RESEARCH ARTICLE

Association between the NQO1 C609T Polymorphism with Hepatocellular Carcinoma Risk in the Chinese Population

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Abstract

Background: Associations between the NQO1 C609T polymorphism and hepatocellular carcinoma (HCC) risk are a subject of debate. We therefore performed the present meta-analysis to evaluate links with HCC susceptibility. Materials and Methods: Several major databases (PubMed, EBSCO), the Chinese national knowledge infrastructure (CNKI) and the Wanfang database were searched for eligible studies. Crude odds ratios (ORs) with 95% confidence intervals (CIs) were used to measure the strength of associations. Results: A total of 4 studies including 1,325 patients and 1,367 controls were identified. There was a significant association between NQO1 C609T polymorphism and HCC for all genetic models (allelic model: OR=1.45, 95%CI=1.23-1.72, $p<0.01$; additive model: OR=1.96, 95%CI=1.57-2.43, $p<0.01$; dominant model: OR=1.62, 95%CI=1.38-1.91, $p<0.01$; and recessive model: OR=1.53, 95%CI=1.26-1.84, $p<0.01$). On subgroup analysis, similarly results were identified in Asians. For Asians, the combined ORs and 95% CIs were (allelic model: OR=1.50, 95%CI=1.24-1.82, $p<0.01$; additive model: OR=2.11, 95%CI=1.48-3.01, $p<0.01$; dominant model: OR=1.69, 95%CI=1.42-2.02, $p<0.01$; and recessive model: OR=1.59, 95%CI=1.16-2.19, $p<0.01$). Conclusions: The current meta-analysis suggested that the NQO1 C609T polymorphism could be a risk factor for developing HCC, particularly in the Chinese population.

Keywords: Hepatocellular carcinoma - NQO1 - polymorphism - meta-analysis - Chinese

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Introduction

Hepatocellular carcinoma (HCC) is the fifth most common cause of cancer and the third leading cause of cancer-related death worldwide (El-Serag and Rudolph, 2007; de Lope et al., 2012). Nearly 700,000 new cases of HCC are diagnosed each year and 600,000 deaths are recorded on the world (Farazi and DePinho, 2006). More than 75% of these cases occur in the Asia-Pacific region and China alone accounts for 55% cases of HCC worldwide (Parkin, 2001; Yuen et al., 2009).

It is mostly important to identify risk factors for HCC, and aim at early develop interventions and promote our understanding of the complicated pathologic mechanisms of this disease. Chronic hepatitis B virus (HBV) and hepatitis C virus (HCV) infection is common environmental factors cause HCC. Moreover, other environmental risk factors including alcohol intake, cigarette smoking, and exposure to aflatoxin B1 are also the risk factor for HCC. However, only a fraction of HBsAg carriers eventually develop HCC and only 2.5% of HCV infected individuals develop HCC later in life (Bowen and Walker, 2005). Thus, host genetic factors may affect the development of HCC. Among the genetic factors, some single nucleotide polymorphisms (SNPs) have been regarded as HCC risk factors (Kato et al., 2005; Kim and Lee, 2005; Liu et al., 2013).

The cytosolic enzyme NAD(P)H: quinone oxidoreductase detoxification of environmental carcinogens(Dinkova-Kostova and Talalay, 2010; Siegel et al., 2012). It can catalyze the two-electron reduction of quinoid compounds and prevent the production of semiquinone free radicals and reactive oxygen species, thus protesting cells from oxidative damage(Siegel et al., 2012). NQO1 polymorphism is a cytosine(C) to thymine(T) transition at nucleotide position 609 exon 6 of NQO1 cDNA that encodes for a proline(P) to serine(S) substitution at position 187 in amino acid sequence of the protein (Traver et al., 1997). NQO1 C609T polymorphism is linked to the enzymatic activity to NQO1 and may affect host's susceptibility to HCC by changing the enzymatic activity. There were several studies performed to detect the association between NQO1 C609T polymorphism and risk of HCC, but no consistent results were reported (Akkiz et al., 2010; Tan et al., 2012; Liu et al., 2013; Wang et al., 2013). We thus conducted a meta-analysis to assess the association between NQO1 C609T polymorphism and HCC risk.

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Materials and Methods

Identification and eligibility of relevant studies

We conducted a computer-assisted search with PubMed, EBSCO, the Chineses national knowledge infrastructure (CNKI), and the Wanfang databases to identify relevant published studies. We searched the databases from inception through 1 May, 2014 with the following keyword combinations: "hepatocellular carcinoma", "HCC", "NQO1 C609T", "rs1800566", and "polymorphism". Manual searches of reference lists from applicable articles were conducted to identify any studies that may have been missed by the computer-assisted search. The search was not restricted by language, but the research had to be conducted on human subjects. This study was conducted in accordance with the PRISMA guidelines (Moher et al., 2009).

Studies in this meta-analysis met the following inclusion criteria: (a) detected the relationship between the NQO1 C609T polymorphism and HCC risk; (b) case control study; (c) provided available genotype data to assess the odds ratios (ORs) and 95% confidence intervals (CIs).

Data extraction

The following data were extracted from the included studies: first author's name, year of publication, country of participants, the number of cases and controls, the number of cases and controls for each polymorphism genotype, and the P value for Hardy-Weinberg equilibrium (HWE). Two investigators extracted the required information from all included studies independently according to the inclusion criteria listed above. Discrepancies were resolved by discussion between two investigators.

Statistical analysis

We performed the association between NQO1 C609T polymorphism and the risk of HCC, using allelic model (T *vs* C), additive model (TT *vs* CC), dominant model (TT+TC *vs* CC), and recessive model (TT *vs* TC+CC). The strength of association between NQO1 C609T polymorphism and hepatocellular carcinoma risk was assessed by ORs and corresponding 95% CIs. A chi-square based Q test was conducted to assess heterogeneity (Lau et al., 1997), and the I^2 was also calculated for heterogeneity analysis (Higgins and Thompson, 2002).

To assess the quality of studies, HWE was performed in the controls using the Fisher's exact test. A P value<0.05 was considered statistically significant, and studies with deviation from HWE were defined as low quality studies. If the result of the Q test was I^2<50%, the fixed-effects model was used; otherwise, the random-effects model was adopted. Publication bias was tested by funnel plot and Egger's linear regression test (p<0.05 was considered representative of statistically significant publication bias) (Egger et al., 1997).

All analyses were performed in STATA 11.0 and RevMan 5 software. All the tests were two-sided and the significant level was 0.05.

Results

Study selection and characteristics

We firstly identified 154 studies through the literature search. However, after reviewing of abstracts and full tests, 150 were excluded, and 4 articles were finally included into the meta-analysis (Akkiz et al., 2010; Tan et al., 2012; Liu et al., 2013; Wang et al., 2013) (Figure 1). The publication of Wang et al. (2013) presented two separate case-control studies, each study in one publication was considered separately for pooling analysis. Therefore, 4 publications including 5 studies were involved in this meta-analysis. Data were collected from 1325 patients and 1367 controls, and the main characteristics of the studies were summarized in Table 1. Among the 5 studies, 4 focused on Asians, and 1 on Caucasian.

Pooled analysis

Table 2 showed the results for the association between NQO1 C609T polymorphism and the risk of HCC. Overall, the combined results showed a significant association between NQO1 C609T polymorphism and HCC for all genetic models (allelic model: OR=1.45, 95%CI=1.23-1.72, p<0.01; additive model: OR=1.96, 95%CI=1.57-2.43, p<0.01; dominant model: OR=1.62, 95%CI=1.38-1.91, p<0.01; and recessive model: OR=1.53, 95%CI=1.26-1.84, p<0.01) (Figure 2-5). In the stratified analyses by ethnicity (Asian or Caucasian), we found that the NQO1 C609T polymorphism significantly increased the risk of HCC in Asian populations in the all genetic models (allelic model: OR=1.50, 95%CI=1.24-1.82, p<0.01; additive model: OR=2.11, 95%CI=1.48-3.01, p<0.01; dominant model: OR=1.69, 95%CI=1.42-2.02, p<0.01; and recessive model: OR=1.59, 95%CI=1.16-2.19, p<0.01) (Table 2).

Heterogeneity analysis

We used Q statistic and I^2 index to assess the heterogeneity between studies. As shown in Table 2, there was some heterogeneity between studies in some comparisons. We conducted a subgroup analysis, and did not identify the source of heterogeneity in the overall meta-analysis.

Publication bias

Begg's funnel plot and Egger's tests were performed

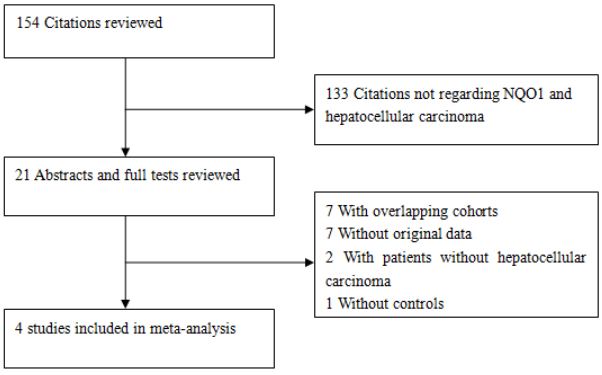

Figure 1. Flow Chat of the Study Extraction

Table 1. Characteristics of the Studies Included in the Meta-analysis

ID	Study	Year	Country	Ethnitity	Cases/ Controls	Case			Control			*p*-HWE Controls
						TT	TC	CC	TT	TC	CC	
1	Akkiz H	2010	Turkish	Caucasian	167/167	10	71	86	9	62	96	0.81
2	Tang SK	2012	China	Asian	400/400	104	201	95	73	175	152	0.07
3	Wang WW1	2013	China	Asian	136/123	54	58	24	22	60	41	0.1
4	Wang WW2	2013	China	Asian	146/151	41	71	34	39	60	52	0.01
5	Liu F	2013	China	Asian	476/526	118	220	138	100	235	191	0.07

Table 2. Meta-analysis of the Association between the NQO1 C609T Polymorphism and HCC

polymorphism	Studies	No. of studies	Test of association			Test of heterogeneity		
			OR	95%CI	*p*-value	Model	*p*-value	I^2 (%)
T *vs* C	Overall	5	1.45	1.23-1.72	<0.01	R	0.08	51
	Asian	4	1.5	1.24-1.82	<0.01	R	0.07	57
TT *vs* CC	Overall	5	1.96	1.57-2.43	<0.01	F	0.12	46
	Asian	4	2.11	1.48-3.01	<0.01	R	0.09	53
dominant model	Overall	5	1.62	1.38-1.91	<0.01	F	0.24	28
	Asian	4	1.69	1.42-2.02	<0.01	F	0.25	27
recessive model	Overall	5	1.53	1.26-1.84	<0.01	F	0.11	47
	Asian	4	1.59	1.16-2.19	<0.01	R	0.07	58

Table 3. Egger's Linear Regression Test to Measure the Funnel Plot Asymmetric

Comparisons	Study	Y axle intercept: a(95%CI)	t	P value
T *vs* C	Overall	-15.59	0.41	0.71
TT *vs* CC	Overall	-12.86	0.26	0.81
dominant model	Overall	-12.64	0.65	0.56
recessive model	Overall	-12.93	0.18	0.87

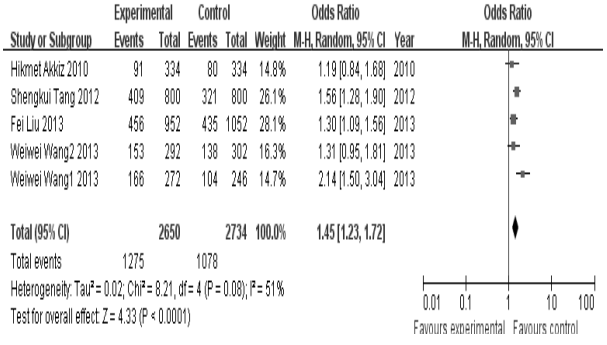

Figure 2. Forest Plots for T *vs* C of the Association between NQO1 C609T Polymorphism and HCC

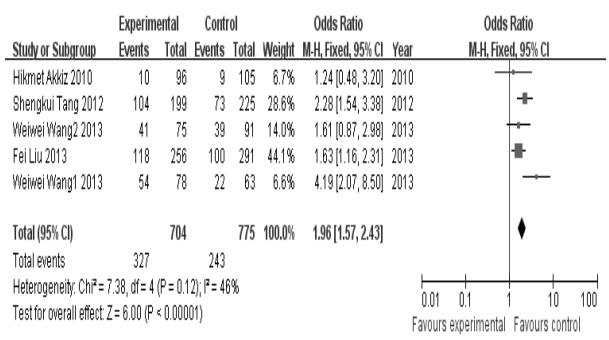

Figure 3. Forest Plots for TT *vs* CC of the Association between NQO1 C609T Polymorphism and HCC

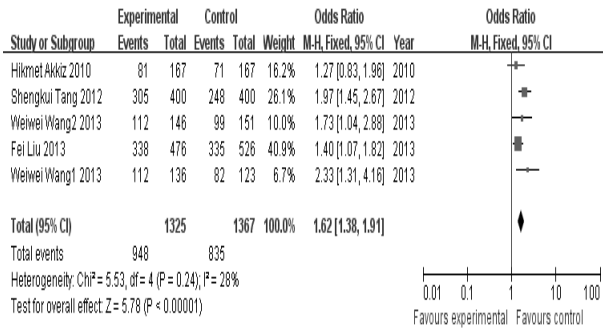

Figure 4. Forest Plots for Dominant Model (TT + TC *vs* CC) of the Association between NQO1 C609T Polymorphism and HCC

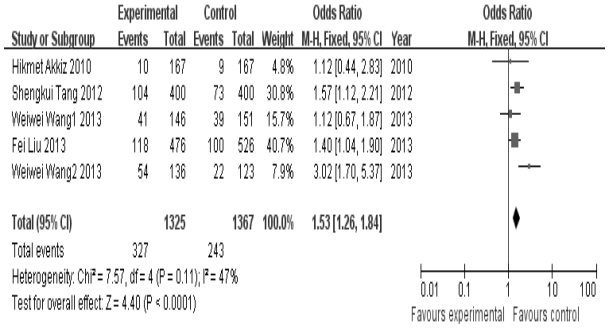

Figure 5. Forest Plots for Recessive Model (TT *vs* TC+CC) of the Association between NQO1 C609T Polymorphism and HCC

to assess publication bias of the literature on HCC. Figure 6 displays a funnel plot that detected the NQO1 C609T polymorphism and risk of overall HCC included in this meta-analysis in all genetic models. The shapes of the funnel plots did not show any evidence of obvious asymmetry. Also there was no statistical evidence of publication bias across studies by using Egger's linear

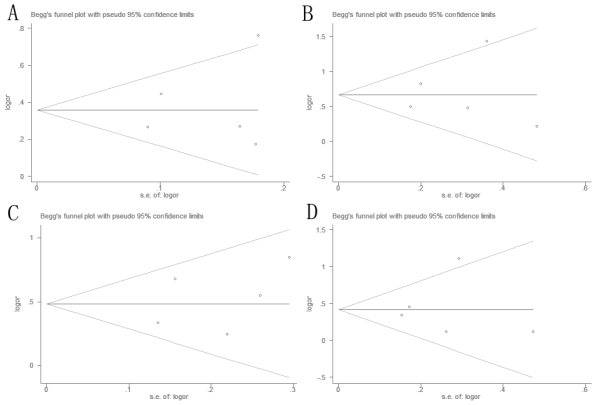

Figure 6. .Funnel Plots of All Individual Studies in the Meta-analysis of Risk of HCC with T *vs* C (A), TT *vs* CC (B), Dominant Model (C), and Recessive Model (D). No evidence of publication bias was found in any of them

regression test (allelic model: p=0.71; additive model: p=0.81; dominant model: p=0.56; and recessive model: p=0.87).

Discussion

The NQO1, which is generally involved in xenobiotic metabolizing, had been extensively studied on its relationship with different types of cancer, such as gastric, colorectal, cervical, prostate, and breast cancer(Hu et al., 2010; Malik et al., 2011; Yuan et al., 2011; Mandal et al., 2012; Peng et al., 2013). There were a number of studies performed to detect the association between NQO1 C609T polymorphism and HCC, but no consistent results were reported, this was largely attributed to the small samples or the relatively low statistical power of published studies. Meta-analysis is a powerful method for resolving inconsistent results with relatively large number of subjects. Therefore, this meta-analysis was conducted to provide a quantitative method for combining the different results. To the best of our knowledge, this was the most comprehensive meta-analysis assessing the genetic susceptibility of the NQO1 C609T gene polymorphism to HCC.

In the present study, we performed a meta-analysis with the 5 studies including 1325 cases and 1367 controls to explore overall effects of NQO1 C609T on HCC risk. Overall, the results of this meta-analysis from the current data suggested that there was a statistically significant association between NQO1 C609T polymorphism and an increased risk of HCC. This might be a reflection of a prominent role for NQO1 C609T variation in the etiology of HCC.

In the subgroup analysis by ethnicity, there was an obvious association between NQO1 C609T polymorphism and HCC in Asians (Table 2). In the meta-analysis, there were only 4 included studies assessing the association between NQO1 C609T polymorphism and HCC which were from Asians, and they all from Chinese. Therefore, more studies with large samples and other regions are needed to produce a more precise estimation on the effect of NQO1 C609T polymorphism on HCC.

Some limitation must be considered with interpreting the results from the meta-analysis. Firstly, although the Begg's test and Egger's test did not suggest any publication bias, selection bias could have occurred, because only studies published in English and Chinese were included in our meta-analysis. Secondly, the single-locus-based meta-analysis precluded the possibility of gene-gene and gene-environment interactions, as well as haplotype-based effects. Thirdly, our results were based on unadjusted estimates, and therefore, they were unable to adjust them by possible confounders such as age, gender, smoking status and environment factors. Considering these potential limitations, our results should be interpreted with caution.

In summary, our pooled data show evidence for major role of NQO1 C609T polymorphism in the carcinogenesis of hepatocellular carcinoma, especially among Chinese populations.

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