Int J Clin Exp Med 2017;10(1):142-152
www.ijcem.com /ISSN:1940-5901/IJCEM0038138

Review Article

The interaction between polymorphisms in MnSOD and prostate cancer risk: a meta-analysis and systematic review

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Received July 19, 2016; Accepted October 29, 2016; Epub January 15, 2017; Published January 30, 2017

Abstract: Previous studies have investigated the relationship between manganese superoxide dismutase (MnSOD) Val16Ala and Ala-9Val polymorphisms and prostate cancer (PCa) risk, but the results remained controversial. This meta-analysis was therefore performed to clarify this association. The databases PubMed, EMbase, Web of Science, Wanfang and China National Knowledge Infrastructure (CNKI) were searched to identify relevant eligible studies. Pooled odds ratios (ORs) with 95% confidence intervals (CIs) were calculated to evaluate the strength of the association. A total of thirteen studies encompassing 4,583 cases and 7,207 controls were eligible for Ala-16Val polymorphism, and five case-control studies were eligible for Ala-9Val polymorphism. Overall, significant relationship between Ala-9Val polymorphism and PCa risk was identified in the allelic, dominant and homozygote models, while no finding of significant association was observed for Ala-16Val polymorphism. However, as for Ala-16Val polymorphism, when the stratification analysis was conducted by Hardy Weinberg Equilibrium (HWE) status, we found that the Ala-16Val polymorphism was significantly associated with an increased risk of PCa among these studies that were not conformed to HWE. To sumup, the present meta-analysis suggests that *MnSOD* Ala-9Val polymorphism is a risk factor for PCa. More studies with larger sample sizes, well controlled confounding factors are warranted to validate our findings.

Keywords: MnSOD, gene, polymorphism, meta-analysis

Introduction

Prostate cancer (PCa) is considered as the highest incidence of cancer in men, and it has become the second largest cause to deaths of males in Western countries [1, 2]. Currently, a large number of studies have found genetic environment factors play significant roles in the pathogenesis of PCa. However, there existed some differences in the risk of different individuals suffering from PCa under the same environmental conditions, indicating that genetic factors may play a critical part in PCa risk. Currently, there are quantities of articles reported the association between genetic polymorphisms and PCa risk [3-6], and the potential mechanisms for pathogenesis remain to be further clarified.

Under normal physiological conditions, several antioxidant enzymes in the body, which are represented by MnSOD, can continue to remove excess reactive oxygen species (ROSs) produced by cells of our body in order to stabilize the oxidation-reduction system [7]. On the contrary, once this dynamic equilibrium is destroyed, possibly causing excess ROS, and then trigger a variety of oxidative stress damages to cells or induce somatic mutations, perhaps leading to tumorigenesis. There has been reported that defective oxidative stress is a common system defects in tumor cells. Excess oxidative stress can carry out oxidative modification on the residues of key amino acid by influencing the intracellular redox state, in order to attack the cell's DNA, lipid quality and protein, and then induce pathogenesis of cancers

and other diseases [7, 8]. In addition, gene mutations in *MnSOD* promoter region which change the sequence of the gene, really become the central component of the results weakening its antioxidant effects. So far, a large number of studies have confirmed that the Ala polymorphism in *MnSOD* promoter region significantly increased the risk of breast cancer (BC) [9], esophageal cancer [10] and cervical cancer (CC) [11], whereas some studies have reported that this mutation reduced the risk of lung cancer (LC) [12] and bladder cancer (BC) [13]. In addition, studies also indicated an increased risk of PCa [14], but results were not consistent.

Individual studies might have underpowered for the purpose to detect the overall results. A quantitative synthesis of the evidence has been deemed critically given the amount of the accumulated data. For more results, we expect to expand the sample sizes, then perform a retrospective study to reduce bias, which is due to insufficient sample sizes or quality differences among similar studies, in purpose of obtaining more credible conclusions. Therefore, we designed this meta-analysis, on the basis of the case-control studies on the associations of *MnSOD* Ala-16Val and Ala-9Val polymorphisms and PCa risk, providing more reliable lines for the evidence-based clinical and basic research.

Materials and methods

Publication search

The following terms were retrieved in electronic databases such as EMbase, PubMed, Web of Science, Wanfang and CNKI databases: "manganese superoxide dismutase OR MnSOD" AND "variant OR mutation OR SNP OR polymorphism OR genotype" AND "prostate cancer OR prostate carcinoma OR prostate adenocarcinoma OR prostate neoplasm OR prostate tumor". Other relevant publications were also identified hand search of the indexed references of enrolled studies. And the last research was up to April 26, 2016.

Inclusion criteria and exclusion criteria

The studies were enrolled based on the following inclusion criteria: 1) studies that evaluate *MnSOD* polymorphisms and PCa risk; 2) PCa cases were verified by histopathological analyses in these studies and 3) Case-control or cohort studies published in official medical journals. Meanwhile, the exclusion criteria were presented as follows: 1) duplicate information, 2) reports of clinical cases, series, comments, systematic reviews and editorial and 3) insufficient information. Studies previously published in some other languages instead of English were also excluded. According to the inclusion criteria, all the records were checked by two authors independently and consensus was reached on each record.

Data extraction

We extracted data on the basis of a standard protocol. Studies that did not conform to the inclusion criteria, or those provided insufficient data, or those confirmed double publications were excluded. The data was included only once when the same data was found in different studies. The following details should be should be extracted from enrolled publications: the name of first author, publication year, ethnicity, frequencies of each genotype, genotyping method, source of control and *P* value of HWE.

Statistical analyses

In our meta-analysis, five genetic models were all used, including allelic contrast (B vs. A), recessive (BB vs. BA+AA), dominant (BA+BB vs. AA), homozygous (BB vs. AA), and heterozygous (BA vs. AA) models [15]. The association between *MnSOD* polymorphisms and PCa risk were compared by ORs and 95% CIs. In addition, Chi-square-based Q-tests were used to assess the heterogeneity among the individual studies, and significance was set at the $P<0.05$ level [16]. The random-effect model was conducted to evaluate the pooled OR (DerSimonian and Laird method) if there existed heterogeneity within individual studies [17]. On the contrary, the fixed-effect model was adopted (the Mantel-Haenszel method). Z test, with significance set at the $P<0.05$ level, was used to determine the pooled OR, and HWE was evaluated by Fisher's exact test with significance set at the $P<0.05$ level. The funnel plot was conducted to assess the potential publication bias [18]. Egger's linear regression test on the natural logarithm scale of the OR was used to evaluate the asymmetry of funnel plot, and significance was set at the $P<0.05$ level [19]. In order

Table 1. Baseline characteristics of eligible studies

SNP	First Author	Year	Ethnicity	Genotyping Method	Source of Control	Cancer Type	Case			Control			
							AA	AB	BB	AA	AB	BB	HWE
Ala-9Val	Ergen *et al.*	2007	Caucasian	PCR-RFLP	H-B	PCa	19	25	6	32	18	0	Y
	Arsova *et al.*	2008	Caucasian	PCR-RFLP	H-B	PCa	19	46	20	41	73	37	Y
	Kucukgergin *et al.*	2012	Caucasian	PCR-RFLP	P-B	PCa	43	65	26	66	69	24	Y
	Eken *et al.*	2013	Caucasian	PCR-RFLP	P-B	PCa	7	17	9	31	37	13	Y
	Parlaktas *et al.*	2015	Caucasian	PCR-RFLP	P-B	PCa	23	23	3	24	20	5	Y
Ala-16Val	Woodson *et al.*	2003	Caucasian	Sequenom	P-B	PCa	43	98	58	49	102	40	Y
	Li *et al.*	2005	Caucasian	PCR-RFLP	P-B	PCa	132	288	147	190	379	195	Y
	Taufer *et al.*	2005	Caucasian	PCR-RFLP	P-B	PCa	24	78	18	40	113	13	N
	Choi *et al.*	2007	Caucasian	Sequenom	P-B	PCa	112	239	104	293	610	311	Y
	Kang *et al.*	2007	African	TaqMan	P-B	PCa	31	57	15	122	194	79	Y
	Kang *et al.*	2007	Caucasian	TaqMan	P-B	PCa	275	578	297	376	686	320	Y
	Choi *et al.*	2008	Mixed	Sequenom	P-B	PCa	119	245	105	327	635	317	Y
	Mikhak *et al.*	2008	Caucasian	TaqMan	P-B	PCa	156	320	166	162	331	159	Y
	Iguchi *et al.*	2008	Caucasian	PCR-RFLP	H-B	PCa	9	24	24	37	59	41	Y
	Bica *et al.*	2009	Caucasian	PCR-RFLP	P-B	PCa	10	32	9	42	105	8	N
	Iguchi *et al.*	2009	Mixed	PCR-RFLP	H-B	PCa	41	86	60	40	96	39	Y
	Hemelrijck *et al.*	2014	Caucasian	Sequenom	P-B	PCa	53	100	50	90	190	80	Y
	Oskina *et al.*	2014	Caucasian	TaqMan	P-B	PCa	94	194	92	99	152	86	Y

HWE: Hardy-Weinberg equilibrium; SNP: single-nucleotide polymorphism; H-B: hospital based; P-B: population based; PCa: prostate cancer; A: wild type; B: mutated type; Y: control conformed to HWE; N: study did not conform to HWE. PCR-RFLP: Polymerase Chain Reaction-Restriction Fragment Length Polymorphism.

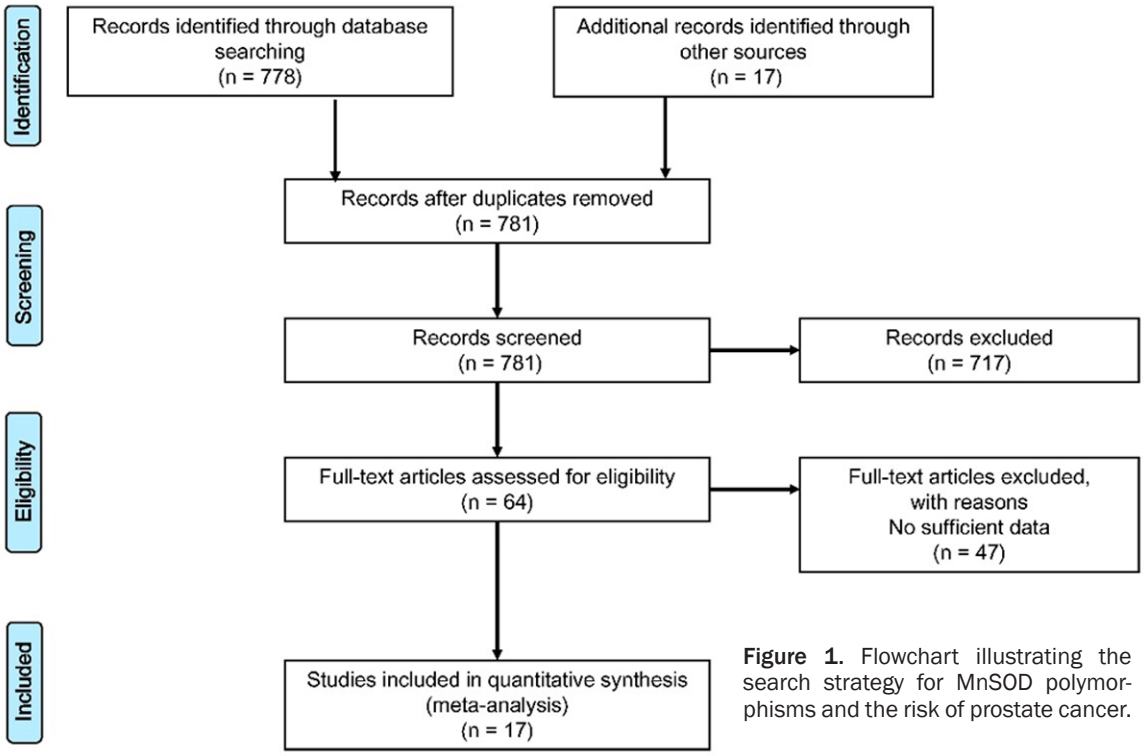

Figure 1. Flowchart illustrating the search strategy for MnSOD polymorphisms and the risk of prostate cancer.

to control the false positive error rate, we used Bonferroni method to adjust for multiple comparisons. Two-sided values of $P<0.05$ were considered to be statistically significant in pres-

Table 2. The overall analyses of *MnSOD* polymorphisms and prostate cancer risk

Polymorphisms	Comparison	Subgroup	N	P_H	P_Z	$P_{(Adjust)}$	Random	Fixed
Ala-16Val	B VS. A	Overall	12	0.212	0.049	0.490	1.059 (0.991-1.132)	1.056 (1.000-1.116)
	B VS. A	Caucasian	9	0.237	0.024	0.240	1.076 (0.999-1.159)	1.073 (1.009-1.140)
	B VS. A	Mixed	2	0.122	0.884	1.000	1.055 (0.825-1.351)	1.010 (0.884-1.154)
	B VS. A	PCR-RFLP	5	0.222	0.014	0.140	1.214 (1.035-1.424)	1.158 (1.030-1.301)
	B VS. A	Sequenom	3	0.824	0.408	1.000	0.960 (0.870-1.058)	0.960 (0.870-1.058)
	B VS. A	TaqMan	4	0.579	0.061	0.610	1.079 (0.997-1.169)	1.079 (0.997-1.169)
	B VS. A	H-B	2	0.331	0.018	0.180	1.342 (1.051-1.715)	1.344 (1.052-1.716)
	B VS. A	P-B	10	0.387	0.141	1.000	1.042 (0.983-1.105)	1.043 (0.986-1.103)
	B VS. A	N	2	0.532	0.033	0.330	1.339 (1.023-1.751)	1.338 (1.023-1.750)
	B VS. A	Y	10	0.284	0.117	1.000	1.044 (0.980-1.113)	1.046 (0.989-1.106)
	BA VS. AA	Overall	12	0.930	0.070	0.700	1.091 (0.992-1.199)	1.091 (0.993-1.199)
	BA VS. AA	Caucasian	9	0.840	0.068	0.680	1.103 (0.992-1.226)	1.104 (0.993-1.227)
	BA VS. AA	Mixed	2	0.516	0.856	1.000	1.021 (0.811-1.286)	1.021 (0.812-1.286)
	BA VS. AA	PCR-RFLP	5	0.773	0.356	1.000	1.100 (0.894-1.354)	1.102 (0.896-1.356)
	BA VS. AA	Sequenom	3	0.789	0.842	1.000	1.017 (0.859-1.203)	1.017 (0.860-1.204)
	BA VS. AA	TaqMan	4	0.634	0.064	0.640	1.138 (0.992-1.305)	1.138 (0.993-1.305)
	BA VS. AA	H-B	2	0.210	0.842	1.000	1.098 (0.598-2.015)	1.046 (0.671-1.631)
	BA VS. AA	P-B	10	0.946	0.070	0.700	1.093 (0.993-1.204)	1.093 (0.993-1.204)
	BA VS. AA	N	2	0.832	0.457	1.000	1.194 (0.746-1.910)	1.195 (0.747-1.911)
	BA VS. AA	Y	10	0.849	0.090	0.900	1.087 (0.987-1.197)	1.087 (0.987-1.197)
	BA+BB VS. AA	Overall	12	0.826	0.040	0.400	1.097 (1.003-1.199)	1.098 (1.004-1.201)
	BA+BB VS. AA	Caucasian	9	0.645	0.029	0.290	1.117 (1.010-1.234)	1.118 (1.012-1.236)
	BA+BB VS. AA	Mixed	2	0.877	0.867	1.000	1.019 (0.819-1.267)	1.019 (0.819-1.267)
	BA+BB VS. AA	PCR-RFLP	5	0.627	0.107	1.000	1.170 (0.960-1.424)	1.175 (0.966-1.429)
	BA+BB VS. AA	Sequenom	3	0.954	0.847	1.000	0.984 (0.839-1.155)	0.984 (0.839-1.155)
	BA+BB VS. AA	TaqMan	4	0.715	0.040	0.400	1.145 (1.006-1.302)	1.145 (1.006-1.303)
	BA+BB VS. AA	H-B	2	0.194	0.261	1.000	1.327 (0.734-2.398)	1.269 (0.837-1.923)
	BA+BB VS. AA	P-B	10	0.870	0.064	0.640	1.090 (0.995-1.194)	1.090 (0.995-1.195)
	BA+BB VS. AA	N	2	0.711	0.193	1.000	1.354 (0.854-2.145)	1.357 (0.857-2.148)
	BA+BB VS. AA	Y	10	0.771	0.067	0.670	1.088 (0.993-1.191)	1.089 (0.994-1.193)
	BB VS. AA	Overall	12	0.059	0.110	1.000	1.142 (0.971-1.342)	1.118 (1.001-1.249)
	BB VS. AA	Caucasian	9	0.064	0.066	0.660	1.194 (0.989-1.442)	1.157 (1.023-1.310)
	BB VS. AA	Mixed	2	0.142	0.940	1.000	1.092 (0.681-1.751)	1.010 (0.771-1.324)
	BB VS. AA	PCR-RFLP	5	0.051	0.016	0.160	1.780 (1.111-2.851)	1.380 (1.082-1.759)
	BB VS. AA	Sequenom	3	0.806	0.402	1.000	0.919 (0.753-1.121)	0.919 (0.753-1.121)
	BB VS. AA	TaqMan	4	0.485	0.071	0.710	1.160 (0.988-1.361)	1.159 (0.988-1.359)
	BB VS. AA	H-B	2	0.385	0.026	0.260	1.737 (1.061-2.845)	1.748 (1.071-2.853)
	BB VS. AA	P-B	10	0.088	0.276	1.000	1.094 (0.931-1.284)	1.091 (0.974-1.222)
	BB VS. AA	N	2	0.337	0.003	0.030	2.979 (1.477-6.009)	2.938 (1.458-5.923)
	BB VS. AA	Y	10	0.312	0.132	1.000	1.086 (0.958-1.231)	1.090 (0.974-1.220)
	BB VS. BA+AA	Overall	12	0.015	0.239	1.000	1.093 (0.942-1.269)	1.056 (0.964-1.155)
	BB VS. BA+AA	Caucasian	9	0.047	0.174	1.000	1.117 (0.952-1.310)	1.080 (0.977-1.195)
	BB VS. BA+AA	Mixed	2	0.020	0.632	1.000	1.163 (0.628-2.153)	1.008 (0.810-1.255)
	BB VS. BA+AA	PCR-RFLP	5	0.023	0.019	0.190	1.641 (1.084-2.484)	1.281 (1.054-1.557)
	BB VS. BA+AA	Sequenom	3	0.471	0.241	1.000	0.908 (0.771-1.069)	0.907 (0.770-1.068)
	BB VS. BA+AA	TaqMan	4	0.322	0.322	1.000	1.058 (0.911-1.229)	1.069 (0.937-1.219)
	BB VS. BA+AA	H-B	2	0.935	0.008	0.080	1.667 (1.141-2.435)	1.666 (1.140-2.434)

	BB VS. BA+AA	P-B	10	0.039	0.639	1.000	1.035 (0.896-1.197)	1.027 (0.935-1.127)
	BB VS. BA+AA	N	2	0.320	0.002	0.020	2.612 (1.425-4.786)	2.561 (1.394-4.705)
	BB VS. BA+AA	Y	10	0.129	0.466	1.000	1.037 (0.918-1.171)	1.035 (0.944-1.134)
Ala-9Val	B VS. A	Overall	6	0.170	0.001	0.010	1.343 (1.080-1.670)	1.320 (1.120-1.555)
	B VS. A	PCR-RFLP	5	0.103	0.004	0.040	1.387 (1.028-1.871)	1.341 (1.096-1.642)
	B VS. A	H-B	2	0.018	0.279	1.000	1.632 (0.673-3.958)	1.364 (0.989-1.881)
	B VS. A	P-B	4	0.543	0.006	0.060	1.305 (1.078-1.579)	1.304 (1.078-1.579)
	BA VS. AA	Overall	6	0.683	0.013	0.130	1.394 (1.069-1.820)	1.397 (1.072-1.821)
	BA VS. AA	PCR-RFLP	5	0.770	0.007	0.070	1.539 (1.122-2.110)	1.541 (1.125-2.111)
	BA VS. AA	H-B	2	0.315	0.050	0.500	1.677 (0.999-2.814)	1.673 (1.000-2.799)
	BA VS. AA	P-B	4	0.698	0.090	0.900	1.304 (0.956-1.780)	1.307 (0.959-1.782)
	BA+BB VS. AA	Overall	6	0.449	0.002	0.020	1.478 (1.148-1.902)	1.483 (1.154-1.906)
	BA+BB VS. AA	PCR-RFLP	5	0.399	0.002	0.020	1.583 (1.170-2.142)	1.587 (1.178-2.138)
	BA+BB VS. AA	H-B	2	0.123	0.028	0.280	1.855 (0.845-4.071)	1.742 (1.063-2.854)
	BA+BB VS. AA	P-B	4	0.624	0.024	0.240	1.393 (1.039-1.869)	1.400 (1.045-1.875)
	BB VS. AA	Overall	6	0.274	0.003	0.030	1.618 (1.061-2.468)	1.670 (1.186-2.352)
	BB VS. AA	PCR-RFLP	5	0.174	0.017	0.170	1.645 (0.884-3.060)	1.680 (1.097-2.573)
	BB VS. AA	H-B	2	0.050	0.384	1.000	3.610 (0.200-65.095)	1.715 (0.858-3.430)
	BB VS. AA	P-B	4	0.462	0.012	0.120	1.665 (1.12-2.475)	1.656 (1.117-2.454)
	BB VS. BA+AA	Overall	6	0.301	0.025	0.250	1.349 (0.955-1.907)	1.393 (1.043-1.859)
	BB VS. BA+AA	PCR-RFLP	5	0.237	0.169	1.000	1.268 (0.775-2.075)	1.297 (0.896-1.879)
	BB VS. BA+AA	H-B	2	0.061	0.482	1.000	2.615 (0.179-38.218)	1.237 (0.697-2.196)
	BB VS. BA+AA	P-B	4	0.574	0.030	0.300	1.456 (1.041-2.038)	1.449 (1.037-2.026)

P_H: *P*-value of heterogeneity test; P_Z: *P*-value of Z test; P-B: population-based; H-B: hospital-based; HWE: Hardy Weinberg Equilibrium; A: wild type; B: mutated type; Y: studies were conformed to HWE; N: studies were not conformed to HWE; PCR-RFLP: polymerase chain reaction-restriction fragment length polymorphism.

ent study. STATA 12.0 software was used for performing the statistical analyses (Stata Corp, College Station, TX, USA).

Results

Characteristics of eligible studies

Details of these enrolled studies were summarized in **Table 1** [14, 20-35], and the data selection process was presented in **Figure 1**. There were five eligible studies identified for Ala-9Val polymorphism in this meta-analysis, including 351 cases and 490 controls. And a total of 13 studies encompassing 4,583 cases and 7,207 controls met the inclusion criteria for Ala-16Val polymorphism. Methods of genotyping were PCR-RFLP, Sequenom and TaqMan, while the most commonly used method was PCR-RFLP (11/21, 52%), and the details of these studies were summarized in **Table 1**. In addition, there were two studies that were deviated from HWE [22, 27].

Main results

The results of the association between *MnSOD* polymorphisms (Ala-9Val and Ala-16Val) and PCa risk was showed in **Table 2**. Overall, significant relationship between Ala-9Val polymorphism in *MnSOD* promoter and PCa risk was identified in the allelic, dominant and homozygote models (B VS. A: OR = 1.343, 95% CI: 1.080-1.670, $P_{(Adjust)}$ = 0.010, **Figure 2A**; BA+BB VS. AA: OR = 1.478, 95% CI: 1.148-1.902, $P_{(Adjust)}$ = 0.020; BB VS. AA: OR = 1.618, 95% CI: 1.061-2.468, $P_{(Adjust)}$ = 0.030, **Table 2** and **Figure 2B**). However, no finding of significant association between Ala-16Val polymorphism and PCa risk was observed in all of the genetic models (**Table 2**).

Subgroup analyses

In the stratified analysis by genotyping method, the Ala-9Val polymorphism was significantly related to an increased risk of PCa in PCR-RFLP group in the allelic and dominant models (B VS. A: OR = 1.387, 95% CI: 1.028-1.871, $P_{(Adjust)}$ = 0.040; BA+BB VS. AA: OR = 1.583, 95% CI: 1.170-2.142, $P_{(Adjust)}$ = 0.020, **Table 2**). For Ala-16Val polymorphism, in the stratification analysis by HWE status, we observed that the Ala-16Val polymorphism was significantly associ-

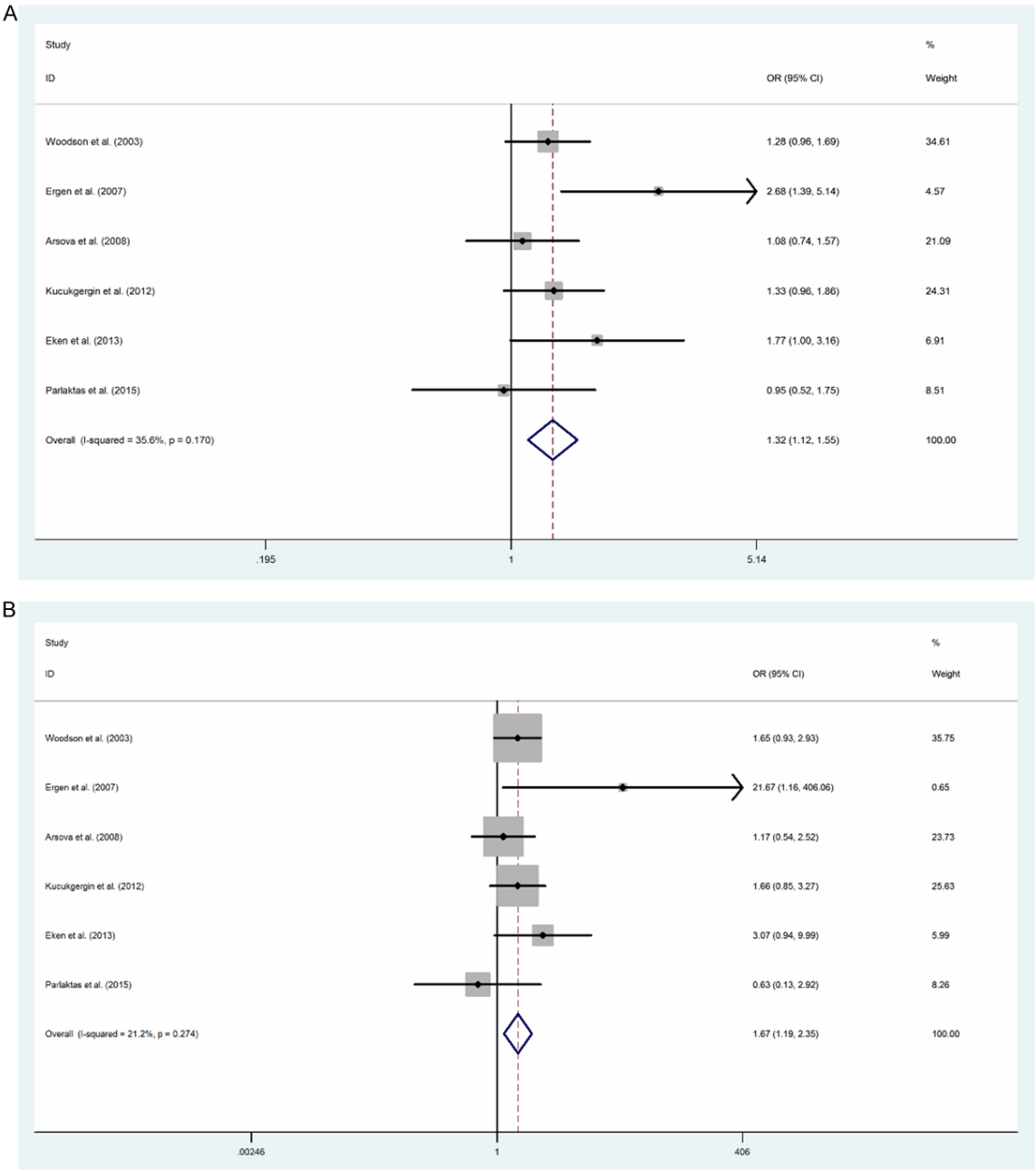

Figure 2. A. Forest plot of prostate cancer risk associated with the MnSOD. Ala-9Val polymorphism (B vs. A) in the whole. The squares and horizontal lines correspond to the study-specific OR and 95% CI. The area of the squares reflects the weight (inverse of the variance). The diamond represents the summary OR and 95% CI. B. Forest plot of prostate cancer risk associated with the MnSOD Ala-9Val polymorphism (BB vs. AA) in the whole. The squares and horizontal lines correspond to the study-specific OR and 95% CI. The area of the squares reflects the weight (inverse of the variance). The diamond represents the summary OR and 95% CI.

ated with an increased risk of PCa among these studies that were not in consistent with HWE (BB VS. AA: OR = 2.979, 95% CI: 1.477-6.009, $P_{(Adjust)}$ = 0.030; BB VS. BA+AA: OR = 2.612, 95% CI: 1.425-4.786, $P_{(Adjust)}$ = 0.020) (showed in **Table 2**).

As showed from Table S1, significant association between *MnSOD* polymorphisms and PCa risk was not observed when stratification analyses were conducted based on the disease status, such as aggressive vs. non-aggressive, the age when the diagnosis was made and *etc*.

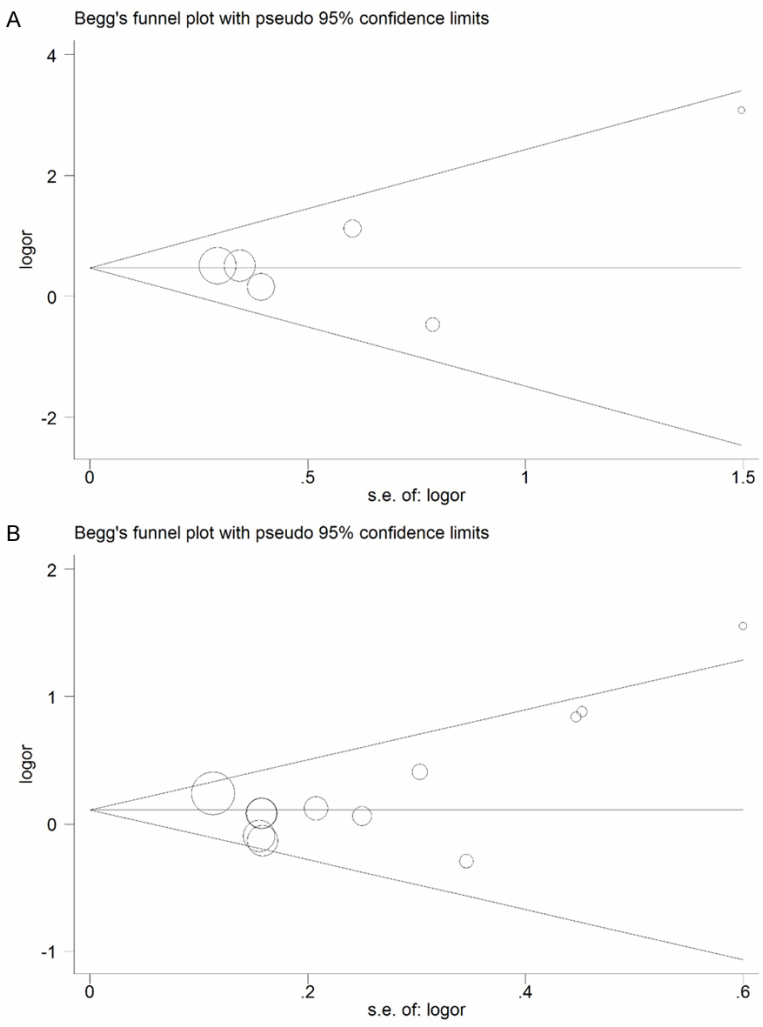

Figure 3. A. Begg's funnel plot for publication bias test (BB VS. AA in MnSOD Ala-9Val polymorphism). Each point represents a separate study for the indicated association. Log [OR], natural logarithm of OR. Horizontal line, mean effect size. B. Begg's funnel plot for publication bias test (BB VS. AA in MnSOD Ala-16Val polymorphism). Each point represents a separate study for the indicated association. Log [OR], natural logarithm of OR. Horizontal line, mean effect size.

Sensitivity analysis and publication bias

For the purpose of testing the robustness of relationship, sensitivity analysis was conducted through excluding studies one after another and analysis about the effect size was carried out among all of the remaining studies. Through excluding any of these studies, sensitivity analysis suggested that there was no study significantly influenced the combined ORs, proving the stability of results. Begg's funnel plot and Egger's test were used to estimate the publication bias. And no evidence of publication bias was visually observed form the funnel plot (**Figure 3A** and **3B**) and statistically verified by the Egger's test in any of the comparison models. In addition, the quality of the enrolled studies was assessed by Newcastle-Ottawa Scale (NOS) (**Table 3**).

Discussion

As an endogenous antioxidant localized in the mitochondria, Manganase superoxide dismutase (MnSOD) may play a key role in affecting PCa. With the Ala variant, *MnSOD* polymorphisms was reported functioned in influencing transportation of the enzyme into mitochondria, accounting for a more efficient importation [36]. Hence, susceptibility of cancers may also be modified by variants in *MnSOD* though improving the gene expression. And the mechanisms are still warranted further investigation.

In present meta-analysis, which included 4,934 PCa cases and 7,697 controls from 17 publications encompassing 18 case-control studies, investigated the association between Ala-9Val and Ala-16Val polymorphisms in *MnSOD* promoter and PCa risk. Considering pooled ORs in overall comparison models, for Ala-9Val polymorphism, a significantly increased risk of PCa was identified in the allelic, dominant and homozygote models, while no significant association was identified for Ala-16Val polymorphism. Besides, in the subgroup analysis stratified by genotyping method, Ala-9Val polymorphism was significantly related to an increased risk of PCa in PCR-RFLP group in allelic and dominant models. When the stratification analysis was conducted based on HWE status, we found that Ala-16Val polymorphism was statistically related to an increased risk of PCa among these studies which were not inconsistent with HWE. Absolutely, deviation from HWE probably

Table 3. Methodological quality of the included studies according to the Newcastle-Ottawa Scale

Polymorphism	Author	Ethnicity	Adequacy of Case Definition	Representativeness of the Cases	Selection of Controls	Definition of Controls	Comparability Cases/Controls	Ascertainment of Exposure	Same Method of Ascertainment	Non-response rate
Ala-9Val	Ergen *et al.*	Caucasian	*	*	NA	NA	**	*	*	*
	Arsova *et al.*	Caucasian	*	*	NA	*	**	*	*	*
	Kucukgergin *et al.*	Caucasian	*	*	NA	*	**	*	*	*
	Eken *et al.*	Caucasian	*	*	*	*	**	*	*	*
	Parlaktas *et al.*	Caucasian	*	*	*	*	**	*	*	*
Ala-16Val	Woodson *et al.*	Caucasian	*	*	NA	*	**	*	*	*
	Li *et al.*	Caucasian	*	*	*	*	**	*	*	*
	Taufer *et al.*	Caucasian	*	*	*	*	**	*	*	*
	Choi *et al.*	Caucasian	*	*	NA	*	**	*	*	*
	Kang *et al.*	Afican	*	*	*	NA	**	*	*	*
	Kang *et al.*	Caucasian	*	*	*	NA	**	*	*	*
	Choi *et al.*	Mixed	*	*	NA	*	**	*	*	*
	Mikhak *et al.*	Caucasian	*	*	NA	*	**	*	*	*
	Iguchi *et al.*	Caucasian	*	*	NA	NA	**	*	*	*
	Bica *et al.*	Caucasian	*	*	NA	*	**	*	*	*
	Iguchi *et al.*	Mixed	*	*	NA	NA	**	*	*	*
	Hemelrijck *et al.*	Caucasian	*	*	NA	*	**	*	*	*
	Oskina *et al.*	Caucasian	*	*	NA	*	**	*	*	*

This table identifies 'high' quality choices with a 'star'. A study can be awarded a maximum of one star for each numbered item within the Selection and Exposure categories. A maximum of two stars can be given for Comparability. *, Yes; NA, not applicable. (http://www.ohri.ca/programs/clinical epidemiology/oxford.htm).

caused by methodological errors may affect the results in original articles [22, 27]. This discrepancy may also be attributed to the different methods implemented to calculate *P* values of HWE (R vs. SPSS).

Till now, four meta-analysis that focused on the association between *MnSOD* polymorphisms and risk of PCa were published. Compared with those previously published meta-analysis, our study have several advantages. Firstly, the study by Mao *et al.* [37] only focus on Ala-16Vla polymorphism in *MnSOD* and PCa risk, while Wang *et al.* [38] performed their study only concerning Ala-16Vla polymorphism. But in our study, we focused on both of them. Secondly, in order to obtain more powerful and convincing evidence, more new studies were added and the results were adjusted according to Bonferroni corrections [39]. At the same time, some mistakes were identified in the study conducted by Mao *et al.* [37], which were revised in our study. Thirdly, Sun *et al.* [40] conducted a systematic analysis discussing the association of *MnSOD* polymorphisms and risk of diverse cancers, such as prostate, esophageal, and lung cancer, while in our work, we only concentrated on PCa, aiming to expand the depth of results and enrich the details of analysis. Fourthly, in our meta-analysis significant association was found between the Ala-9Val polymorphism in *MnSOD* and PCa risk, whereas this conclusion was different from the study by Li and his colleagues [41]. Finally, subgroup analyses stratified by age at diagnosis and disease status (aggressive vs. non-aggressive prostate cancer) were performed by, making contents more diversified.

Certainly, several limitations should be acknowledged. First, the number of studies and the sample sizes were relatively small, contributing to an insufficient power to identify slightly influence of these polymorphisms on PCa risk. Second, the ethnicity of most studies were Caucasians and few studies were conducted in Asians or Africans, therefore, we were unable to perform a stratification analysis based on ethnicity to identify underlying risk in these ethnicities. Third, the studies included all types of PCa, which may be familial, inherited or sporadic PCa. It could affect our conclusion. Fourthly, conclusion was not totally adjusted. Various details should be adjusted such as family history, lifestyle and environmental factors, so as to acquire more accurate results.

In conclusion, our meta-analysis suggests that the *MnSOD* Ala-9Vla polymorphism may be a risk factor for PCa. Future well-designed studies with larger sample sizes, well controlled confounding factors are warranted to further verify our findings.

Acknowledgements

The present work was supported by Shenzhen Science and Technology Innovation Committee (Grant number: JCYJ20140414170821327).

Disclosure of conflict of interest

None.

Authors' contribution

X.Z. and Y.W., accessed information from literature for this article. M.Z., H.W. and Z.C. contributed towards writing, discussing, and editing the manuscript.

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Table S1. Subgroup analyses according to the clinical characteristics

Status	P_H	P_Z	$P_{(A)}$	Random	Fixed	Comparison	Subgroup	N	Status	P_H	P_Z	$P_{(A)}$	Random	Fixed
Ala-16Val														
Aggressive	0.96	0.327	1.000	1.057 (0.946-1.180)	1.057 (0.946-1.180)	B VS. A	Overall	4	Non-Aggressive	0.946	0.321	1.000	1.051 (0.952-1.160)	1.051 (0.952-1.160)
	0.868	0.371	1.000	1.062 (0.931-1.210)	1.062 (0.931-1.210)	B VS. A	Caucasian	3		0.973	0.570	1.000	1.034 (0.922-1.159)	1.034 (0.922-1.159)
	0.82	0.695	1.000	1.034 (0.874-1.224)	1.034 (0.874-1.224)	B VS. A	PCR-RFLP	2		0.842	0.763	1.000	1.026 (0.869-1.211)	1.026 (0.869-1.211)
	0.711	0.339	1.000	1.074 (0.928-1.243)	1.074 (0.928-1.243)	B VS. A	Other methods	2		0.653	0.312	1.000	1.065 (0.942-1.204)	1.065 (0.942-1.204)
	0.913	0.3	1.000	1.064 (0.946-1.196)	1.064 (0.946-1.196)	B VS. A	P-B	3		0.831	0.348	1.000	1.051 (0.947-1.165)	1.051 (0.947-1.165)
	0.827	0.794	1.000	1.025 (0.845-1.245)	1.026 (0.846-1.245)	BA VS. AA	Overall	4		0.867	0.303	1.000	1.094 (0.920-1.301)	1.095 (0.921-1.302)
	0.734	0.95	1.000	0.992 (0.788-1.248)	0.993 (0.789-1.249)	BA VS. AA	Caucasian	3		0.986	0.641	1.000	1.049 (0.859-1.281)	1.049 (0.859-1.281)
	0.805	0.675	1.000	1.065 (0.792-1.432)	1.065 (0.793-1.431)	BA VS. AA	PCR-RFLP	2		0.876	0.716	1.000	1.055 (0.790-1.410)	1.055 (0.790-1.410)
	0.396	0.986	1.000	0.996 (0.771-1.288)	0.998 (0.772-1.289)	BA VS. AA	Other methods	2		0.436	0.311	1.000	1.116 (0.900-1.386)	1.118 (0.901-1.386)
	0.642	0.782	1.000	1.029 (0.837-1.263)	1.029 (0.838-1.264)	BA VS. AA	P-B	3		0.722	0.287	1.000	1.103 (0.919-1.323)	1.104 (0.920-1.324)
	0.964	0.559	1.000	1.055 (0.880-1.265)	1.056 (0.880-1.266)	BA+BB VS. AA	Overall	4		0.878	0.256	1.000	1.098 (0.932-1.293)	1.099 (0.934-1.294)
	0.914	0.747	1.000	1.036 (0.836-1.284)	1.036 (0.836-1.284)	BA+BB VS. AA	Caucasian	3		1.000	0.573	1.000	1.056 (0.875-1.274)	1.056 (0.875-1.274)
	0.784	0.65	1.000	1.066 (0.808-1.406)	1.066 (0.808-1.406)	BA+BB VS. AA	PCR-RFLP	2		0.978	0.700	1.000	1.055 (0.804-1.384)	1.055 (0.804-1.384)
	0.657	0.704	1.000	1.047 (0.823-1.332)	1.048 (0.824-1.333)	BA+BB VS. AA	Other methods	2		0.460	0.258	1.000	1.123 (0.915-1.378)	1.125 (0.917-1.379)
	0.888	0.535	1.000	1.063 (0.876-1.289)	1.063 (0.876-1.290)	BA+BB VS. AA	P-B	3		0.725	0.256	1.000	1.104 (0.929-1.311)	1.105 (0.930-1.312)
	0.967	0.337	1.000	1.113 (0.894-1.385)	1.113 (0.894-1.385)	BB VS. AA	Overall	4		0.941	0.320	1.000	1.106 (0.907-1.348)	1.106 (0.907-1.348)
	0.88	0.385	1.000	1.120 (0.867-1.449)	1.120 (0.867-1.449)	BB VS. AA	Caucasian	3		0.977	0.571	1.000	1.068 (0.850-1.342)	1.068 (0.850-1.342)
	0.808	0.696	1.000	1.068 (0.766-1.490)	1.068 (0.766-1.489)	BB VS. AA	PCR-RFLP	2		0.871	0.767	1.000	1.051 (0.758-1.457)	1.051 (0.758-1.457)
	0.748	0.351	1.000	1.148 (0.859-1.535)	1.148 (0.859-1.535)	BB VS. AA	Other methods	2		0.638	0.306	1.000	1.139 (0.888-1.460)	1.139 (0.888-1.460)
	0.933	0.306	1.000	1.129 (0.895-1.426)	1.129 (0.895-1.426)	BB VS. AA	P-B	3		0.822	0.342	1.000	1.107 (0.898-1.365)	1.107 (0.898-1.365)
	0.693	0.316	1.000	1.096 (0.918-1.308)	1.095 (0.917-1.306)	BB VS. BA+AA	Overall	4		0.984	0.627	1.000	1.040 (0.887-1.220)	1.040 (0.887-1.220)
	0.549	0.264	1.000	1.127 (0.916-1.387)	1.126 (0.914-1.386)	BB VS. BA+AA	Caucasian	3		0.929	0.716	1.000	1.035 (0.860-1.246)	1.035 (0.860-1.245)
	0.911	0.855	1.000	1.025 (0.784-1.341)	1.025 (0.784-1.341)	BB VS. BA+AA	PCR-RFLP	2		0.749	0.917	1.000	1.014 (0.778-1.322)	1.014 (0.778-1.322)
	0.311	0.238	1.000	1.153 (0.909-1.463)	1.151 (0.911-1.455)	BB VS. BA+AA	Other methods	2		0.996	0.595	1.000	1.056 (0.865-1.289)	1.056 (0.865-1.289)
	0.517	0.285	1.000	1.109 (0.919-1.339)	1.108 (0.918-1.337)	BB VS. BA+AA	P-B	3		0.939	0.688	1.000	1.035 (0.875-1.225)	1.035 (0.875-1.225)
Ala-9Val														
Aggressive	0.05	0.07	1.000	1.758 (0.955-3.237)	1.545 (1.109-2.151)	B VS. A	Overall	3	Non-Aggressive	0.268	0.101	1.000	1.493 (0.897-2.484)	1.437 (0.932-2.216)
	0.018	0.272	1.000	1.775 (0.637-4.943)	1.449 (0.988-2.125)	B VS. A	H-B	2		0.107	0.175	1.000	1.533 (0.674-3.489)	1.403 (0.860-2.288)
	0.834	0.004	1.000	2.333 (1.319-4.126)	2.326 (1.313-4.119)	BA VS. AA	Overall	3		0.411	0.496	1.000	1.264 (0.641-2.493)	1.265 (0.643-2.492)
	0.594	0.016	1.000	2.234 (1.172-4.259)	2.219 (1.161-4.242)	BA VS. AA	H-B	2		0.186	0.491	1.000	1.315 (0.485-3.568)	1.302 (0.614-2.760)
	0.488	0.002	1.000	2.415 (1.393-4.188)	2.402 (1.383-4.173)	BA+BB VS. AA	Overall	3		0.333	0.242	1.000	1.466 (0.750-2.866)	1.462 (0.773-2.765)
	0.488	0.011	1.000	2.321 (1.134-4.749)	2.254 (1.203-4.223)	BA+BB VS. AA	H-B	2		0.138	0.292	1.000	1.511 (0.525-4.350)	1.466 (0.719-2.989)
	0.093	0.119	1.000	3.101 (0.749-12.849)	2.398 (1.141-5.037)	BB VS. AA	Overall	3		0.223	0.165	1.000	2.108 (0.608-7.310)	1.849 (0.776-4.405)
	0.042	0.351	1.000	4.506 (0.191-106.274)	2.077 (0.870-4.958)	BB VS. AA	H-B	2		0.092	0.407	1.000	3.136 (0.21-46.808)	1.706 (0.626-4.646)
	0.072	0.412	1.000	1.673 (0.489-5.726)	1.294 (0.723-2.318)	BB VS. BA+AA	Overall	3		0.323	0.117	1.000	1.816 (0.757-4.354)	1.811 (0.862-3.808)
	0.035	0.522	1.000	2.82 (0.118-67.439)	1.138 (0.572-2.262)	BB VS. BA+AA	H-B	2		0.146	0.228	1.000	2.617 (0.302-22.678)	1.696 (0.719-4.000)
<65	0.071	0.088	1.000	1.764 (0.920-3.383)	1.524 (1.056-2.199)	B VS. A	Overall	3	>65	0.158	0.280	1.000	1.188 (0.770-1.834)	1.183 (0.872-1.605)
	0.032	0.307	1.000	1.757 (0.596-5.179)	1.396 (0.915-2.131)	B VS. A	P-B	2		0.795	0.041	1.000	1.510 (1.016-2.243)	1.510 (1.016-2.243)
	0.233	0.002	1.000	2.782 (1.141-6.786)	2.564 (1.397-4.707)	BA VS. AA	Overall	3		0.994	0.534	1.000	1.173 (0.710-1.937)	1.173 (0.710-1.937)
	0.095	0.166	1.000	3.656 (0.585-22.853)	2.464 (1.259-4.823)	BA VS. AA	P-B	2		0.925	0.595	1.000	1.185 (0.633-2.217)	1.185 (0.634-2.217)
	0.124	0.003	1.000	2.960 (0.996-8.794)	2.394 (1.342-4.271)	BA+BB VS. AA	Overall	3		0.707	0.333	1.000	1.258 (0.790-2.003)	1.258 (0.791-2.003)
	0.056	0.233	1.000	3.549 (0.443-28.435)	2.195 (1.158-4.161)	BA+BB VS. AA	P-B	2		0.958	0.204	1.000	1.452 (0.817-2.584)	1.452 (0.816-2.584)
	0.04	0.213	1.000	3.078 (0.524-18.083)	1.975 (0.882-4.426)	BB VS. AA	Overall	3		0.174	0.265	1.000	1.397 (0.613-3.188)	1.399 (0.775-2.525)
	0.03	0.498	1.000	2.683 (0.154-46.666)	1.461 (0.570-3.744)	BB VS. AA	P-B	2		0.751	0.044	1.000	2.195 (1.016-4.741)	2.199 (1.020-4.741)
	0.131	0.698	1.000	1.325 (0.462-3.799)	1.148 (0.571-2.308)	BB VS. BA+AA	Overall	3		0.089	0.602	1.000	1.252 (0.537-2.917)	1.207 (0.728-1.999)
	0.099	0.959	1.000	1.040 (0.236-4.590)	0.868 (0.373-2.021)	BB VS. BA+AA	P-B	2		0.682	0.048	1.000	1.995 (1.000-3.980)	2.003 (1.007-3.984)

H-B: hospital based; P-B: population based; A: wild type; B: mutated type; PCR-RFLP: Polymerase Chain Reaction-Restriction Fragment Length Polymorphism; P_H: *P* value of heterogeneity; *P* value of Z test; *P* value of Adjusted *P* value.