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Toll-like receptor 2 gene polymorphisms and cancer susceptibility: A meta-analysis

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To date, epidemiological studies have assessed the association between Toll-like receptor 2 (TLR2) gene polymorphisms and cancer risk. However, the results of these studies remain controversial. We aimed to examine the associations between three SNPs (Delta22, rs3804099 and rs3804100) of TLR2 gene and cancer risk by conducting a meta-analysis of case-control studies. A total of eight studies eligible for TLR2 Delta22 polymorphism (2,061 cancer cases and 3,490 controls), six studies for rs3804099 polymorphism (1,681 cases and 1,996 controls), and five studies for rs3804100 polymorphism (3,131 cases and 2,969 controls) were included in this meta-analysis. Our results suggested that Delta22 represented a risk factor on cancers (del-allele versus ins-allele, OR=1.35, 95% CI: 1.05-1.72; del/del versus ins/ins, OR=1.91, 95% CI: 1.03-3.56; del/del + del/ins versus ins/ins, OR=1.33, 95% CI: 1.02-1.73; del/del versus del/ins + ins/ins, OR=1.79, 95% CI: 1.02-3.13), especially in Caucasian population and among population-based studies. For TLR2 rs3804099 polymorphism, we found a decreased cancer risk associated with CC/CT genotype only in Asians compared with TT genotype. TLR2 rs3804100 polymorphism was significantly associated with an elevated cancer risk in overall analysis (CC versus CT, OR=1.70, 95% CI: 1.20-2.42; CC versus CT/TT, OR=1.61, 95% CI: 1.15-2.25). In a stratified analysis, a statistically significant correlation was also observed in non-Caucasian population and population-based studies. In conclusion, the Delta22 and rs3804100 polymorphisms in TLR2 are risk factors for cancer susceptibility while the TLR2 rs3804099 dominant genotype is a protective factor, especially in Asians. Further large and well-designed studies are needed to confirm these conclusions.

Key words: TLR2, polymorphism, cancer risk, meta-analysis

Cancer is a major public health problem worldwide. It is a multistep process resulting from complex interactions between genetic and environmental factors [1]. Host genetic factors may play a critical role in the pathophysiology of many human cancers [2]. The genetic susceptibility to cancer is multifactorial, with some factors known to function in the cell cycle, apoptosis, or cell differentiation. In addition, the risk may also comprise additional factors that are related to the activation of the immune system and inflammation. Consequently, common single nucleotide polymorphisms (SNPs) of a series of low penetrance alleles, modified during the above-mentioned processes, are likely to play an important role in cancer susceptibility.

Toll-like receptors (TLRs) constitute a family of receptors directly recognizing a wide spectrum of exogenous and endogenous ligands playing the key role in realization of innate and adaptive immune response, and participating in the processes of cell proliferation, survival, apoptosis, angiogenesis, tissue remodeling and repair [3-5]. There are 10 TLRs expressed in human beings [6], and Toll-like receptor 2 (TLR2) is one of the most actively investigated TLRs. Dysregulation of the TLR2 signaling owing to SNPs may shift balance between pro- and anti-inflammatory cytokines, modulating the risk of infection, chronic inflammation and cancer. The human TLR2 gene is located on chromosome 4q32 and is composed of 2 non-coding exons and 1coding exon [7]. Genetic studies on the TLR2 gene have identified a number of polymorphisms which have been shown to affect host defense and disease progression [8, 9]. A 22-bp nucleotide deletion at position -196 to -174 of the untranslated 5'-region is associated with reduced TLR2 transcriptional activity compared to the wild type allele in luciferase reporter assays [10]. This polymorphism has already been shown to be associated with an increased risk of noncardiac gastric cancer and susceptibility to cervical cancer [11,

12]. Another 2 polymorphisms within the TLR2 exon region, namely, it has been reported, that TLR2 rs3804099 T/C and rs3804100 T/C can alter the risk of hepatocellular carcinoma development [13].

Despite a series of molecular epidemiological studies aiming to examine the association between these three polymorphisms and the susceptibility of different cancer types, the available results remain conflicting. Therefore, it is highly necessary to perform a quantitative and systematic investigation with rigorous methods. To further evaluate the association between TLR2 polymorphisms (-196 to 174 del/ins, rs3804099 and rs3804100) and the risk of cancer, a meta-analysis was conducted on all eligible published studies in current study.

Materials and methods

Literature search and data extraction. PubMed, Embase, China National Knowledge Infrastructure, and Chinese Biomedicine Database were searched (the last search update was October 20, 2012) using the following query: ('toll-like receptor 2' or 'TLR2') and ('cancer' or 'tumor' or 'neoplasm' or 'malignancy' or 'carcinoma') and 'polymorphism' by two independent investigators (XP Wang and J Li). All published papers without language restrictions matching the eligible criteria were retrieved. Additional studies were identified by a manual search of references of original or review articles on this topic. Studies included in our meta-analysis have to meet the following criteria: (a) evaluated the relationship of the TLR2 polymorphisms TLR2 -196 to 174 del/ins (Delta22) or rs3804099 or rs3804100 and cancer risk; (b) in a case-control study design; (c) contained available genotype frequency; (d) excluded benign tumors, precancerous lesions. Major reasons for exclusion of studies were (a) only case population; (b) the study did not have the outcomes of comparison reported or it was not possible to determine them; (c) duplicate of previous publication.

Data extraction. Two of the authors (XP Wang and W Xie) extracted all data independently using a standardized extraction form and reached a consensus on all items. In the present study, the following information was extracted: first author, year of publication, country, ethnicity, cancer type, numbers of cases and controls, source of control groups (population- or hospital-based controls), genotyping methods and evidence of Hardy-Weinberg equilibrium (HWE). Meanwhile, gastric cancer was regarded as an independent cancer type in Delta22 analysis; cervical cancer, gallbladder cancer, prostate cancer and hepatocellular carcinoma were merged into the "other cancers" group.

Statistical analysis. Firstly, the strength of the association between the TLR2 polymorphisms (Delta22, rs3804099 and rs3804100) and cancer risk was measured by ORs with 95% confidence intervals (CIs). The statistical significance of the OR was determined using the Z test. Statistical heterogeneity between studies was assessed with the χ^2-based Q test and I^2, heterogeneity was considered significant when $P<0.05$, and I^2 was used to quality variation in OR attributable to heterogeneity. When heterogeneity was not an issue, fixed effect model with Mantel-Haenszel method was used [14]. Otherwise, a random effect model with inverse variance method was used. Generally, we first evaluated the risks of variant allele versus wild-type allele (del-allele vs. ins-allele for Delta22; C allele vs. T allele for rs3804099 and rs3804100). As to genotype comparison, the risks of the heterozygote and variant homozygote compared with the wild-type homozygote was estimated (del/ins vs. ins/ins and del/del vs. ins/ins for Delta22; CT vs. TT and CC vs. TT for rs3804099 and rs3804100), respectively. Then we evaluated the risks of the dominant and recessive effects of the variant allele (del/del + del/ins vs. ins/ins and del/del vs. del/ins + ins/ins for Delta22; CC+CT vs. TT and CC vs. CT+TT for rs3804099 and rs3804100), respectively. In addition, we also performed stratification analyses on cancer type (divided into gastric cancer and other cancers for TLR2 Delta22) and ethnicity, as well as source of control. Publication bias was tested graphically by using funnels plots, in which the standard error was plotted against the log (OR) to form a simple scatter plot, and the funnel plot asymmetry was assessed by the method of Egger's test. Asymmetric plots could indicate potential existing publication bias. The statistical analyses were performed using Stata Statistical package (version 11.0; Stata Corp., College Station, TX) and Review Manager (Version 4.2, the Cochrane Collaboration). All P values were two-sided.

Results

Characteristics of studies. According to the searching strategy, 58 papers were found. We reviewed the titles, abstracts and the full texts of all retrieved articles through defined criteria. Finally, 15 studies including a total of 5,443 cancer cases and 6,793 controls were selected in our meta-analysis [11-13, 15-26] (Figure 1). The characteristics of the selected studies are listed in Table 1. Among these studies, Delta22 polymorphism was investigated in eight studies with 2061 cases and 3490 controls, whereas six studies were included into rs3804099 polymorphism analysis, with 1681 cases and 1996 controls and five studies into rs3804100 polymorphism analysis, with 3131 cases and 2969 controls. The distribution of genotypes in the controls was consistent with the Hardy-Weinberg equilibrium for all selected studies, except for two studies [20, 26] for rs3804099 polymorphism.

Meta-analysis. The evaluation of the association between these three polymorphisms and cancer risk is presented in Table 2. In the overall analysis, significant association could be observed between cancer risk and the variant genotypes of TLR2 Delta22 polymorphism in different genetic models (del-allele versus ins-allele, OR=1.35, 95% CI: 1.05-1.72; del/del versus ins/ins, OR=1.91, 95% CI: 1.03-3.56; dominant model, OR=1.33, 95% CI: 1.02-1.73; recessive model, OR=1.79, 95% CI: 1.02-3.13) (Table 2 and Figure 2). In a stratified analysis by specific cancer type, no association was found among studies of gastric cancer in all genetic models. However, a significant

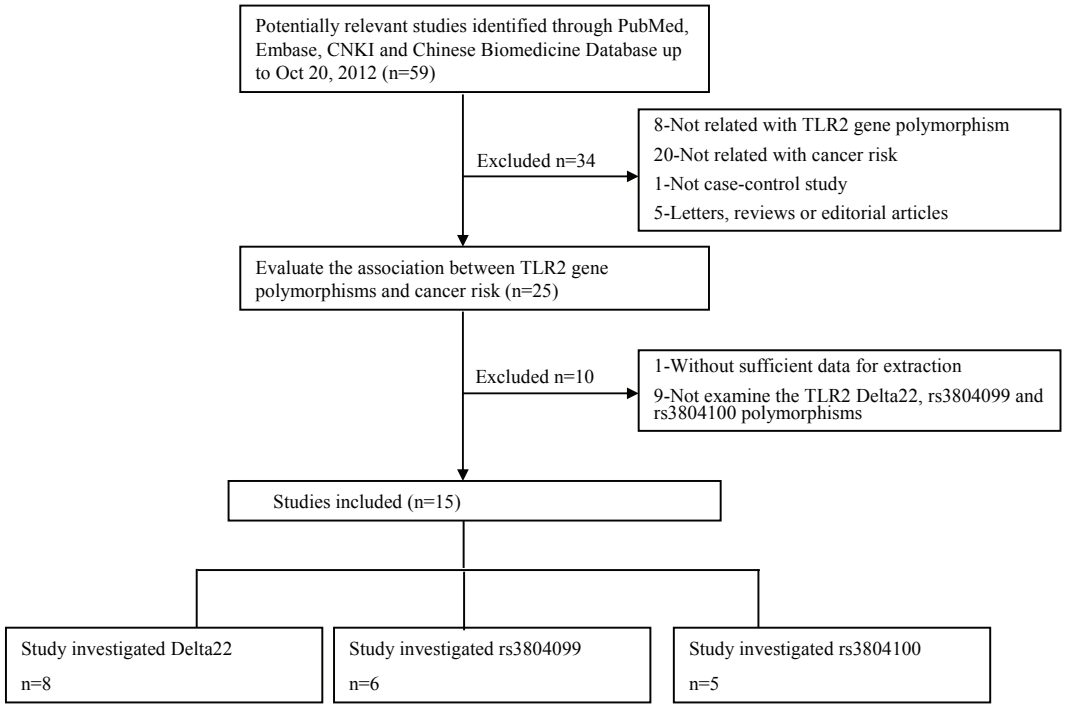

Figure 1. Study flow chart for the process of selecting the final 15 publications.

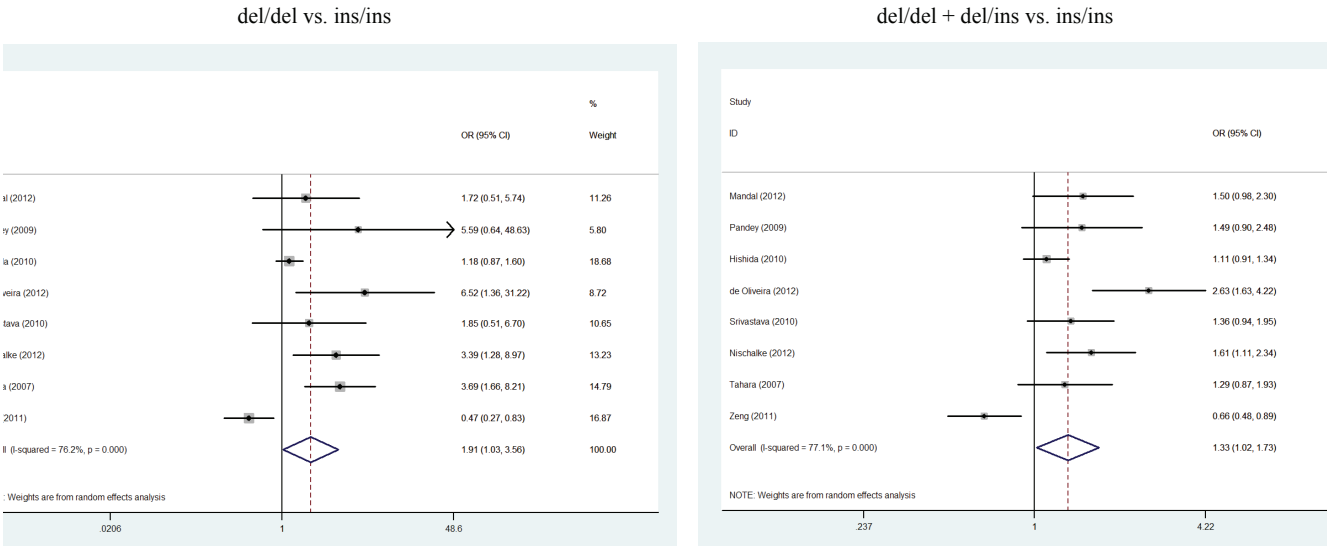

Figure 2. Forest plot of overall cancer risk associated with TLR2 -196 to 174 del (Delta 22) polymorphism for homozygote comparison (del/del vs. ins/ins) and dominant genetic model (del/del + del/ins vs. ins/ins).

association was found in other cancer types. In the subgroup analysis by ethnicity, Delta22 polymorphism had a significant increased risk of cancer in Caucasian population in all genetic models (del-allele versus ins-allele, OR=1.97, 95% CI: 1.27-3.08; del/del versus ins/ins, OR=4.07, 95% CI: 1.78-9.29; del/del versus del/ins, OR=2.41, 95% CI: 1.03-5.63; dominant model, OR=2.01, 95% CI: 1.25-3.23; recessive model, OR=3.53, 95% CI: 1.55-8.01) (Table 2), but not in Asian population. Meanwhile, in the stratified analysis by source of controls, we found elevated risk among studies from population-based controls in all genetic models.

For rs3804099 polymorphism, in the overall analysis, no association was found in any genetic models (data not shown). In the stratified analysis by ethnicity, we found a decreased

Table 1. Characteristics of literatures included in the meta-analysis between TLR-2 gene polymorphisms and cancer risk

First author	Year	Country	Ethnicity	Source of cases	Source of controls	Matching criteria	Genotyping methods	Cancer type	HWE	Cases/controls	TLR-2 Gene
Tahara	2007	Japan	Asian	289 patients who were diagnosed with histologically confirmed gastric cancer were enrolled	HB	NA	Allele-specific PCR	Gastric cancer	YES	289/146	-196 to -174 del
Etokebe	2009	Croatia	Caucasian	130 breast cancer patients with radiologically and biochemically diagnosis were selected	HB	Age, sex	TaqMan/PCR	Breast cancer	YES YES	89/89 89/89	rs3804099 rs3804100
Pandey	2009	India	Asian	150 patients diagnosed with histopathologically confirmed cervical cancer were included	NA	Age, ethnicity	PCR-RFLP	Cervical cancer	YES	150/150	-196 to -174 del
Purdue	2009	USA	Mixed	A total of 1961 histologically confirmed non-Hodgkin lymphoma patients were enrolled	PB	Age	Oligonucleotide ligase assay	Non-Hodgkin lymphoma	YES	1942/1798	rs3804100
Hishida	2010	Japan	Asian	583 patients with histologically diagnosed gastric cancer were enrolled	HB	Age, sex	PCR	Gastric cancer	YES	583/1636	-196 to -174 del
Srivastava	2010	India	Asian	233 individuals with histopathologically confirmed gallbladder cancer were enrolled	PB	Age, sex	PCR-RFLP	Gallbladder cancer	YES	233/257	-196 to -174 del
Gast	2011	Germany	Caucasian	763 patients with histologically confirmed cutaneous melanoma were enrolled	HB	NA	Sequenom Massarray	Malignant melanoma	YES YES	761/729 700/667	rs3804099 rs3804100
Miedema	2011	Netherlands	Caucasian	192 patients diagnosed with pathologically confirmed ALL were enrolled	HB	NA	Allele-Specific PCR	ALL	NO YES	182/178 189/183	rs3804099 rs3804100
Zeng	2011	China	Asian	248 patients with histological confirmation of gastric cancer diagnosis were enrolled	HB	Age, sex	PCR-RFLP	Gastric cancer	YES	248/496	rs3804099
Zeng-2	2011	China	Asian	248 patients with pathologically confirmed gastric cancer were enrolled	HB	Age, sex	PCR-RFLP	Gastric cancer	YES	248/496	-196 to -174 del
de Oliveira	2012	Brazil	Caucasian	174 patients with pathologically confirmed gastric cancer were enrolled	PB	Age	Allele-specific PCR	Gastric cancer	YES	174/208	-196 to -174 del
Mandal	2012	India	Asian	195 age matched patients with histological confirmation of prostate cancer diagnosis were enrolled	HB	Age, ethnicity	PCR	Prostate cancer	YES	195/250	-196 to -174 del
Nischalke	2012	Germany	Caucasian	197 patients diagnosed with pathologically confirmed HCV-associated HCC were enrolled	PB	Age, sex	Real-time PCR	Hepatocellular carcinoma	YES	189/347	-196 to -174 del
Pimentel-Nunes	2012	Portugal	Caucasian	193 patients with pathologically conformed colorectal cancer were enrolled	HB	Gender	PCR-RFLP	colorectal cancer	NO	190/272	rs3804099
Junjie	2012	China	Asian	211 patients diagnosed with pathologically, radiologically and biochemically diagnosis confirmed HCC were enrolled	PB	Ethnicity	SNaPshot	Hepatocellular carcinoma	YES YES	211/232 211/232	rs3804099 rs3804100

Abbreviations: PCR-RFLP, polymerase chain reaction-restriction fragments length polymorphism; PB, population-based; HB, hospital-based; HWE: Hardy-Weinberg equilibrium of controls; NA, not applicable. ALL, acute lymphoblastic leukemia; HCC, hepatocellular carcinoma; HCV, hepatitis C virus;

Table 2. Stratified analyses of the TLR2 gene polymorphisms and cancer risk

Polymorphisms	Variables	N[a]	Cases/controls	D-allele versus I-allele		D/D versus I/I		D/D versus D/I		DD/DI versus II (dominant)		DD versus DI/II (recessive)	
				OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b
-196 to -174 del (Delta22)	Total	8	2061/3490	1.35 (1.05-1.72)[c,d]	0.000	1.91 (1.03-3.56)[c,d]	0.000	1.50 (0.94-2.39)[c]	0.026	1.33 (1.02-1.73)[c,d]	0.000	1.79 (1.02-3.13)[c,d]	0.001
	Cancer Types												
	Gastric cancer	4	1294/2486	1.27 (0.84-1..93)[c]	0.000	1.60 (0.66-3.90)[c]	0.000	1.42 (0.72-2.81)[c]	0.005	1.22 (0.77-1.92)[c]	0.000	1.57 (0.71-3.47)[c]	0.000
	Other types*	4	767/1004	1.45 (1.22-1.73)[d]	0.795	2.55 (1.37-4.77)[d]	0.685	1.78 (0.94-3.37)	0.705	1.48 (1.21-1.82)[d]	0.935	2.29 (1.23-4.26)[d]	0.686
	Ethnicity												
	Asian	6	1698/2935	1.19 (0.93-1.51)[c]	0.000	1.49 (0.77-2.90)[c]	0.001	1.35 (0.79-2.30)[c]	0.022	1.16 (0.90-1.49)[c]	0.006	1.45 (0.80-2.64)[c]	0.003
	Caucasian	2	363/555	1.97 (1.27-3.08)[c,d]	0.093	4.07 (1.78-9.29)[d]	0.484	2.41 (1.03-5.63)[d]	0.860	2.01 (1.25-3.23)[d]	0.114	3.53 (1.55-8.01)[d]	0.532
	Source of controls												
	Population-based	3	596/812	1.69 (1.18-2.42)[c,d]	0.042	3.23 (1.61-6.48)[d]	0.471	2.04 (1.00-4.15)[d]	0.773	1.74 (1.22-2.49)[c,d]	0.093	2.83 (1.42-5.64)[d]	0.513
	Hospital-based	4	1315/2528	1.12 (0.82-1.53)[c]	0.000	1.29 (0.60-2.79)[c]	0.000	1.27 (0.68-2.37)[c]	0.009	1.07 (0.77-1.49)[c]	0.005	1.30 (0.65-2.59)[c]	0.001
rs-3804099 (T>C)	Variables	N[a]	Cases/controls	C-allele versus T-allele		CC versus TT		CC versus CT		CC/CT versus TT (dominant)		CC versus CT/TT (recessive)	
				OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b
	Total	6	1681/1996	1.03 (0.84-1.27)[c]	0.013	1.09 (0.87-1.35)	0.171	1.26 (0.91-1.74)[c]	0.056	0.90 (0.66-1.22)[c]	0.022	0.96 (0.62-1.48)[c]	0.000
	Ethnicity												
	Asian	2	459/728	0.99 (0.55-1.76)[c]	0.002	0.69 (0.44-1.09)	0.277	1.31 (0.54-3.16)[c]	0.014	0.68 (0.51-0.90)[d]	0.939	1.15 (0.46-2.84)[c]	0.008
	Caucasian	4	1222/1268	1.21 (0.99-1.27)	0.682	1.26 (0.97-1.62)	0.813	1.12 (0.88-1.41)	0.618	1.15 (0.94-1.40)	0.353	0.84 (0.46-1.56)[c]	0.002
rs-3804100 (T>C)	Variables	N[a]	Cases/controls	C-allele versus T-allele		CC versus TT		CC versus CT		CC/CT versus TT (dominant)		CC versus CT/TT (recessive)	
				OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b	OR (95% CI)	P^b
	Total	5	3131/2969	1.13 (0.99-1.29)	0.680	1.05 (0.60-1.83)	0.815	1.70 (1.20-2.42)[d]	0.482	1.07 (0.92-1.24)	0.835	1.61 (1.15-2.25)[d]	0.634
	Ethnicity												
	Caucasian	3	978/939	1.04 (0.80-1.36)	0.853	0.96 (0.22-4.23)	0.416	0.94 (0.21-4.24)	0.406	1.04 (0.79-1.38)	0.865	0.96 (0.22-4.22)	0.414
	Non-Caucasian	2	2153/2030	1.18 (0.97-1.45)	0.222	1.07 (0.59-1.94)	0.616	1.76 (1.23-2.53)[d]	0.291	1.08 (0.90-1.29)	0.287	1.65 (1.17-2.34)[d]	0.471
	Source of controls												
	Population-based	2	2153/2030	1.16 (1.00-1.35)	0.222	1.07 (0.59-1.94)	0.616	1.76 (1.23-2.53)[d]	0.291	1.08 (0.90-1.29)	0.287	1.65 (1.17-2.34)[d]	0.471
	Hospital-based	3	978/939	1.04 (0.80-1.36)	0.853	0.96 (0.22-4.23)	0.416	0.94 (0.21-4.24)	0.406	1.04 (0.79-1.38)	0.865	0.96 (0.22-4.22)	0.414

[a]Number of comparisons. [b]*P* value of *Q*-test for heterogeneity test. [c]Random-effects model was used when *P* value for heterogeneity test ≤ 0.10; otherwise, fixed-effects model was used; [d]There was significance between compared model and cancer risk. * Cervical cancer, gallbladder cancer, prostate cancer and hepatocellular carcinoma were included.

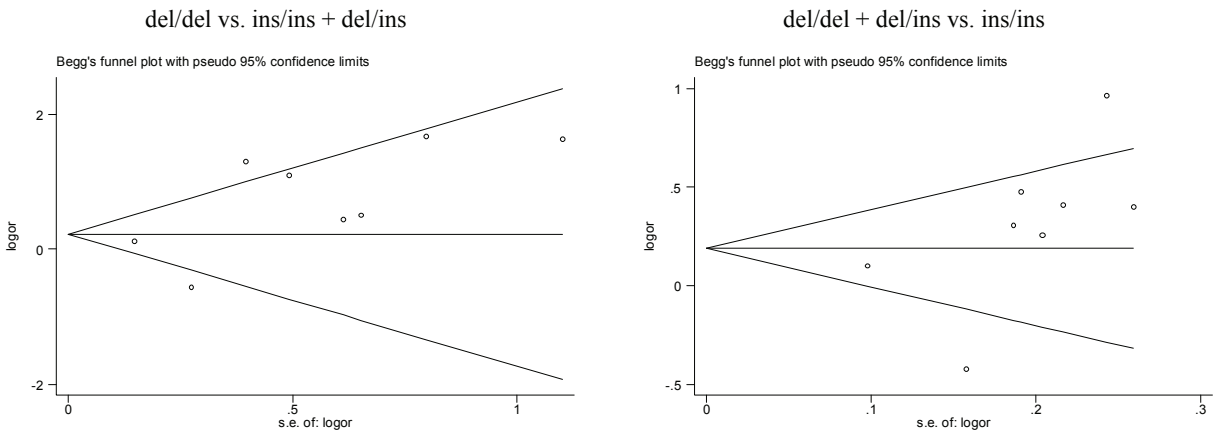

Figure 3. Begg's funnel plots to examine publication bias for reported comparisons of TLR2 -196 to 174 del (Delta 22) polymorphism. Plots are shown with pseudo 95% confidence limits. S.E., standard error. Each point represents a separate study for the indicated association.

cancer risk associated with CC/CT genotype only in Asians compared with TT genotype (OR=0.68, 95% CI: 0.51-0.90), but not in Caucasian population (Table 2). Six studies reported rs3804099 polymorphism in the overall analysis, only one for population-based controls and the other five for hospital-based. Source of controls subgroup analysis was not performed due to limited studies.

For rs3804100 polymorphism, the overall analysis showed significant increased risk in heterozygous comparison (CC versus CT, OR=1.70, 95% CI: 1.20-2.42). This association was observed in recessive model (OR=1.61, 95% CI: 1.15-2.25) (Table 2). When evaluating the effect of the polymorphism by different source of control, we found that rs3804100 significantly increased cancer risk in heterozygous comparison and recessive model among population-based studies. Because of the inadequate sample populations available for Asian and mixed groups, different ethnicities were categorized as Caucasian and non-Caucasian. We also performed sub-analysis stratified by ethnicity and found that rs3804100 polymorphism increased the cancer risk in non-Caucasians.

Sensitivity analyses and publication bias. Sensitivity analysis excluding two studies [20, 26] (for rs3804099 polymorphism) that did not reach HWE in control group, the estimated pooled OR still did not change the results appreciably, indicated that the final results of this meta-analysis were relatively stable and reliable. Begg's and Egger's test were conducted to evaluate publication bias. These different test methods have come to the same conclusion. Both of them revealed no statistical significance for publication bias in all comparison models in this meta-analysis, and the results were shown in Figure 3 and Table 3.

Discussion

Toll-like receptors (TLRs) are involved in innate immunity defence against microorganisms. The ability to properly re-

Table 3 The results of Begg's and Egger's test for publication bias

Polymorphism	Comparison type	Begg's test		Egger's test	
		Z value	P value	t value	P value
TLR2 Delta22	del vs. ins	1.61	0.108	1.93	0.102
	del/del vs. ins/ins	0.87	0.386	1.70	0.139
	del/del vs. del/ins	0.87	0.386	1.60	0.162
	del/del +del/ins vs. ins/ins	1.61	0.108	1.57	0.168
	del/del vs. del/ins +ins/ins	0.87	0.386	1.82	0.118
TLR2 rs3804099	C vs. T	0.24	0.806	-0.41	0.712
	CC vs. TT	0.24	0.806	-1.22	0.311
	CC vs. CT	-0.24	1.000	0.11	0.918
	CC/CT vs. TT	0.73	0.462	-1.09	0.355
	CC vs. CT/TT	1.50	0.133	-1.28	0.269
TLR2 rs3804100	C vs. T	0.24	0.806	0.36	0.746
	CC vs. TT	0.34	0.734	0.33	0.771
	CC vs. CT	0.34	0.734	-0.93	0.449
	CC/CT vs. TT	0.24	0.806	-0.83	0.469
	CC vs. CT/TT	0.34	0.734	-0.84	0.490

spond to TLR ligands may be impaired by SNPs within TLR genes, causing an altered susceptibility to disease. Aberrant signaling in TLR pathway activation is involved in the pathogenesis of autoimmune, chronic inflammatory, infectious diseases [6, 27], and even more seriously involved in cancers. Among TLRs, TLR2 is a central player in response to infection by Gram-positive bacteria and thus an important candidate inflammatory gene. The importance of TLR2 polymorphisms' impact on multiple types of cancer has been a concern in recent years; the most intensively concerned ones are Delta22, rs3804099 and rs3804100, however, the existing data were contradictory. Hence, it is necessary to provide a quantitative approach for combining the results of various studies, and for estimating and explaining their diversity. To the best of our knowledge, this is the first meta-analysis associating TLR2 polymorphisms with cancer risk.

We specifically conducted a systematic search of the literatures and combined the available results concerning on these three potentially functional TLR2 polymorphisms for elucidating genetic factors in cancer. Finally, the present meta-analysis included a total 5,443 cancer cases and 6,793 controls from 15 case-control studies. In the overall analysis, significant association was observed between cancer risk and the variant genotypes of TLR2 Delta22 and rs3804100 polymorphism in different genetic models, however, no association was found in any genetic models of rs3804099 polymorphism. Despite a noteworthy risk effect, the association studies on Delta22 and rs3804100 polymorphism and cancer risk were relatively limited (8 studies for Delta22; 5 studies for rs3804100). In the future study, Delta22 and rs3804100 should be paid far more attention as a most interesting TLR2 polymorphism.

As to the cancer-subgroup analyses, our results suggested that Delta22 was a risk effect on other cancer types but not on gastric cancer. The role of the TLR2 Delta22 polymorphisms in genetic susceptibility to gastric cancer has been intensively investigated. Unexpectedly, data obtained from these studies [12, 17, 21, 23] are contradictory. According to the investigation of Noguchi et al. [10], the del allele of the indicated SNP reduced the transcriptional activity of the TLR2 gene, and, because the TLR2-mediated immune response is important for the response against Helicobacter pylori (HP) infection [28], it may be associated with increased risk of such infection and with severe HP-related disease. Thus, it is interesting that this polymorphism was not associated with the risk of HP infection. Possible explanations may include the following: (i) a different prevalence of HP infection in study samples can occur even in the same population if it is large, like the Japanese population (Delta22 may be associated with higher cancer risk only or more strongly in the presence of HP because it correlates with the outcome of HP infection but does not correlate with the risk of such infection); (ii) Tahara et al. [12] recruited patients with noncardia gastric cancer only, and Hishida et al. [17] recruited cases with all types of gastric cancer, which could lead to discrepancies in the results of these 2 studies because the role of HP infection in the etiology of cardia gastric cancer is less significant than in the case with noncardia gastric cancer, according to many studies [29, 30]; and (iii) chance or any unknown bacterial, host, or environmental factor. It is also possible that this SNP is associated with the risk of gastric cancer independently of HP infection, which is partially proven by the results of Tahara et al. [12], and other bacteria or endogenous ligands (possibly damage associated molecular patterns such as heat shock proteins or extracellular matrix fragments) may also play a role in a correlation with gastric cancer risk. In addition, the del allele of the Delta22 polymorphism correlated with a higher gallbladder cancer risk in the study of Srivastava et al. [18] and with increased cervical cancer risk in the study of Pandey et al. [11]. This SNP seems to be significantly associated with the risk of many cancer types, which was partially consistent with our present analysis results. However, what plays a major role in this risk-modulating effect/impairment of TLR2-mediated immune response to pathogen-associated molecular patterns or certain alterations in TLR2-endogenous ligands interaction is unclear.

Different ethnicities may have different genetic backgrounds, which influence the association between polymorphism and cancer susceptibility. We also conducted stratified analysis by ethnicity, positive relationship between Delta22 and cancer risk was found in Caucasian population. Meanwhile, we found rs3804100 polymorphism increased cancer risk in non-Caucasian population. However, the sub-analysis of rs3804099 revealed a decreased risk of cancer in Asian population. There are significant geographic and ethnic differences in the intercontinental incidence of Delta22, rs3804099 and rs3804100 polymorphisms. The ethnic differences in the allele frequencies may be a result of natural selection, or balance to other related genetic variants. So, further investigations are warranted to validate ethnic difference in the effect of these functional polymorphisms on cancer risk especially in Asians and Caucasians in the future.

Our results indicated that all genetic models of Delta22 and CC versus CT, as well as CC versus CT/TT genotypes of rs3804100 increased cancer risk among population-based studies but not among hospital-based studies. This reason may be that hospital-based studies have a high risk of producing unreliable results because hospital-based controls may not always be truly representative of the general population. Therefore, a methodologically preferable design, such as using a proper and representative population-based study, is crucial to avoid selection bias.

Some limitations of the present meta-analysis should be addressed. First, the controls were not uniformly defined. Both healthy individuals and patients without cancers in hospital were included in the control group. Thus, the controls may not always be truly representative in the underlying source populations, especially when the polymorphism is also expected to affect the risk of other diseases. Second, the lack of the original data on genotypes and environmental risk factors of the included studies limited our evaluation on the potential gene–environment interaction. Third, our meta-analysis was

based on unadjusted estimates, adjusted ORs for age and sex ought to be pooled to provide exact summary estimates if more individual data of studies are available.

In conclusion, this meta-analysis provided evidence of the association between the TLR2 Delta22, rs3804099 and rs3804100 polymorphisms and cancer risk, especially in Caucasians and Asians. However, further large, well-designed, comprehensive studies in various populations; with more detailed individual data are needed to confirm our results. Moreover, to have a better, in-depth understanding of the association between TLR2 polymorphisms and cancer risk, sophisticated gene-gene and gene-environment interactions should also be considered in future analysis.

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