RESEARCH ARTICLE

The MDM2 SNP309T>G Polymorphism Increases Bladder Cancer Risk among Caucasians: a Meta-analysis

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Abstract

Published studies have evaluated associations between the MDM2 SNP309T>G polymorphism and bladder cancer susceptibility. However, these generated inconsistent results. The aim of the present investigation was to quantify the strength of association between MDM2 SNP309T>G polymorphism and bladder cancer risk by conducting a meta-analysis. We searched PubMed and Embase for related studies that had been published in English before April 1, 2014 and associations were assessed by summarizing the odds ratios (ORs) with the corresponding 95% confidence intervals (CIs). Five case-control studies with a total of 972 cases and 1,012 controls were finally identified to be eligible for the meta-analysis. Overall, the results indicated that there was no significant association between the MDM2 SNP309T>G polymorphism and bladder cancer risk (for the allele model G vs. T: OR=1.08, 95% CI 0.85-1.36, *p*=0.54; for the co-dominant model GG vs. TT: OR=1.20, 95% CI 0.74-1.93, *p*=0.46; for the dominant model GG+GT vs. TT: OR=0.98, 95% CI 0.80-1.20, *p*=0.83; for the recessive model GG vs. GT+TT: OR=1.20, 95% CI 0.83-1.74, *p*=0.33). However, on subgroup analysis by ethnicity, significant associations were found in Caucasians in three models (for the allele model G vs. T: OR=1.41, 95% CI 1.10-1.81, *p*=0.006; for the co-dominant model GG vs. TT: OR=2.16, 95% CI 1.28-3.63, *p*=0.004; for the recessive model GG vs. GT+TT: OR=2.06, 95% CI 1.31-3.22, *p*=0.002). In summary, the present meta-analysis provides evidence that the genotype for the MDM2 SNP309T>G polymorphism may be associated with genetic susceptibility to bladder cancer among Caucasians.

Keywords: Bladder cancer - MDM2 - polymorphism - meta-analysis - ethnic variation

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Introduction

According to the GLOBOCAN 2012 report from the World Health Organization's International Agency for Research on Cancer (IARC), bladder cancer is the ninth most common cancer in the world and there are nearly 429793 new cases and 165068 deaths die of bladder cancer in 2012 (GLOBOCAN2012, IARC). The majority of bladder cancer occurs in males and the highest incidence rates are found in the countries of Europe, North America, and Northern Africa (Jemal et al. 2011). Although the development of bladder cancer is associated smoking, occupational exposure and chronic infection with Schistosoma hematobium, not all of these populations will ever develop bladder cancer (Mommsen and Aagaard 1983; Olfert et al. 2006; Murta-Nascimento et al. 2007). The genetic susceptibility, environmental and lifestyle factors also play an important role in the development of the disease (McConkey et al. 2010).

P53 tumor suppressor pathway plays an important role in cell cycle, apoptosis and inhibition of angiogenesis. The mutation that inactivates the p53 gene has been found in at least half of all human cancers (Greenblatt et al. 1994). The human mouse double minute 2 (MDM2) gen located on chromosome 12q13 to 14 with a genomic length of 34 kb is a major negative regulator of the p53 network (Kubbutat et al. 1997). The over expression of MDM2 can decrease the level of p53 protein and eventually results in the dysfunction of the P53 pathway, in turn, contribute to the development of cancers. A functional T to G polymorphism at nucleotide 309 in the promoter region of the MDM2 gene has been identified (Whibley et al. 2009). The G allele of SNP309 confers an increased binding affinity to the Sp1 transcriptional activator which can increase transcription of the MDM2 gene (Pietsch et al. 2006). Thus the SNP309 GG and TG genotypes seem to express higher levels of MDM2 protein than those with the TT genotypes, eventually causes a relative decrease of p53 protein. This highly suggests that MDM2 variant may be a potential modulator of cancer susceptibility.

Published studies have evaluated the association between MDM2 SNP309T>G polymorphism and bladder

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cancer susceptibility (Onat et al. 2006; Horikawa et al. 2008; Wang et al. 2008; Gangwar and Mittal 2010; Hitzenbichler et al. 2014). However, these studies showed inconsistent results. To derive a more precise estimation of this association, a meta-analysis was performed to estimate the effect of MDM2 SNP309T>G polymorphism on bladder cancer risk.

Materials and Methods

Identification and eligibility of relevant studies

Electronic searches of PubMed and Embase databases (up to October 2013) were performed for all possible publications on the association between MDM2 SNP309T>G polymorphism and bladder cancer risk. The initial search terms were "MDM2," "rs2279744," "T309G," "polymorphism," "variant," "mutation," "bladder," "cancer" and "carcinoma." The search was limited to English-language studies. In addition, the reference lists of identified studies were manually checked to include other potentially eligible studies.

Inclusion and exclusion criteria

Studies were selected according to the following inclusion criteria: case-control studies in design; investigating the association between MDM2 SNP309T>G polymorphism and bladder cancer risk; with genotype distribution data to calculate combined ORs and 95% CIs. The major exclusion criteria were: no control population; abstract, comment, and review; duplicate of earlier publication and no usable genotype frequency data.

Data extraction

Two investigators (Huai-Gao Wang and Qing-Yun Wu) independently extracted the data according to the listed inclusion and exclusion criteria above. Disagreements were resolved by discussion among the investigators. The following information was extracted from each eligible study: name of the first author, year of publication, country of origin, ethnicity, numbers of genotyped cases and controls and genotype frequency in cases and controls. Different ethnic descents were categorized as Asian and Caucasian.

Statistical analysis

In this meta-analysis, the association strength between MDM2 SNP309T>G polymorphism and bladder cancer risk was measured by the pooled odds ratios (ORs) with 95% confidence intervals (CIs). The alleles model (G vs. T), the co-dominant model (GG vs. TT), the dominant model (GG+GT vs. TT), and the recessive model (GG vs. GT+TT) were performed respectively. Subgroup analysis by ethnicity was also performed. The Z-test was performed to assess the significance of the pooled ORs and $p \leq 0.05$ was considered as statistically significant. The Q test and I^2 test were performed to assess the heterogeneity between studies. According to the heterogeneity, the fixed-effect or random-effect model would be used to calculate the pooled odds ratios (ORs) with 95% confidence intervals (CIs). If $P_Q < 0.10$ and $I^2 > 50\%$, indicating the presence of heterogeneity, the random-effects model (the DerSimonian and Laird method) was used to calculate the pooled OR. Otherwise, the fixed-effects model (Mantel-Haenszel) was selected. The Hardy-Weinberg equilibrium (HWE) was determined in the control groups. Sensitivity analyses were performed to identify one single study' effect on pooled results and test the reliability of results. Publication bias among the included studies was investigated by Begger's funnel plots and Egger' linear regression test, and a $p<0.05$ was considered as statistically significant. Data analysis was performed using the software STATA (version 12.0) and Review Manager (version 5.0).

Results

Characteristics of published studies

Based on the inclusion and exclusion criteria, five studies (972 cases and 1012 controls) published from 2006 to 2014 were identified to be eligible studies (Onat et al. 2006; Horikawa et al. 2008; Wang et al. 2008; Gangwar and Mittal 2010; Hitzenbichler et al. 2014). All of the studies were published in English. The characteristics of all the studies that were included in the meta-analysis were listed in Table 1. There were two studies on Caucasians, three studies on Asians. Among them, one study was from Turkey (Onat et al. 2006), one study was from Japan (Horikawa et al. 2008), one study was from China (Wang et al. 2008), one study was from India (Gangwar and Mittal

Table 1. Association Between Individual Study Characteristics and MDM2 T309G Polymorphism

Reference	Year	Area	Ethnicity	Cases	Controls	Cases			Controls			HWE(control)
						TT	TG	GG	TT	TG	GG	*p*
Gangwar et al.	2010	India	Asian	212	250	70	89	53	62	113	75	0.162
Hitzenbichler et al.	2014	Germany	Caucasians	224	140	75	101	48	51	70	19	0.594
Horikawa et al.	2008	Japan	Asian	227	266	44	116	67	55	132	79	1.000
Onat et al.	2006	Turkey	Caucasians	75	103	13	36	26	29	57	17	0.317
Wang et al.	2008	China	Asian	234	253	62	121	51	64	134	55	0.379

Table 2. Meta-analysis of MDM2 T309G Polymorphism and Bladder Cancer Risk in Each Subgroup

Category	G vs. T			GG vs. TT			TT			GT+TT		
	OR (95%CI)	P_{OR}	I^2 (%)	OR (95%CI)	P_{OR}	I^2 (%)	OR (95%CI)	P_{OR}	I^2 (%)	OR (95%CI)	P_{OR}	I^2 (%)
Ethnicity												
Asian	0.92 (0.79,1.06)	0.26	0.28	0.85 (0.64,1.14)	0.28	0.17	0.87 (0.68,1.11)	0.25	0.26	0.92 (0.73,1.16)	0.48	0
Caucasian	1.41 (1.10,1.81)	0.006	0.45	2.16 (1.28,3.63)	0.004	0.33	1.30 (0.90,1.90)	0.17	0.22	2.06 (1.31,3.22)	0.002	0

**CI* 95% confidence intervals, *OR* odds ratio

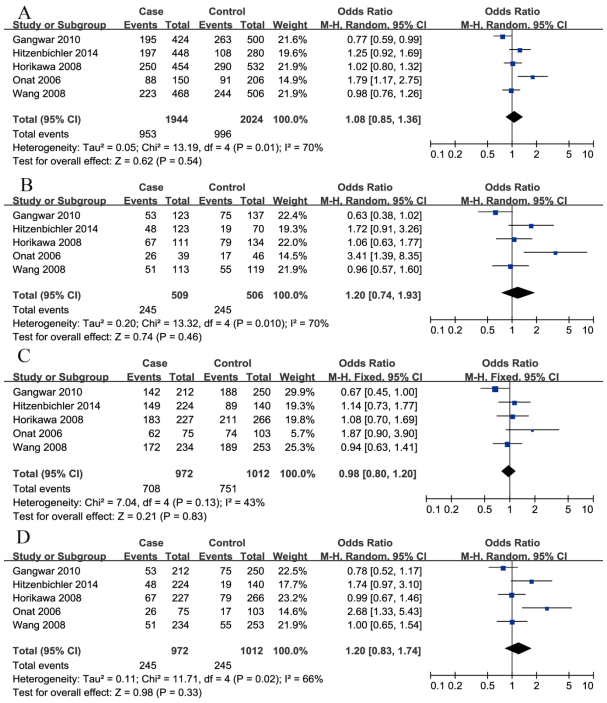

Figure 1. Forest Plot of the Meta-analysis on the Association Between MDM2 SNP309T>G Polymorphism and Bladder Cancer Risk. No significant association was found between MDM2 SNP309T>G polymorphism and bladder cancer risk in all genetic models. A) the allele model (G vs. T); B) the co-dominant model (GG vs. TT); C) the dominant model (GG+GT vs. TT); D) the recessive model (GG vs. GT+TT). Error bars indicate 95% CI. Solid squares represent each study in the meta-analysis. Solid diamonds represent pooled OR.

2010), one study was from Germany (Hitzenbichler et al. 2014). The genotype distributions of all studies in the control groups conformed to the HWE equilibrium.

Meta-analysis results

Overall, the combined results indicated that MDM2 SNP309T>G polymorphism was not significantly associated with bladder cancer risk in all genetic models (for the allele model G vs. T: OR=1.08, 95% CI 0.85-1.36, p=0.54; for the co-dominant model GG vs. TT: OR=1.20, 95% CI 0.74-1.93, p=0.46; for the dominant model GG+GT vs. TT: OR=0.98, 95% CI 0.80-1.20, p=0.83; for the recessive model GG vs. GT+TT: OR=1.20, 95% CI 0.83-1.74, p=0.33) (Figure 1). Subgroup analyses by ethnicity further showed that there was also no significant association between MDM2 SNP309T>G polymorphism and susceptibility to bladder cancer in Asians (for the allele model G vs. T: OR=0.92, 95% CI 0.79-1.06, p=0.26; for the co-dominant model GG vs. TT: OR=0.85, 95% CI 0.64-1.14, p=0.28; for the dominant model GG+GT vs. TT: OR=0.87, 95% CI 0.68-1.11, p=0.25; for the recessive model GG vs. GT+TT: OR=0.92, 95% CI 0.73-1.16, p=0.48) (Table 2). However, significant associations were found in

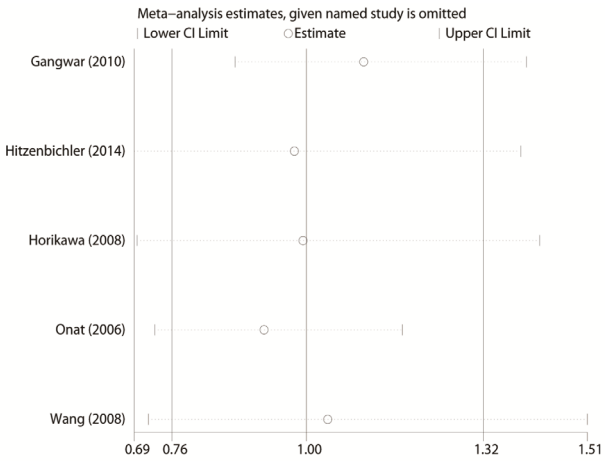

Figure 2. Influence Analysis for the Dominant Model (GG+GT vs. TT) in the Overall Meta-analysis. This figure shows the influence of individual studies on the summary OR. The middle vertical axis indicates the overall OR and the two vertical axes indicate its 95% CI. Every hollow round indicates the pooled OR when the left study is omitted in this meta-analysis. The two ends of the dotted lines represent the 95% CI.

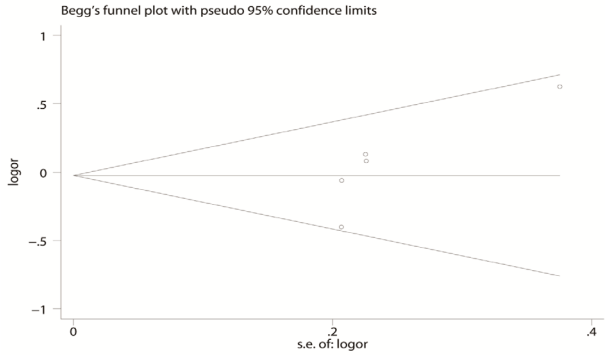

Figure 3. Begger's Funnel Plots for Publication Bias of the Meta-analysis on the Association Between MDM2 SNP309T>G Polymorphism and Bladder Cancer Risk in Dominant Model (GG+GT vs. TT). Begger's funnel plots for publication bias of the meta-analysis on the association between MDM2 SNP309T>G polymorphism and bladder cancer risk in dominant model (GG+GT vs. TT).

Caucasians in three models (for the allele model G vs. T: OR=1.41, 95% CI 1.10-1.81, p=0.006; for the co-dominant model GG vs. TT: OR=2.16, 95% CI 1.28-3.63, p=0.004; for the recessive model GG vs. GT+TT: OR=2.06, 95% CI 1.31-3.22, p=0.002) (Table 2). In the sensitivity analysis, we excluded one single study from the overall pooled analysis each time to check the influence of the removed data set to the overall ORs. The pooled ORs and 95 % CIs were not significantly changed when any part of the study was omitted, which indicated that omission of any single study did not have significant impact on the combined ORs (Figure 2).

Publication bias

Begg's funnel plot and Egger' linear regression

test were performed to estimate the publication bias of the meta-analysis. The shapes of the funnel plots of the all genetic did not reveal any evidence of obvious asymmetry in all comparison models, and Egger's test further indicated no considerable publication bias in this meta-analysis (Figure 3).

Discussion

To the best of our knowledge, this is the first meta-analysis to assess the association between MDM2 SNP309T>G polymorphism and bladder cancer risk. We aimed to bring together the results of all eligible case-control studies to quantify the strength of association between MDM2 SNP309T>G polymorphism and bladder cancer risk. The present meta-analysis included five studies with a total of 972 cases and 1012 controls on MDM2 SNP309T>G polymorphisms were finally identified to be eligible studies. Our result suggested that there was no significant association between MDM2 SNP309T>G polymorphism and bladder cancer risk. Sensitivity analysis also showed that omission of any single study did not have significant impact on the combined ORs, which adds robustness to our finding. We also performed a subgroup analysis by ethnicity and found that there was also no significant association between MDM2 SNP309T>G polymorphism and bladder cancer risk in Asians, however there was a significant difference was found in Caucasians. There is a difference between Asians and Caucasians in the 309G minor allele frequency: 0.51 in Asians and 0.45 in Caucasians. The different genetic backgrounds may contribute to the discrepancy.

The formation of bladder cancer is a complex process associated with the interaction of environmental factors and genetic susceptibility (Knowles 2008; Fang et al. 2013). Identifications of risk factors of bladder cancer can help us find some effective and preventive interventions to decrease the incidence of the disease. As a tumor suppressor gene, p53 can inhibit abnormal cell proliferation and promote tumor cells apoptosis. Decreased expression of p53 can possibly results in the dysfunction of cell-cycle checkpoint control and eventually contributed to cancer development (Levine et al. 1991). MDM2 is a negative regulator of p53 which can result in the reduction of p53 expression. Considering the important role of MDM2 in P53 pathway, the polymorphisms in the MDM2 is supposed to be associated with risk of various types of cancers. Ma et al. suggested that a significant association between the MDM2 SNP309 T/G polymorphism and liver cancer risk, the MDM2 SNP309 G allele contributing to increased risk in both Asians and Caucasians in a graded, dose-dependent fashion (Ma et al. 2012). Zhao et al. suggests that MDM2 SNP309 polymorphism may increase the risk to breast cancer in Asian and African population (Zhao et al. 2012). Wang et al. provided evidence for a major role of MDM2 rs2279744 polymorphism in the carcinogenesis of endometrial cancer, especially among Caucasian populations (Wang et al. 2014b). Wang et al. indicated that MDM2 polymorphisms have some effects on the risk of osteosarcoma (Wang et al. 2014a). Cao et al. suggested that the MDM2 SNP309 polymorphism is a risk factor for the development of colorectal cancer, particularly among Asians(Cao et al. 2012).Ma et al. and Tian et al. provided evidence of the association between MDM2 SNP309 polymorphism and gastric cancer risk (Ma et al. 2013; Tian et al. 2013). The findings above suggest that MDM2 SNP309 polymorphism was a risk factor for certain types of cancer development and the different ethnicity may be associated with genetic susceptibility of bladder. In addition, published studies also have evaluated the association between MDM2 SNP309T>G polymorphisms and risk of bladder cancer. Due to the relatively small sample size and different population, these studies showed inconsistent results. To clarify the effect of MDM2 SNP309T>G polymorphism on the risk of bladder cancer, we performed this meta-analysis. We found that MDM2 SNP309T>G polymorphism may be associated with genetic susceptibility of bladder cancer among Caucasians.

In this meta-analysis, obvious heterogeneity was observed for the association between MDM2 SNP309T>G polymorphism and bladder cancer risk. However, all of the I^2 values were less than 50% and P_Q were greater than 0.1 in the subgroup by ethnicity. These results indicated that the ethnicity might be the major source of the heterogeneity for the association between MDM2 SNP309T>G polymorphism and bladder cancer risk.

There are some limitations to this meta-analysis. Firstly, the searches in our study were limited in the English literature. Besides, due to the limited data, we did not carry out subgroup analysis to other factors, which may participate in the progression of disease, such as age, gender and other lifestyle. Secondly, the sample sizes of cases and controls were small, especially in the subgroup of Caucasians. There were only two studies on Caucasians and it might result in the false positive findings in the meta-analysis. Therefore, the positive results of the Caucasians should be interpreted with caution. Thirdly, this meta-analysis was just based on unadjusted estimates which might bias the results. Finally, obvious heterogeneity was observed for the association between MDM2 SNP309T>G polymorphism and bladder cancer risk. Although the ethnicity contributed to the potential heterogeneity, the populations, habits, lifestyle, geographical location and study designs may also contribute to the heterogeneity.

In summary, this meta-analysis suggested that MDM2 SNP309T>G polymorphism may be associated with genetic susceptibility of bladder cancer among Caucasians based on the current published studies. However, future studies with large sample sizes, gene-gene, gene-environment interactions and well-designs are needed to give a more reliable estimation of the association with the bladder cancer susceptibility.

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