

TLR4 polymorphism and periodontitis susceptibility

A meta-analysis

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Abstract

Background: Many primary and secondary studies reported the association between Toll-like receptor 4 (TLR4) polymorphism and periodontitis susceptibility, which mainly focused on TLR4-299A>G or TLR4-399C>T of Caucasian, however, these studies had different conclusions. The aim of this study was to reassess relative studies about TLR4 polymorphism and periodontitis susceptibility, and update meta-analysis.

Methods: We searched the electronic database including CNKI (Chinese National Knowledge Infrastructure), PubMed, Embase, and hand searched relative studies until January 4, 2016. Two authors selected studies according to inclusion and exclusion criteria, assessed studies using Newcastle-Ottawa Scale case control study (NOS), and calculated the combined effect size using STATA software, version 12.0.

Results: This meta-analysis included 18 studies, containing 2453 healthy participants and 2987 patients with chronic periodontitis (CP) and 462 patients with aggressive periodontitis (AP). There was a significance between TLR4C>G (rs7873784) allele and CP in Asian, and its recessive model was also significant (for C vs G: odds ratio [OR]=0.72, 95% confidence interval [CI]=0.54–0.95, $I^2=0\%$; for CC+CG vs GG: OR=0.66, 95% CI=0.49–0.89, $I^2=0\%$). However, we did not detect any significant relevance between other TLR4 polymorphism and periodontitis susceptibility in overall and subgroup analyses. The sensitive analysis showed that dropping any single studies did not affect the pooled-analysis results. Publication bias was not detected.

Conclusions: The meta-analysis found association between TLR4C>G (rs7873784) allele and CP in Asian and it may passed on to offsprings in the form of recessiveness. However, further studies about the association between TLR4C>G (rs7873784) and CP is warranted to confirm.

Abbreviations: AP = aggressive periodontitis, CI = confidence interval, CNKI = Chinese National Knowledge Infrastructure, CP = chronic periodontitis, HWE = Hardy–Weinberg equilibrium, NOS = Newcastle-Ottawa Scale case control study, OR = odds ratio, TLR4 = Toll-like receptor 4.

Keywords: meta-analysis, periodontal disease, periodontitis, polymorphism, TLR4

1. Introduction

Periodontitis is a kind of chronic disease affected by multiple factors such as microorganism, host and environment.^[1] Periodontitis was identified into 3 types: chronic periodontitis (CP), aggressive periodontitis (AP), and periodontitis as a

manifestation of systemic disease.^[2] Kassebaum et al^[3] predicted an increasing global burden of severe periodontitis on account of growing life expectancy resulted in decreased tooth loss and increasing world population during 1990 to 2010. Susin et al^[4] summarized the features of epidemiology and demographics in AP which clarified the prevalence of AP varied significantly in

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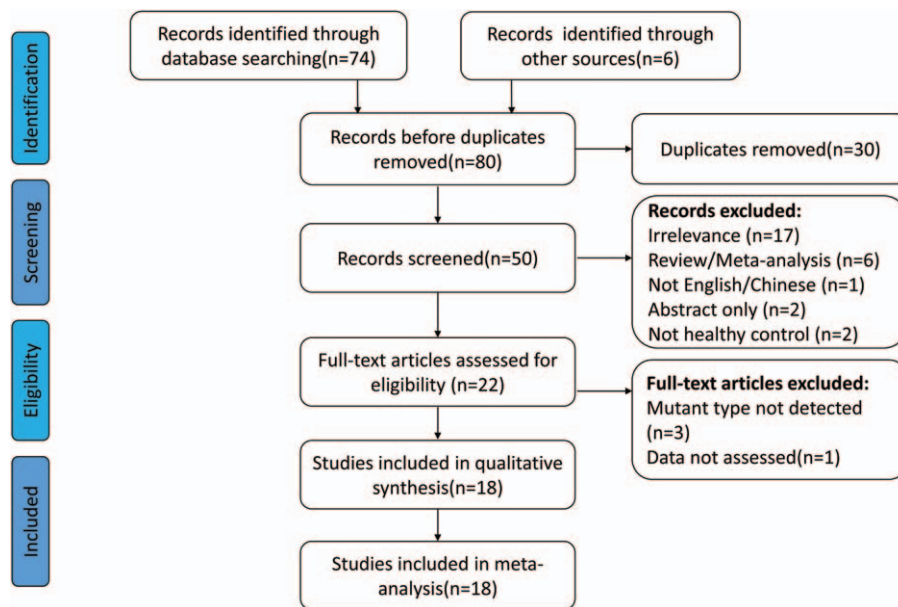


Figure 1. Flow diagram of study selection in the meta-analysis.

different regions and races. Thus, periodontitis has become one of the hot research fields all over the world.

The traditional method for dealing with periodontitis mainly focused on removing pathogenic bacteria, which resulted in bacteria resistance and disease recurrence. Besides, the host inflammatory response plays a critical role in the destruction of periodontal tissue. In the recent decades, the development of sequencing technology enabled us to discuss whether the variations of host's immune-related Deoxyribose Nucleic Acid molecules affected the occurrence and development of diseases. Thus, there is a great significance to discuss the gene variants of immune-related molecules for the prevention and treatment of periodontitis. Luigi^[5] elucidated host genetic variants may work in the occurrence and development of AP through selectively participating in the dysbiotic process. Hajishengallis and Sahingur^[6] reported a polymorphic site in the TLR9 gene promoter region differentially expressed and TLR9 gene and protein expression increased in CP. Toll-like receptor 4 (TLR4) was a pattern-recognition receptor, which played an important part in innate immunity by realizing lipid-based structures of bacteria and mediating intracellular signaling.^[7,8] Furthermore, Many studies reported the association between TLR4 polymorphism and periodontitis susceptibility, and they mainly focused on TLR4-299A>G or TLR4-399C>T of Caucasian but conducted different conclusions. Therefore, this meta-analysis and subgroup analyses were carried out to further illuminate the relationship between TLR4 polymorphism and periodontitis susceptibility based on the currently available studies.

2. Materials and methods

This review is not a primary research, so ethical approval and informed consent are not necessary. This meta-analysis was reported following the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) statement.^[9]

2.1. Inclusion and exclusion criteria

All retrieved literatures fit the following criteria should be included: the study tested TLR4 polymorphism and periodontitis susceptibility, participants in studies must be explicitly diagnosed with CP or AP, the numbers of every genotypes were available both in case and control groups. The study met the following criteria should be excluded: the review or meta-analysis about this theme, the article not described in Chinese or English, no control subjects or without healthy control group, participants who were pregnant or lactating, mutant type not detected.

2.2. Search strategy

The authors performed a systematic search in CNKI, PubMed, and Embase up to January 4, 2016 using key words: “polymorphism OR mutation OR variant” AND “TLR4 OR Toll like receptor 4” AND “periodontal disease OR periodontitis.” We also selected references of the relative reviews by hand searching. The details of search strategy in PubMed are summarized in “Appendix 1.”

2.3. Data extraction

Two authors extracted the useful information independently and any disagreement was solved by discussing until reaching an agreement or consulting a third author if needed. The extracted data were as follows: the first author's name and the publication date, the country or ethnicity of study participants, polymorphisms under investigation, disease type, the numbers of every genotypes both in case and control groups, genotyping method, the Hardy-Weinberg equilibrium (HWE) of the controls.

2.4. Quality assessment

Two of us conducted the quality assessment of included studies according to the Newcastle-Ottawa Scale case control study

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

Refs.	Country (ethnicity)	Polymorphism	Disease type	Case/control (n)				Genotype method	HWE
				11	12	22	Total		
Garlet et al ^[15]	Brazil (mixed)	TLR4-299A>G (rs4986790)	CP	6/12	56/74	135/131	197/217	TaqMan	Yes
Sahingur et al ^[16]	USA (Caucasian)			95/59	19/17	0/1	114/77	TaqMan	Yes
Schulz et al ^[17]	Germany (Caucasian)			52/73	8/7	0/0	60/80	Taq-polymerase	Yes
Izakovicova Holla et al ^[18]	Czech (Caucasian)			147/195	24/23	0/0	171/218	Taq-polymerase	Yes
Brett et al ^[19]	UK (Caucasian)			47/90	6/7	0/0	53/97	PCR	Yes
Berdeli et al ^[20]	Turkey (Caucasian)			79/100	4/6	0/0	83/106	PCR-RFLP	Yes
James et al ^[21]	UK (Caucasian)			77/78	17/16	1/0	95/94	PCR	Yes
Laine et al ^[22]	Netherlands (Caucasian)			90/90	10/8	0/1	100/99	PCR	Yes
Noack et al ^[23]	Germany (Caucasian)			96/68	12/8	0/0	108/76	PCR	Yes
Folwaczny et al ^[24]	Germany (Caucasian)			234/236		10/8	244/244	PCR-RFLP	NR
Tervonen et al ^[25]	Finland (Caucasian)			38/142		13/36	51/178	PCR	NR
Schulz et al ^[17]	Germany (Caucasian)		AP	66/73	7/7	0/0	73/80	Taq-polymerase	Yes
Noack et al ^[26]	Germany (Caucasian)			100/71	11/9	0/0	111/80	PCR	Yes
Emingil et al ^[27]	Turkey (white)			86/147	4/7	0/1	90/155	PCR-RFLP	Yes
Brett et al ^[19]	UK (Caucasian)			37/90	8/7	0/0	45/97	PCR	Yes
James et al ^[21]	UK (Caucasian)			69/103	4/20	0/0	73/123	PCR	Yes
Melanie ^[28]	USA (Africa)			59/44	9/7	2/2	70/53	TaqMan	Yes
Schulz et al ^[17]	Germany (Caucasian)	TLR4-399C>T (rs4986791)	CP	52/73	8/7	0/0	60/80	Taq-polymerase	Yes
Izakovicova Holla et al ^[18]	Czech (Caucasian)			147/196	24/22	0/0	171/218	Taq-polymerase	Yes
Brett et al ^[19]	UK (Caucasian)			50/78	4/17	0/0	54/95	PCR	Yes
Berdeli et al ^[20]	Turkey (Caucasian)			80/101	3/5	0/0	83/106	PCR-RFLP	Yes
James et al ^[21]	UK (Caucasian)			73/74	20/18	1/0	94/92	PCR	Yes
Laine et al ^[22]	Dutch (Caucasian)			90/90	10/8	0/1	100/99	PCR	Yes
Noack et al ^[23]	Germany (Caucasian)			96/68	12/8	0/0	108/76	PCR	Yes
Reddy et al ^[29]	India (Asian)			56/59	3/1	1/0	60/60	PCR-RFLP	Yes
Folwaczny et al ^[24]	Germany (Caucasian)			233/235		11/9	244/244	PCR-RFLP	NR
Schulz et al ^[17]	Germany (Caucasian)		AP	66/73	7/7	0/0	73/80	Taq-polymerase	Yes
Noack et al ^[26]	Germany (Caucasian)			100/71	11/9	0/0	111/80	PCR	Yes
Emingil et al ^[27]	Turkey (white)			88/148	2/7	0/0	90/155	PCR-RFLP	Yes
Brett et al ^[19]	UK (Caucasian)			46/78	3/17	0/0	45/97	PCR	Yes
Yu et al ^[30]	China (Asian)	TLR4C>G (rs11536889)	CP	32/6	212/52	379/80	623/138	PCR-SNP	Yes
Fukusaki et al ^[31]	Japan (Asian)			10/2	18/24	69/73	97/99	PCR	Yes
Ding et al ^[32]	China (Asian)			34/11	220/97	406/149	660/257	PCR	Yes
Yu et al ^[30]	China (Asian)	TLR4C>G (rs7873784)		4/0	82/27	536/112	622/139	PCR-SNP	Yes
Ding et al ^[32]	China (Asian)			4/0	86/49	565/201	655/250	PCR	Yes
Yu et al ^[30]	China (Asian)	TLR4C>T (rs10759930)		84/18	295/68	248/54	627/140	PCR-SNP	Yes
Ding et al ^[32]	China (Asian)			90/33	311/123	262/101	663/257	PCR	Yes
Yu et al ^[30]	China (Asian)	TLR4A>G (rs10983755)		56/13	246/52	322/74	624/139	PCR-SNP	Yes
Ding et al ^[32]	China (Asian)			60/24	259/96	341/136	660/256	PCR	Yes
Yu et al ^[30]	China (Asian)	TLR4A>G (rs11536879)		500/112	112/24	13/4	625/140	PCR-SNP	Yes
Ding et al ^[32]	China (Asian)			526/205	119/44	13/8	658/257	PCR	Yes
Yu et al ^[30]	China (Asian)	TLR4A>G (rs1927907)		64/15	232/54	292/62	588/131	PCR-SNP	Yes
Ding et al ^[32]	China (Asian)			70/29	250/103	315/118	635/250	PCR	Yes

11 = Wild homozygous genotype, 12 = mutant heterozygote, 22 = mutant homozygote, AP = aggressive periodontitis, control = healthy group, CP = chronic periodontitis, HWE = Hardy-Weinberg equilibrium, NR = not reported, PCR = polymerase chain reaction, RFLP = restriction fragment length polymorphism, SNP = single nucleotide polymorphism, TLR4 = Toll-like receptor 4.

(NOS).^[10] This standard assessed 3 sections (selection, comparability, exposure) and 8 items. In the selection and exposure categories, a quality research item received 1 star, and a comparable category could receive at most 2 stars. The quality assessment values ranged from 0 to 9 stars. Generally, the study which scored at least 5 points was considered to be included in meta-analysis.

2.5. Data analyses

The authors calculated the odds ratios (ORs) and corresponding 95% confidence interval (CI) to estimate the association between TLR4 polymorphism and periodontitis in 5 genetic models: allele

comparison (1 vs 2), homozygote comparison (11 vs 22), heterozygote comparison (12 vs 11), dominant model (22 + 12 vs 11), recessive model (11 + 12 vs 22). Heterogeneity was tested using *I*² statistics. Values of *P* < 0.1 or *I*² > 50% indicated obvious heterogeneity and the random effects model was considered to use; otherwise, use the fixed effects model.^[11] Subgroup analyses were performed according to ethnicity, smoking status. Sensitive analysis was carried out to test the stabilization of the pooled results.^[12] Publication bias was detected with funnel plot in visual and with quantitative method of Begg and Egger linear regression.^[13,14] All data analyses were conducted by STATA software, version 12.0. Using 2-sided *P*-values and *P* < 0.05 was supposed to have a statistically significance.

Table 2**Nonsmokers: characteristics of studies included in the meta-analysis.**

Refs.	Country (ethnicity)	Polymorphism	Disease type	Case/control (n)				Genotype method	HWE
				11	12	22	Total		
Berdeli et al ^[20]	Turkey (Caucasian)	TLR4-299A>G (rs4986790)	CP	50/95	4/6	0/0	54/101	Taq-polymerase	Yes
Izakovicova Holla et al ^[18]	Czech (Caucasian)			94/127	16/18	0/0	110/145	PCR-RFLP	Yes
Berdeli et al ^[20]	Turkey (Caucasian)	TLR4-399C>T (rs4986791)	CP	51/96	3/5	0/0	54/101	Taq-polymerase	Yes
Izakovicova Holla et al ^[18]	Czech (Caucasian)			94/128	16/17	0/0	110/145	PCR-RFLP	Yes

11 = Wild homozygous genotype, 12 = mutant heterozygote, 22 = mutant homozygote, control = healthy group, CP = chronic periodontitis, HWE = Hardy-Weinberg equilibrium, PCR = polymerase chain reaction, RFLP = restriction fragment length polymorphism, TLR4 = Toll-like receptor 4.

3. Results

3.1. Study selection and characteristics

Seventy-four studies from database searching and 6 studies from manual retrieval, counting up to 80 studies, were identified in this meta-analysis. After removing 30 duplicates, the remaining 50 studies need further screening. According to inclusion and exclusion criteria, this meta-analysis ultimately included 18 studies,^[15–32] which included 2453 healthy participants and 2987 patients with CP and 462 patients with AP. The flow diagram of study selection is shown in Fig. 1.

The basic characteristics of included studies are shown in Table 1. Among the 18 included studies, 4 studies were about Asian,^[29–32] 12 studies were about Caucasian,^[16–27] 1 study was conducted in Brazil (mixed),^[15] and 1 in USA (Africa).^[28] Only 2 studies^[18,20] reported the relationship between gene polymorphism and nonsmokers, and the basic characteristics are summarized in Table 2. The controls of 2 studies' HWE were not reported, and we could not calculate them in any way.^[24,25]

3.2. Quality assessment

The details of quality assessment based on the NOS are shown in Table 3. The last column in each row listed the total score of each study. One study had low score of 2 points, which would be considered to conduct the sensitive analysis. The percentage of different star numbers involving in all included studies is revealed in Fig. 2. Except for item "Non-response rate" which the percentage of 0 star was 100%, the remaining items has low percentage of 0 star. In general, the quality of included studies was good and fair.

3.3. Meta-analyses

In the association between TLR4C>G (rs7873784) polymorphism and CP susceptibility, there was a significance between TLR4C>G (rs7873784) allele and CP in Asian, and its recessive model was also significant (for C vs G: OR=0.72, 95% CI=0.54–0.95, $I^2=0\%$; for CC+CG vs GG: OR=0.66, 95% CI=0.49–0.89, $I^2=0\%$) (Figs. 3 and 4). All the genetic models of other TLR4 gene polymorphism included in this article in overall and subgroup revealed no significance, and the results are listed in Table 4. The result of sensitive analysis was still nonsignificant but the heterogeneity significantly decreased, which was conducted by removing the study of low quality (Table 5).

3.4. Publication bias

The funnel plot method was used when the number of included studies at least 10 at which point it had relatively high test efficiency.^[33] The 11 included studies about TLR4-299A>G

polymorphism with CP had nearly symmetric funnel plot, which indicated no obvious publication bias (Fig. 5). The studies which had small quantity of 2 could not be detected the publication bias. Egger and Begg test did not indicate obvious publication bias for other studies (for TLR4-299A>G with AP: A vs G: $P=0.64$; AG vs AA: $P=0.95$; GG+AG vs AA: $P=0.90$. for TLR4-399C>T with CP: C vs T: $P=0.88$; CC vs TT: $P=0.41$; AG vs AA: $P=0.78$; GG+AG vs AA: $P=0.49$; AA+AG vs GG: $P=0.60$. for TLR4-399C>T with AP: C vs T: $P=0.38$; AG vs AA: $P=0.36$. for TLR4C>G (rs11536889) with CP: C vs G: $P=0.19$; CC vs GG: $P=0.23$; CG vs CC: $P=0.27$; GG+CG vs CC: $P=0.25$; CC+CG vs GG: $P=0.15$).

4. Discussion

This meta-analysis systematically collected studies about TLR4 gene polymorphism associated with periodontitis from frequently used databases and manual retrieval. We analyzed the existing different TLR4 gene associated with different periodontitis and ultimately found that there was a significance between TLR4C>G (rs7873784) polymorphism and CP about C and G allele and its recessive model was also significant in Asian, which indicated that the Asian people suffered from CP might due to TLR4C>G (rs7873784) and it possibly passed on to offsprings in the form of recessiveness. On account of only 2 included studies about this gene, the reliability of this conclusion still need more research to demonstrate.

Previous meta-analyses about TLR4 gene polymorphism mainly focused on TLR4-299A>G or TLR4-399C>T of Caucasian, which made different conclusions. Ozturk and Vieira^[34] included 7 studies and calculated the OR value of major allele versus minor allele for both TLR4-299A>G and TLR4-399C>T polymorphisms, and concluded that the TLR4-299A>G may be a risk factor against CP (OR=1.43, 95% CI=1.04–1.97) and the TLR4-399C>T appeared to be a protective factor to AP (OR=0.29, 95% CI=0.13–0.61). Song et al^[35] and Zheng et al^[36] used differently 4 genetic models, and their overall results associated with ethnic analysis all failed to reveal any association between TLR4-299A>G or TLR4-399C>T and periodontitis. Besides, Zheng et al^[36] found a significantly increased risk for periodontitis in recessive models of TLR4-299A>G. Han et al^[37] indicated that both TLR4-299A>G and TLR4-399C>T showed elevated risk of CP in Caucasians.

Our meta-analyses have many superiorities. This pooled analysis systematically included 18 studies, excluded studies lack of specific statistics and conducted quality assessment for every included study. Besides, this study analyzed five genetic models to explore inheritance patterns of genes. We also carried out subgroup analyses of participants' ethnicity and smoking status. Based on the current studies, the authors did not detect any

Table 3

Evaluations of the qualities of the included studies based on the Newcastle-Ottawa Scale.

Refs.	Selection			Comparability			Exposure		Total
	(1) Is the case definition adequate	(2) Representativeness of the cases	(3) Selection of controls	(4) Definition of controls	(1) Comparability of cases and controls on the basis of the design or analysis	(1) Ascertainment of exposure	(2) Same method of ascertainment for cases and controls	(3) Nonresponse rate	
Garlet et al ^[15]	*	*	*	*	**	*	*	0	8
Sahingur et al ^[16]	*	*	*	*	*	*	*	0	7
Schulz et al ^[17]	*	*	*	*	**	*	*	0	8
Izakovicova Holla et al ^[18]	*	*	*	*	**	*	*	0	8
Brett et al ^[19]	*	*	0	0	0	0	0	0	2
Berdelli et al ^[20]	*	*	*	*	*	*	*	0	7
James et al ^[21]	*	*	*	*	*	*	*	0	7
Laine et al ^[22]	*	*	*	*	*	*	*	0	7
Noack et al ^[23]	*	*	*	*	*	0	*	0	6
Folwaczny et al ^[24]	*	*	*	*	0	*	*	0	6
Tervonen et al ^[25]	*	*	*	*	*	*	*	0	7
Noack et al ^[26]	*	*	*	*	*	0	*	0	6
Emingil et al ^[27]	*	*	*	*	*	0	*	0	6
Melanije ^[28]	*	*	*	*	*	*	*	0	7
Reddy et al ^[29]	*	*	*	*	0	*	*	0	6
Yu et al ^[30]	*	*	*	*	*	*	*	0	7
Fukusaki et al ^[31]	*	*	*	*	*	*	*	0	7
Ding et al ^[32]	*	*	*	*	*	*	*	0	7

In the selection and exposure categories, a quality research item following (studies that precisely described or eligible) received 1 star, and a comparable category could receive at most 2 stars. The quality assessment values ranged from 0 to 9 stars.

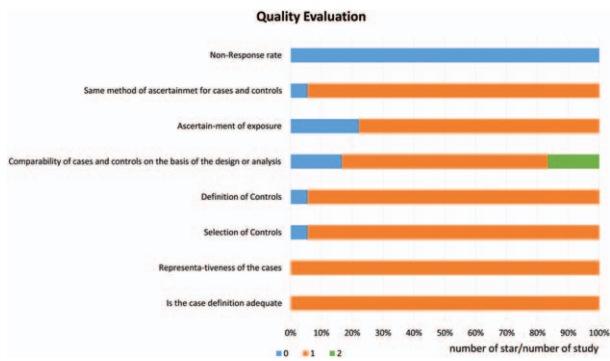


Figure 2. Quality evaluation of the included studies based on the Newcastle-Ottawa Scale.

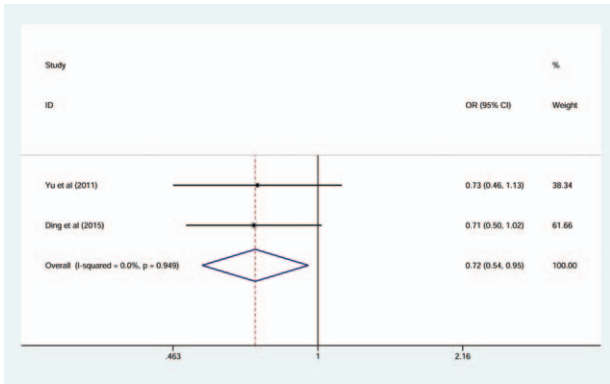


Figure 3. Forest plot for TLR4C>G (rs7873784) associated with chronic periodontitis in C versus G comparison (fixed-effect model).

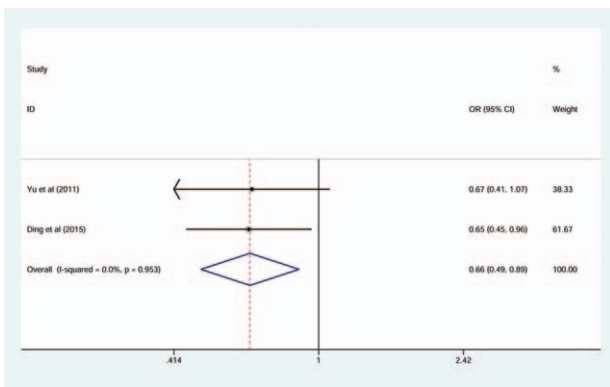


Figure 4. Forest plot for TLR4C>G (rs7873784) associated with chronic periodontitis in CC+CG versus GG comparison (fixed-effect model).

Table 4
Meta-analysis of the association between the TLR4 gene polymorphism and periodontitis.

Variables	Overall and subgroup analysis	Disease type	No. of studies	1 vs 2	11 vs 22	12 vs 11	22+12 vs 11	11+12 vs 22
				OR (95% CI)	OR (95% CI)	OR (95% CI)	OR (95% CI)	OR (95% CI)
TLR4-299A>G (rs4986790)	Total	CP	11	0.90 (0.72, 1.12)	0.68 (0.30, 1.56)	1.16 (0.86, 1.55)	1.21 (0.94, 1.57)	0.74 (0.50, 1.09)
	Caucasian		10	1.05 (0.79, 1.39)	1.61 (0.32, 8.04)	1.13 (0.83, 1.53)	1.17 (0.90, 1.53)	1.09 (0.71, 1.67)
	Brazil (mixed)		1	0.66 (0.32, 1.34)	0.49 (0.18, 1.29)	1.51 (0.54, 4.28)	1.86 (0.69, 5.06)	0.70 (0.46, 1.05)
	Nonsmokers		2	0.83 (0.45, 1.54)	—	1.19 (0.64, 2.24)	—	—
	Total	AP	6	1.15 (0.77, 1.71)	1.46 (0.27, 7.86)	0.90 (0.59, 1.38)	0.88 (0.58, 1.34)	1.45 (0.27, 7.80)
TLR4-399C>T (rs4986791)	Total	CP	5	1.16 (0.74, 1.81)	1.76 (0.07, 43.66)	0.95 (0.48, 1.91)	0.93 (0.47, 1.86)	1.76 (0.07, 43.59)
	Caucasian		1	1.13 (0.49, 2.59)	1.34 (0.23, 7.91)	0.95 (0.34, 2.68)	0.91 (0.36, 2.34)	1.33 (0.23, 7.84)
	USA (Africa)		9	0.88 (0.65, 1.20)	0.70 (0.14, 3.58)	1.13 (0.81, 1.56)	1.15 (0.85, 1.56)	0.72 (0.14, 3.67)
	Total	CP	8	0.92 (0.67, 1.26)	0.99 (0.14, 7.13)	1.10 (0.79, 1.53)	1.11 (0.82, 1.51)	1.02 (0.14, 7.28)
	Caucasian		1	0.19 (1.28, 1.28)	0.32 (0.01, 7.93)	3.16 (0.32, 31.29)	4.21 (0.46, 38.87)	0.33 (0.01, 8.21)
TLR4C>G (rs11536889)	Total	AP	2	0.81 (0.43, 1.53)	—	1.25 (0.65, 2.41)	—	—
	Total (Caucasian)		4	1.50 (0.87, 2.58)	—	0.66 (0.38, 1.14)	—	—
	Total (Asian)		3	0.98 (0.82, 1.18)	1.42 (0.84, 2.39)	0.61 (0.36, 1.03)	0.67 (0.39, 1.11)	0.90 (0.73, 1.12)
	Total (Asian)		2	0.72 (0.54, 0.95)	2.15 (0.32, 19.84)	0.25 (0.03, 2.01)	0.37 (0.05, 2.92)	0.66 (0.49, 0.89)
	Total (Asian)		2	1.00 (0.85, 1.18)	1.04 (0.72, 1.49)	0.93 (0.65, 1.32)	0.94 (0.67, 1.32)	0.98 (0.78, 1.23)
TLR4A>G (rs10983755)	Total	CP	2	1.03 (0.86, 1.23)	0.99 (0.66, 1.49)	1.09 (0.72, 1.65)	1.04 (0.70, 1.54)	1.06 (0.85, 1.34)
	Total (Asian)		2	1.06 (0.82, 1.36)	0.98 (0.68, 1.41)	1.05 (0.78, 1.42)	0.99 (0.75, 1.32)	1.51 (0.75, 3.05)
	Total (Asian)		2	0.78 (0.55, 1.09)	0.91 (0.62, 1.33)	1.01 (0.68, 1.48)	1.06 (0.74, 1.53)	0.91 (0.72, 1.15)
	Total (Asian)		2	0.78 (0.55, 1.09)	0.91 (0.62, 1.33)	1.01 (0.68, 1.48)	1.06 (0.74, 1.53)	0.91 (0.72, 1.15)
	Total (Asian)		2	0.78 (0.55, 1.09)	0.91 (0.62, 1.33)	1.01 (0.68, 1.48)	1.06 (0.74, 1.53)	0.91 (0.72, 1.15)

11 = Wild homozygous genotype, 12 = mutant heterozygote, 22 = mutant homozygote, AP = aggressive periodontitis, CP = chronic periodontitis, OR = odds ratio, TLR4 = Toll-like receptor 4. Bold values mean that the data have statistical significance.

Table 5

Sensitive analysis by removing the study of low quality.

Variables	Disease type	No. of studies	1 vs 2		11 vs 22		12 vs 11		22 + 12 vs 11		11 + 12 vs 22	
			OR (95% CI)	I ² (%)	OR (95% CI)	I ² (%)	OR (95% CI)	I ² (%)	OR (95% CI)	I ² (%)	OR (95% CI)	I ² (%)
TLR4-299A>G (rs4986790)	CP	10	0.91 (0.73, 1.14)	21.0	0.68 (0.30, 1.56)	0	1.13 (0.83, 1.53)	0	1.19 (0.91, 1.56)	0	0.74 (0.50, 1.09)	0
	AP	5	1.38 (0.89, 2.13)	0	1.46 (0.27, 7.86)	0	0.74 (0.46, 1.18)	0	0.72 (0.46, 1.14)	0	1.45 (0.27, 7.80)	0
TLR4-399C>T (rs4986791)	CP	8	0.78 (0.56, 1.09)	0	0.70 (0.14, 3.58)	0	1.28 (0.90, 1.81)	0	1.28 (0.93, 1.77)	0	0.72 (0.14, 3.67)	0
	AP	3	1.18 (0.63, 2.20)	0	—	—	0.84 (0.44, 1.60)	0	—	—	—	—

11 = Wild homozygous genotype, 12 = mutant heterozygote, 22 = mutant homozygote, AP = aggressive periodontitis, CP = chronic periodontitis, CI = confidence interval, OR = odds ratio, TLR4 = Toll-like receptor 4.

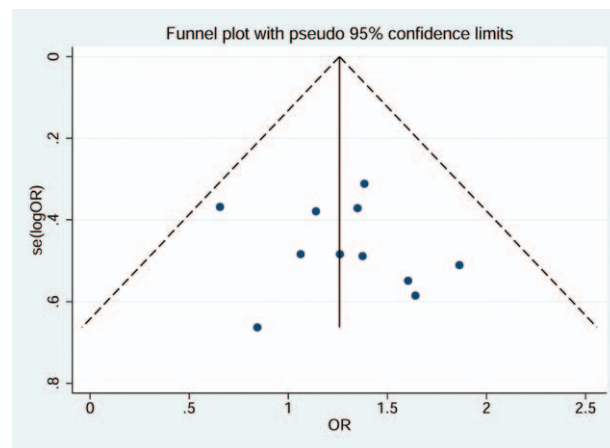


Figure 5. Funnel plot of included studies for TLR4-299A>G associated with chronic periodontitis.

association between TLR4 polymorphisms and periodontitis susceptibility except for TLR4C>G (rs7873784). Through removing the low quality studies, the sensitive analysis only decreased heterogeneity, but could not change the results.

The limitation of this meta-analysis should not be ignored. First, language limitation made us cannot obtain more relative studies. Second, the relatively clinical research in Asian lacked. Third, the case-control study about the association between TLR4 polymorphisms and smoking status was few, which made us cannot extract data directly. Fourth, whether periodontal treatment could influence gene mutation should be seriously considered, but the studies included in this meta-analysis did not make the unified description, which may bring in the bias. Finally, many risk factors of periodontitis were not studied in clinical trials that led to the limitation of subgroup analyses. In conclusion, the unified baseline of studies and more well-designed clinical trials were expected.

5. Conclusions

We found that there was a significance between TLR4C>G (rs7873784) polymorphism and CP about C and G allele and its recessive model was also significant in Asian, which indicated that the Asian people suffered from CP may due to TLR4C>G (rs7873784) and it possibly passed on to offsprings in the form of recessiveness. However, the overall and subgroup analyses in other TLR4 polymorphism included in this study found no significance. Large quantity and high quality researches were expected to explore the pathogenesis of periodontitis.

Appendix 1: PubMed search terms

#1 Search ((((((Polymorphisms, Genetic[Title/Abstract]) OR Genetic Polymorphism[Title/Abstract]) OR Polymorphism (Genetics)[Title/Abstract]) OR Genetic Polymorphisms[Title/Abstract]) OR gene polymorphism[Title/Abstract]) OR mutation [Title/Abstract]) OR variant[Title/Abstract]) OR “Polymorphism, Genetic”[Mesh]

#2 Search (((((Toll Like Receptor 4[Title/Abstract]) OR Toll4 Receptor[Title/Abstract]) OR Toll 4 Receptor[Title/Abstract]) OR TLR4 Receptor[Title/Abstract]) OR Receptor, TLR4[Title/Abstract]) OR “Toll-Like Receptor4”[Mesh]

#3 Search (((((((Disease, Periodontal[Title/Abstract]) OR Diseases, Periodontal[Title/Abstract]) OR Periodontal Disease [Title/Abstract]) OR Parodontosis[Title/Abstract]) OR Pyorrhoea Alveolaris[Title/Abstract]) OR Periodontitides[Title/Abstract]) OR Pericementitis[Title/Abstract]) OR Pericementitides[Title/Abstract]) OR “Periodontal Diseases”[Mesh]) OR “Periodontitis”[Mesh]

#4 Search #1 AND #2 AND #3

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