28
Wongpiyabovorn, 2008	Thailand	Asian	154, mean 45 (SD 44.1) mean 19 (SD 36.5)	234, mean 26 (SD 10.9)	Ethnicity	PCR-RFLP	rs1570360 rs2010963 rs833061	-1154G>A +405G>C -460T>C	29
Lu, 2009	China	Asian	1103, mean 31.9 (SD 13.1)	1104, mean 27.4 (SD 8.9)	Ethnicity	MassARRAY	rs699947 rs2010963	-2578C>A +405G>C	24
Wu, 2010	China	Asian	257, NA	258, NA	Age, gender and ethnicity	PCR-RFLP	rs2010963	+405G>C	26
Zablota, 2013	Poland	Caucasian	189, mean 41.3 (SD 20.1)	215, mean 37.9 (SD 17.6)	Ethnicity	ARMS-PCR PCR-RFLP	rs2010963 rs833061	+405G>C -460T>C	28
Xu, 2013	China	Asian	246, NA	271, NA	Age, gender and ethnicity	ARMS-PCR PCR-RFLP	rs1570360 rs699947	-1154G>A -2578C>A	23

NA, not available; PCR, polymerase chain reaction; RFLP, restriction fragment length polymorphism; ARMS, amplification refractory mutation system; SNPs, single-nucleotide polymorphisms.

TABLE 2. META-ANALYSIS OF THE ASSOCIATION BETWEEN THE VEGF POLYMORPHISMS AND PSORIASIS RISK

SNPs/ subgroups	No. of study (case/control)	2 allele vs. 1 allele (allele model)				1/2 + 2/2 vs. 1/1 (dominant model)				2/2 vs. 1/1 + 1/2 (recessive model)				2/2 vs. 1/1 (homozygous model)				2/2 vs. 1/2 (heterozygous model)			
		OR	95% CI	p	Ph	OR	95% CI	p	Ph	OR	95% CI	p	Ph	OR	95% CI	p	Ph	OR	95% CI	p	Ph
+405C>G																					
Overall	8 (2279/2335)	0.96	0.81–1.15	0.664	0.003	0.91	0.68–1.21	0.510	0.007	0.91	0.61–1.36	0.659	0.039	0.80	0.62–1.05	0.110	0.013	0.96	0.65–1.43	0.849	0.066
Asian	4 (1581/1682)	0.84	0.65–1.08	0.169	0.005	0.64	0.50–0.82	<0.001	0.698	0.76	0.54–1.07	0.115	0.566	0.58	0.40–0.84	0.005	0.805	0.93	0.65–1.33	0.674	0.449
Caucasian	4 (698/605)	1.13	0.88–1.45	0.332	0.091	1.19	0.94–1.52	0.157	0.308	1.18	0.56–2.49	0.666	0.013	1.15	0.78–1.71	0.471	0.016	1.08	0.50–2.32	0.843	0.017
–460T>C																					
Overall	5 (698/966)	0.78	0.67–0.89	0.001	<0.001	0.74	0.60–0.91	0.024	0.170	0.67	0.51–0.88	0.004	0.001	0.56	0.41–0.76	<0.001	0.001	0.75	0.57–1.00	0.052	<0.001
Asian	2 (255/435)	0.59	0.47–0.75	<0.001	<0.001	0.73	0.55–0.97	0.077	0.035	0.18	0.09–0.34	<0.001	0.001	0.20	0.10–0.38	<0.001	<0.001	0.19	0.10–0.36	<0.001	0.002
Caucasian	3 (443/531)	0.91	0.76–1.09	0.318	0.089	0.75	0.55–1.03	0.154	0.371	1.12	0.81–1.55	0.490	0.007	0.88	0.60–1.28	0.509	0.170	1.27	0.90–1.78	0.170	0.143
–2578C>A																					
Overall	4 (758/772)	0.97	0.83–1.14	0.724	0.765	0.95	0.77–1.18	0.652	0.990	0.99	0.70–1.40	0.973	0.255	1.00	0.68–1.48	0.980	0.320	1.02	0.71–1.47	0.899	0.240
Asian	3 (501/622)	1.01	0.83–1.22	0.933	0.707	0.97	0.76–1.23	0.779	0.980	1.27	0.74–2.17	0.385	0.257	1.23	0.71–2.14	0.451	0.299	1.31	0.75–2.29	0.337	0.237
Caucasian	1 (257/150)	0.90	0.67–1.19	0.449	na	0.90	0.56–1.44	0.658	na	0.84	0.54–1.31	0.442	na	0.81	0.47–1.42	0.468	na	0.85	0.53–1.38	0.514	na
–1154G>A																					
Overall	3 (546/465)	0.80	0.66–0.97	0.024	0.680	0.76	0.58–1.00	0.050	0.625	0.75	0.53–1.06	0.104	0.396	0.67	0.44–1.01	0.054	0.409	0.80	0.55–1.16	0.245	0.388
Asian	1 (100/103)	0.75	0.42–1.36	0.351	na	0.67	0.35–1.29	0.230	na	1.08	0.19–2.33	0.552	na	1.87	0.17–21.1	0.611	na	1.05	0.25–2.59	0.384	na
Caucasian	2 (446/362)	0.81	0.66–0.99	0.040	0.395	0.78	0.57–1.06	0.109	0.816	0.73	0.51–1.04	0.082	0.283	0.65	0.43–0.98	0.041	0.300	0.22	0.02–2.21	0.186	0.375

na, not applicable; 1, wild allele; 2, mutant allele; 1/1, wild homozygote; 1/2, heterozygote; 2/2, mutant homozygote; p_h , p -value of heterogeneity test; OR, odds ratio; CI, confidence interval.

FIG. 2. Forest plot of ORs for the association of *VEGF* +405C>G polymorphism with psoriasis risk in subgroup analysis based on ethnicity (A) dominant model; (B) homozygous model. CI, confidence interval; ORs, odds ratios.

~20–45% decrement in the odds (allele model: OR=0.79, 95% CI: 0.67–0.92, $p=0.002$; dominant model: OR=0.77, 95% CI: 0.61–0.97, $p=0.024$; recessive model: OR=0.67, 95% CI: 0.50–0.89, $p=0.007$; homozygous model: OR=0.57, 95% CI: 0.41–0.80, $p=0.001$). In addition, separate meta-analyses stratified by ethnicity found that the magnitude of the overall effect was similar in Asians (allele model: OR=0.59, 95% CI: 0.47–0.75, $p<0.001$; recessive model: OR=0.18, 95% CI: 0.09–0.34, $p<0.001$; homozygous model: OR=0.20, 95% CI: 0.10–0.38, $p<0.001$; heterozygous model: OR=0.19, 95% CI: 0.10–0.36, $p<0.001$) (Fig. 3), whereas the favorable trend disappeared in Caucasians ($p>0.05$ for all comparisons).

VEGF –2578C>A polymorphism effects on psoriasis

VEGF –2578C>A polymorphism was investigated in four studies with a total of 758 cases and 772 controls. Since no significant between-study heterogeneity was found in any comparison, the fixed effects model was conducted. Un-

fortunately, genotype distributions in –2578C>A polymorphism did not show significant difference between patients and controls under any genetic model (allele model: OR=0.97, 95% CI: 0.83–1.14, $p=0.724$; dominant model: OR=0.95, 95% CI: 0.77–1.18, $p=0.652$; recessive model: OR=0.99, 95% CI: 0.70–1.40, $p=0.973$; homozygous model: OR=1.00, 95% CI: 0.68–1.48, $p=0.980$; heterozygous model: OR=1.02, 95% CI: 0.71–1.47, $p=0.899$), even when the groups were stratified by ethnicity (data not shown).

VEGF –1154G>A polymorphism effects on psoriasis

Only three studies with a combination of 546 cases and 465 controls investigated the association of –1154G>A with psoriasis susceptibility. There was no evidence of significant between-study heterogeneity. The pooled analysis has shown that the –1154A allele had a slight but statistically significant preventive effect on the development of psoriasis (allele model: OR=0.80, 95% CI: 0.66–0.97,

FIG. 3. Forest plot of ORs for the association of *VEGF* -460C>T polymorphism with psoriasis risk in subgroup analysis based on ethnicity (A) allele model; (B) homozygous model.

$p=0.024$; dominant model: OR=0.76, 95% CI: 0.56–1.00, $p=0.050$). In the ethnicity-stratified analysis, this borderline statistically significant reduction in risk was found to be associated with the allele and dominant variant genotypes among Caucasians (for allele model: OR=0.81, 95% CI: 0.66–0.99, $p=0.040$; dominant model: OR=0.65, 95% CI: 0.43–0.98, $p=0.041$) but not in Asians ($p>0.05$ for all comparisons).

Sensitivity analysis and publication bias

Sensitivity analysis was also performed by excluding one study at a time to assess whether any single study had a strong influence on the pooled OR. As shown in Figure 4, sensitivity analyses indicate that no single study significantly influenced the pooled OR qualitatively (data not shown), suggesting that the studies used were statistically accurate. Furthermore, Begg's funnel plot and Egger's linear regression test were performed on the metadata to assess the publication bias of individual studies. The shapes of the funnel plots show no evidence of publication bias (Fig. 5). The Egger test also did not displayed any evidence of

publication bias (+405C>G: $t=0.46$, $p=0.664$; -460T>C: $t=-0.54$, $p=0.625$; -2578C>A: $t=0.26$, $p=0.186$; -1154G>A: $t=0.79$, $p=0.576$).

Discussion

A sustained increase in new blood vessel formation, known as cutaneous angiogenesis, can be seen in cutaneous disease, including primary and metastatic cutaneous malignant melanoma (Hubler and Wolf, 1976; Marcoval *et al.*, 1997), rosacea (Aroni *et al.*, 2008), and psoriasis (Creamer and Barker, 1995). Evidence from clinical trials suggests that VEGF, as a key cytokine in skin inflammation, is a main angiogenic mediator in psoriasis (Detmar *et al.*, 1994; Kunstfeld *et al.*, 2004). Angiogenesis-associated skin diseases have been confirmed to correlate with the magnitude of VEGF protein produced. Furthermore, high/low VEGF expression appears to be under genotypic control (Malhotra *et al.*, 1989). Thus, in recent years, the associations between SNPs of the *VEGF* gene and psoriasis risk have been extensively investigated, obtaining conflicting results. This led us to undertake the present meta-analysis, which aims to

FIG. 4. Sensitivity analysis of the summary odds ratio coefficients of *VEGF* gene polymorphisms are illustrated under the dominant model. Results were computed by omitting each study in turn. The two ends of the dotted lines represent the 95% CI (A) +405C>G; (B) -460C>T; (C) -2578C>A; (D) -1154G>A.

derive a more precise evaluation of these issues. To the best of our knowledge, this is the first meta-analysis on the topic.

This meta-analysis, involving 2279 patients and 2335 controls in a total of nine case-control studies, explored the correlation of four common *VEGF* polymorphisms with psoriasis risk. The main findings of this meta-analysis are that the two independent alleles +405G and -460C may have a correlation with decreased psoriasis risk in Asians, and the -1154A allele is associated with borderline significant decrease in psoriasis among Caucasians. It is likely that there is an ethnic difference in the *VEGF* genotype that is associated with psoriasis, which also can be showed in subcategory meta-analysis. These findings may also be biologically plausible since previous studies reported that the +405C allele is associated with increased levels of circulating VEGF and might contribute to individually increased psoriasis susceptibility (Awata *et al.*, 2002; Lambrechts *et al.*, 2003). In addition, the +405G>C locus has also been found to be associated with a great number of inflammatory or neoplastic diseases (Gentilini *et al.*, 2008; Hu *et al.*, 2012). Another two SNPs at positions -460 and -1154, located within the promoter region, have been shown to be associated with VEGF production in stimulated peripheral mononuclear cells (Shahbazi *et al.*, 2002). Based on previous reports and our analysis, we find that the -460T allele is associated not only with high VEGF production but

also with a genetic susceptibility to develop psoriasis. Finally, although the current meta-analysis indicated that the -2578C>A locus alone may be insufficient to influence susceptibility to psoriasis, even when the groups were stratified by ethnicity, a previous research suggested that the CTG (-2578/-460/+1405) haplotype is a marker for genetic susceptibility to psoriasis (Wongpiyabovorn *et al.*, 2008). It is possible that our findings are attributable to chance due to the relatively small sample size. Thus, whether these polymorphisms are truly functional requires further investigation with large-scale sample size.

Several limitations should also not be ignored when interpreting the results. First, the sample size in our study was relatively modest, especially in ethnicity subgroup analysis, so the power of the association analysis was inevitably low. Second, as with other complex traits, psoriasis is polygenic and may also be modulated by several other genetic markers beyond *VEGF*, including HLA-Cw6, TNF- α , IL23R, and several other candidate genes (Li *et al.*, 2007; Wu *et al.*, 2011; Zhu *et al.*, 2012). Thus, our meta-analysis emphasizes that elucidating the pathogenesis of psoriasis would demand further evaluation of the potential gene-gene interactions. Third, the case subjects were simply defined as psoriasis patients, with both early- and late-onset psoriasis or mild/moderate and severe psoriasis patients enrolled in few of the studies, which limited further stratified analysis based on the

FIG. 5. Begg's funnel plot of publication bias in selection of studies on *VEGF* gene polymorphisms are illustrated (A) +405C>G; (B) -460C>T; (C) -2578C>A; (D) -1154G>A. Each point represents a separate study by the indicated association. Log(OR), natural logarithm of OR. Horizontal line, mean magnitude of the effect.

age of onset and disease severity. Finally, our meta-analysis is based on unadjusted ORs estimates because the lack of potential confounders, such as age, gender, smoking status, or other factors, prevented a more precise evaluation with adjusted ORs. Aside from the limitations listed above, our meta-analysis still has some strength. To the best of our knowledge, this is the first meta-analysis on the correlation between *VEGF* polymorphisms and psoriasis.

Despite these remarks, some interesting conclusions have emerged. From the results of this quantitative meta-analysis, it appears that the *VEGF* +405C>G and -460C>T may be protective factors for psoriasis, especially in Asians. With regard to *VEGF* -1154G>A, a borderline significant decrease in risk was also found to be associated with psoriasis in Caucasians. The *VEGF* -2578C>A polymorphism, however, did not appear to have any effect on psoriasis risk, even when the groups were stratified by ethnicity. Thus, our results do support the hypothesis that the SNP markers at the +405C>G, -460C>T, and -1154G>A of the *VEGF* gene may serve as biological markers of psoriasis. Additional large and well-designed epidemiological studies on the role of proangiogenic markers in psoriasis would be required to delineate pathogenic pathways in this disease.

Disclosure Statement

All authors declare there are no conflicts of interest.

References

- Aroni, K., Tsagrioni, E., Kavantzias, N., Patsouris, E., and Ioannidis, E. (2008). A study of the pathogenesis of rosacea: how angiogenesis and mast cells may participate in a complex multifactorial process. *Arch Dermatol Res* **300**, 125–131.
- Awata, T., Inoue, K., Kurihara, S., Ohkubo, T., Watanabe, M., Inukai, K., *et al.* (2002). A common polymorphism in the 5'-untranslated region of the VEGF gene is associated with diabetic retinopathy in type 2 diabetes. *Diabetes* **51**, 1635–1639.
- Barbera-Guillem, E., Nyhus, J.K., Wolford, C.C., Friece, C.R., and Sampsel, J.W. (2002). Vascular endothelial growth factor secretion by tumor-infiltrating macrophages essentially supports tumor angiogenesis, and IgG immune complexes potentiate the process. *Cancer Res* **62**, 7042–7049.
- Barile, S., Medda, E., Nistico, L., Bordinon, V., Cordiali-Fei, P., Carducci, M., *et al.* (2006). Vascular endothelial growth factor gene polymorphisms increase the risk to develop psoriasis. *Exp Dermatol* **15**, 368–376.
- Braverman, I.M., and Sibley, J. (1982). Role of the microcirculation in the treatment and pathogenesis of psoriasis. *J Invest Dermatol* **78**, 12–17.
- Brenchley, P.E. (2000). Angiogenesis in inflammatory joint disease: a target for therapeutic intervention. *Clin Exp Immunol* **121**, 426–429.
- Butt, C., Lim, S., Greenwood, C., and Rahman, P. (2007). VEGF, FGF1, FGF2 and EGF gene polymorphisms and psoriatic arthritis. *BMC Musculoskelet Disord* **8**, 1.

- Costa, C., Incio, J., and Soares, R. (2007). Angiogenesis and chronic inflammation: cause or consequence? *Angiogenesis* **10**, 149–166.
- Creamer, D., Sullivan, D., Bicknell, R., and Barker, J. (2002). Angiogenesis in psoriasis. *Angiogenesis* **5**, 231–236.
- Creamer, J.D., and Barker, J.N. (1995). Vascular proliferation and angiogenic factors in psoriasis. *Clin Exp Dermatol* **20**, 6–9.
- da Costa, B.R., Cevallos, M., Altman, D.G., Rutjes, A.W., and Egger, M. (2011). Uses and misuses of the STROBE statement: bibliographic study. *BMJ Open* **1**, e000048.
- Detmar, M., Brown, L.F., Claffey, K.P., Yeo, K.T., Kocher, O., Jackman, R.W., *et al.* (1994). Overexpression of vascular permeability factor/vascular endothelial growth factor and its receptors in psoriasis. *J Exp Med* **180**, 1141–1146.
- Elder, J.T. (2005). Psoriasis clinical registries, genetics, and genomics. *Ann Rheum Dis* **64 Suppl 2**, ii106–ii107.
- Evensen, L., Micklem, D.R., Blois, A., Berge, S.V., Aarsaether, N., Littlewood-Evans, A., *et al.* (2009). Mural cell associated VEGF is required for organotypic vessel formation. *PLoS One* **4**, e5798.
- Ferrara, N., Houck, K., Jakeman, L., and Leung, D.W. (1992). Molecular and biological properties of the vascular endothelial growth factor family of proteins. *Endocr Rev* **13**, 18–32.
- Gentilini, D., Somigliana, E., Vigano, P., Vignali, M., Busacca, M., and Di Blasio, A.M. (2008). The vascular endothelial growth factor +405G>C polymorphism in endometriosis. *Hum Reprod (Oxford, England)* **23**, 211–215.
- Henseler, T., and Christophers, E. (1985). Psoriasis of early and late onset: characterization of two types of psoriasis vulgaris. *J Am Acad Dermatol* **13**, 450–456.
- Ho-Tin-Noe, B., and Michel, J.B. (2011). Initiation of angiogenesis in atherosclerosis: smooth muscle cells as mediators of the angiogenic response to atheroma formation. *Trends Cardiovasc Med* **21**, 183–187.
- Hu, K., Zhang, Y., Wang, R., Li, G., and Zhang, D. (2012). Current evidence on VEGF+405G/C polymorphism and malignancy susceptibility: a meta-analysis involving 30 studies. *Twin Res Hum Genet* **15**, 496–502.
- Hubler, W.R., Jr., and Wolf, J.E., Jr. (1976). Melanoma. Tumor angiogenesis and human neoplasia. *Cancer* **38**, 187–192.
- Jackson, D., White, I.R., and Riley, R.D. (2012). Quantifying the impact of between-study heterogeneity in multivariate meta-analyses. *Stat Med* **31**, 3805–3820.
- Kunstfeld, R., Hirakawa, S., Hong, Y.K., Schacht, V., Lange-Asschenfeldt, B., Velasco, P., *et al.* (2004). Induction of cutaneous delayed-type hypersensitivity reactions in VEGF-A transgenic mice results in chronic skin inflammation associated with persistent lymphatic hyperplasia. *Blood* **104**, 1048–1057.
- Lambrechts, D., Storkebaum, E., Morimoto, M., Del-Favero, J., Desmet, F., Marklund, S.L., *et al.* (2003). VEGF is a modifier of amyotrophic lateral sclerosis in mice and humans and protects motoneurons against ischemic death. *Nat Genet* **34**, 383–394.
- Li, C., Wang, G., Gao, Y., Liu, L., and Gao, T. (2007). TNF- α gene promoter -238G>A and -308G>A polymorphisms alter risk of psoriasis vulgaris: a meta-analysis. *J Invest Dermatol* **127**, 1886–1892.
- Lu, X., Tang, X.F., Quan, C., Zhou, F.S., Zheng, H.F., Sun, L.D., *et al.* (2009). [Study on association between polymorphism of VEGF gene and psoriasis susceptibility in Chinese Han population]. *Acta Univ Med Anhui* **44**, 318–321.
- Malhotra, R., Stenn, K.S., Fernandez, L.A., and Braverman, I.M. (1989). Angiogenic properties of normal and psoriatic skin associate with epidermis, not dermis. *Lab Invest* **61**, 162–165.
- Marcovall, J., Moreno, A., Graells, J., Vidal, A., Escriba, J.M., Garcia-Ramirez, M., *et al.* (1997). Angiogenesis and malignant melanoma. Angiogenesis is related to the development of vertical (tumorigenic) growth phase. *J Cutan Pathol* **24**, 212–218.
- Peters, J.L., Sutton, A.J., Jones, D.R., Abrams, K.R., and Rushton, L. (2006). Comparison of two methods to detect publication bias in meta-analysis. *JAMA* **295**, 676–680.
- Ragaz, A., and Ackerman, A.B. (1979). Evolution, maturation, and regression of lesions of psoriasis. New observations and correlation of clinical and histologic findings. *Am J Dermatopathol* **1**, 199–214.
- Rueda, B., Lopez-Nevot, M.A., Lopez-Diaz, M.J., Garcia-Porrúa, C., Martin, J., and Gonzalez-Gay, M.A. (2005). A functional variant of vascular endothelial growth factor is associated with severe ischemic complications in giant cell arteritis. *J Rheumatol* **32**, 1737–1741.
- Rueda, B., Perez-Armengol, C., Lopez-Lopez, S., Garcia-Porrúa, C., Martin, J., and Gonzalez-Gay, M.A. (2006). Association between functional haplotypes of vascular endothelial growth factor and renal complications in Henoch-Schönlein purpura. *J Rheumatol* **33**, 69–73.
- Ruggiero, D., Dalmasso, C., Nutile, T., Sorice, R., Dionisi, L., Aversano, M., *et al.* (2011). Genetics of VEGF serum variation in human isolated populations of cileto: importance of VEGF polymorphisms. *PLoS One* **6**, e16982.
- Schon, M.P., Boehncke, W.H., and Brocker, E.B. (2005). Psoriasis: clinical manifestations, pathogenesis and therapeutic perspectives. *Discov Med* **5**, 253–258.
- Shahbazi, M., Fryer, A.A., Pravica, V., Brogan, I.J., Ramsay, H.M., Hutchinson, I.V., *et al.* (2002). Vascular endothelial growth factor gene polymorphisms are associated with acute renal allograft rejection. *J Am Soc Nephrol* **13**, 260–264.
- Trembath, R.C., Clough, R.L., Rosbotham, J.L., Jones, A.B., Camp, R.D., Frodsham, A., *et al.* (1997). Identification of a major susceptibility locus on chromosome 6p and evidence for further disease loci revealed by a two stage genome-wide search in psoriasis. *Hum Mol Genet* **6**, 813–820.
- Wang, Z., Liang, W., Zhang, B., Lv, M., Wang, J., and Zhang, L. (2008). Single nucleotide polymorphisms of VEGF gene and Psoriasis risk. *J Dermatol Sci* **49**, 263–265.
- Watson, C.J., Webb, N.J., Bottomley, M.J., and Brenchley, P.E. (2000). Identification of polymorphisms within the vascular endothelial growth factor (VEGF) gene: correlation with variation in VEGF protein production. *Cytokine* **12**, 1232–1235.
- Wongpiyabovorn, J., Yooyongsatit, S., Ruchusatsawat, K., Avihingsanon, Y., and Hirankarn, N. (2008). Association of the CTG (-2578/-460/+405) haplotype within the vascular endothelial growth factor gene with early-onset psoriasis. *Tissue Antigens* **72**, 458–463.
- Wu, D., Wu, Y., Liu, J.L., Wang, B., and Zhang, X.D. (2011). Association between HLA-Cw*0602 polymorphism and psoriasis risk: a meta-analysis. *Genet Mol Res* **10**, 3109–3120.
- Wu, J., Ren, X., Zhang, X., Li, C., Li, Y., Ma, H., *et al.* (2010). The vascular endothelial growth factor +405 G/C polymorphism in psoriasis. *J Dermatol Sci* **57**, 62–63.
- Xu, H.L., Tan, H.N., Wang, F.S., and Tang, W. (2008). Research advances of endostatin and its short internal fragments. *Curr Protein Pept Sci* **9**, 275–283.
- Xu, J., Huang, C.Z., Huang, C.B., Qiao, Z.X., and Sun, W.J. (2013). Association between the single nucleotide polymor-

- phism in vascular endothelial growth factor gene and susceptibility of psoriasis. *Prog Mod Biomed* **13**, 678–680.
- Young, H.S., Summers, A.M., Bhushan, M., Brenchley, P.E., and Griffiths, C.E. (2004). Single-nucleotide polymorphisms of vascular endothelial growth factor in psoriasis of early onset. *J Invest Dermatol* **122**, 209–215.
- Young, H.S., Summers, A.M., Read, I.R., Fairhurst, D.A., Plant, D.J., Campalani, E., *et al.* (2006). Interaction between genetic control of vascular endothelial growth factor production and retinoid responsiveness in psoriasis. *J Invest Dermatol* **126**, 453–459.
- Zablotna, M., Sobjanek, M., Nedoszytko, B., Lange, M., Kozicka, D., Glen, J., *et al.* (2013). Association of psoriasis with the VEGF gene polymorphism in the northern Polish population. *J Eur Acad Dermatol Venereol* **27**, 319–323.
- Zhu, K.J., Zhu, C.Y., Shi, G., and Fan, Y.M. (2012). Association of IL23R polymorphisms with psoriasis and psoriatic arthritis: a meta-analysis. *Inflamm Res* **61**, 1149–1154.
- Zintzaras, E., and Ioannidis, J.P. (2005). Heterogeneity testing in meta-analysis of genome searches. *Genet Epidemiol* **28**, 123–137.

Address correspondence to:
Jianglin Zhang, MD
Department of Dermatology
Xiangya Hospital
Central South University
No. 87 Xiangya Road
Changsha
Hunan 410008
China

E-mail: zhangjl_csxy@126.com

Received for publication November 3, 2013; received in revised form February 14, 2014; accepted February 26, 2014.