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RESEARCH ARTICLE

NQO1 C609T polymorphism is associated with esophageal cancer risk among Chinese: a meta-analysis

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Abstract NAD(P)H: quinone oxidoreductase 1 (NQO1) C609T gene polymorphism has been reported to influence the risk for esophageal cancer (EC) in many studies; however, the results remain controversial and ambiguous. We therefore carried out a meta-analysis of published case–control studies to investigate the association between NQO1 C609T polymorphism and EC susceptibility. Electronic searches were conducted on links between this variant and EC in several databases. Odds ratios (ORs) and 95 % confidence intervals (CIs) for homozygous, dominant model, recessive model and allele were calculated to estimate the strength of associations in fixed and random effect models. Heterogeneity and publication bias were also assessed. A total of 11 case–control studies were identified, including1,619 cases and 2,101 controls. C allele was associated with a decreased susceptibility risk of EC compared with the T allele among Chinese (OR= 0.70; 95 % CI=0.59–0.84). The contrast of homozygotes and the recessive and dominant models produced the same pattern of results as the allele contrast. Our pooled data suggest a significant association exists between NQO1 C609T polymorphism and EC among Chinese.

Keywords NQO1 polymorphism · Esophageal cancer · Meta-analysis

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Introduction

Esophageal cancer (EC) is the sixth leading cause of cancer death in the world, and 5-year survival rate is less than 10 % [1, 2]. There is considerable heterogeneity in incidence of EC, with highest rate recorded from northern Iran through the central Asian republics to north-central China, referred to as the "esophageal cancer belt" [3]. Although significant improvements in diagnosis and treatment of EC have been made over the past several decades, the etiology of most cases of EC remains unknown due to probable multifactorial mechanisms of pathogenesis. Susceptibility to EC appears to be determined by multiple genetic and environmental risk factors, such as tobacco/alcohol consumptions and exposure to dietary carcinogens [1]. There are convincing data that genetic susceptibility plays an important role [4, 5].

Quinone oxidoreductases (NAD(P)H: quinone oxidoreductase 1 (NQO1) is phase II cytosolic protein that catalyzes two-electron metabolic reduction of quinones or its derivatives into less toxic hydroquinone and prevent the generation of semiquinone free radicals or reactive oxygen species, thus may provide necessary protection for cells against free radical damage, oxidative stress, and neoplasia [6]. The NQO1 gene is located on chromosome 16q22.1. A nonsynonymous SNP has been described at nucleotide position 609 (rs1800566) [7]. The variant is a C-to-T transition and results in a proline-to-serine amino acid substitution at codon 187 in the protein. Compared with the wild-type (CC), the homozygous variant (TT) has only 2–4 % of the quinone reductase activity, whereas the heterozygote variant (CT) has a threefold decrease in enzyme activity [8]. Therefore, it has been hypothesized to affect cancer susceptibility by modifying the internal exposure to bioactivated carcinogens. There are several studies reporting association of NQO1 C609T polymorphism with EC [9, 10]; however, the results remain controversial and ambiguous.

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Meta-analysis is an accepted method to evaluate the association between gene polymorphism and disease susceptibility. A recent meta-analysis revealed that the NQO1 C609T polymorphism considerably increases the risk of esophageal cancer [11]. Another meta-analysis of NQO1 polymorphism published in 2012 indicated null association of the polymorphism with EC overall or with cancer cases stratified by tumor histopathology/ethnicity [10]. Thus, the exact relationship between NQO1 C609T polymorphism and EC susceptibility is not well established. Therefore, we performed a meta-analysis of all eligible studies to obtain a more precise estimation of the association between NQO1 C609T polymorphism and EC susceptibility.

Methods

Publication search and data extraction

The electronic databases PubMed, Embase, Web of Science, and CNKI were searched for studies to include in the present meta-analysis using the following terms: ("esophageal cancer" or "esophagus" or "ESCC" or "EAC" or "oesophagus") and ("polymorphism" or "SNP" or "allele" or "variant") and ("NQO1" or "NAD(P)H: quinine oxidoreductase 1" or "NAD(P)H dehydrogenase, quinone 1" or "DHQU"). An upper date limit of August 31, 2013, was used, but no early date limit was applied. The search was conducted without any restrictions on language but focused on studies that had been conducted on human subjects. The reference lists were screened of all of the identified studies and of the comprehensive reviews in the field. The number of cases and controls for the C609T genotype was extracted from each study. We did not define a minimum number of patients as a criterion for a study's inclusion in our meta-analysis. The studies included must meet the following criteria: (1) evaluation of the NQO1 C609T polymorphism and esophageal cancer risk; (2) case–control study (esophageal cancer group vs. control group).

Statistical analysis

The meta-analysis examined the overall association for the allele contrasts, the contrast of homozygotes, and the recessive and dominant models. The effect of the association was indicated as an odds ratio (OR) with its corresponding 95 % confidence interval (CI). The random effects model adjusts for the variability of results among trials and provides a more conservative estimate of an effect using a wider CI [12]. However, a random effects analysis will give more weight to smaller trials, which, as it appears, overestimate the benefit of treatment, leading to biased overall results [13]. Therefore, the pooled OR estimate of each study was calculated by both the fixed effects model (the Mantel–Haenszel method) [14] and the random effects model (the DerSimonian and Laird method) [15]. The heterogeneity between studies was tested using the Q statistic [16]. Heterogeneity was considered statistically significant if $P<0.10$. Begg's funnel plot and Egger's test were performed to assess the publication bias in the literature. Hardy–Weinberg equilibrium (HWE) was checked in the control group of the eligible studies by the chi-square test. All calculations were performed using ReviewManager 5.0.

Table 1 Characteristics of studies included in the meta-analysis

First author	Year	Country	Ethnicity	Histology	Sample size		Genotype frequency of cases/controls			OR (95%CI) allele contrast	HWE
					cases	controls	CC	CT	TT		
di Martino [17]	2007	UK	Caucasian	100%EAC	141	93	96/55	43/33	2/5	1.50 (0.95–2.39)	0.99
Feng [18]	2008	China	Asian	NR	201	201	28/59	131/109	42/33	0.67 (0.51–0.89)	0.14
Hamajima [19]	2002	Japan	Asian	NR	102	241	37/86	52/107	13/48	1.18 (0.84–1.64)	0.17
Malik [20]	2012	Indian	Asian	76.3%ESCC, 23.7%EAC	135	195	68/112	43/68	24/15	0.66 (0.47–0.93)	0.31
Marjani [21]	2010	Iran	Asian	100%ESCC	93	50	51/22	35/24	7/4	1.32 (0.77–2.24)	0.47
Sarbia [9]	2003	Germany	Caucasian	100%EAC	61	252	30/185	29/63	2/4	0.44 (0.28–0.71)	0.60
Umar [10]	2012	Indian	Caucasian	94.5%ESCC, 5.5%EAC	200	200	92/93	93/86	15/21	1.06 (0.79–1.43)	0.87
von Rahden [22]	2005	Germany	Caucasian	100%EAC	140	260	91/185	42/65	7/10	0.78 (0.54–1.14)	0.17
Zhang [23]	2003	Germany	Caucasian	100%ESCC	257	252	183/185	56/63	18/4	0.75 (0.54–1.05)	0.56
Zhang [24]	2003	China	Asian	100%ESCC	193	165	51/52	92/86	50/27	0.74 (0.55–1.00)	0.39
Zhang [25]	2006	China	Asian	NR	96	192	26/63	42/96	28/33	0.70 (0.49–0.99)	0.73

ESCC esophageal squamous cell cancer; EAC esophageal adenocarcinoma

Results

Twelve relevant studies describing the association between NQO1 C609T and esophageal cancer were identified [9, 10, 17–26]. However, in the study of Zhou et al. on the C609T polymorphism [26], the distributions of genotypes in the control groups were not in HWE ($p<0.05$), indicating genotyping errors and/or population stratification. This study was excluded from this meta-analysis. Finally, 11 studies met the inclusion criteria and were included (Table 1) [9, 10, 17–25]. Among these, five were on Caucasians and six were on Asians. All the included studies were case-controlled, comprising 1,619 cases and 2,101 controls. For case groups, the frequency of CC-homozygous individuals was 46.5 %. However, 40.6 % of CT-heterozygous individuals and 12.9 % of TT-homozygous individuals displayed the C609T polymorphism. In control groups, the frequencies of CC-homozygous individuals, CT-heterozygous individuals, and TT-homozygous individuals were 52.2, 38.1, and 9.7 %, respectively. The C allele frequencies in the case and control groups were 66.8 and 71.3 %, respectively.

All comparisons were listed in Table 2. Overall, the C allele was marginally associated with a decreased risk of EC compared with the T allele (OR=0.83; 95 % CI=0.69–1.00). However, the contrast of homozygotes and the recessive and dominant models produced the null results. Large heterogeneity (I^2=65 %, P=0.01) was detected among the 11 studies. To eliminate heterogeneity, we divided the 11 studies into different tumor histopathological and ethnic subgroups. Subsequently, heterogeneity disappeared in subgroups of Chinese subjects, which revealed that most of the studies could not be grouped based on ethnicity. In the analysis stratified by Chinese ethnicity, C allele was associated with a decreased susceptibility risk of EC compared with the T allele (OR=0.70;

Table 2 Odds ratios (ORs) and heterogeneity results for the genetic contrasts of NQO1 C609T polymorphism for esophageal cancer

	Population	OR				I^2 (%)	P value Q test
		Fixed effects (95 % CI)	P	Random effects (95 % CI)	P		
Alleles	All	0.82 (0.74–0.91)	0.0002	0.83 (0.69–1.00)	0.04	65	0.001
	Caucasian	0.87 (0.74–1.03)	0.10	0.84 (0.60–1.18)	0.33	75	0.003
	Asian	0.79 (0.69–0.91)	0.0007	0.81 (0.65–1.01)	0.06	58	0.04
	East Asian	0.79 (0.67–0.92)	0.002	0.79 (0.62–1.01)	0.07	59	0.06
	Chinese	0.70 (0.59–0.84)	<0.0001	0.70 (0.59–0.84)	<0.0001	0	0.88
	EAC	0.76 (0.61–0.95)	0.02	0.80 (0.44–1.45)	0.46	84	<0.0001
	ESCC	0.86 (0.74–1.01)	0.06	0.87 (0.73–1.04)	0.12	22	0.27
CC to TT	All	0.63 (0.50–0.81)	0.0002	0.65 (0.42–1.02)	0.06	64	0.002
	Caucasian	0.79 (0.50–1.23)	0.29	0.72 (0.27–1.92)	0.51	71	0.007
	Asian	0.58 (0.44–0.77)	0.0002	0.60 (0.38–0.97)	0.04	60	0.03
	East Asian	0.60 (0.44–0.83)	0.002	0.61 (0.34–1.11)	0.11	69	0.02
	Chinese	0.46 (0.32–0.66)	<0.0001	0.46 (0.32–0.66)	<0.0001	0	0.72
	EAC	0.66 (0.37–1.18)	0.16	0.66 (0.17–2.54)	0.55	74	0.004
	ESCC	0.61 (0.43–0.87)	0.006	0.62 (0.35–1.10)	0.10	55	0.06
CC to (CT+TT)	All	0.80 (0.70–0.93)	0.003	0.80 (0.63–1.02)	0.07	63	0.003
	Caucasian	0.85 (0.69–1.04)	0.11	0.82 (0.56–1.20)	0.31	71	0.008
	Asian	0.76 (0.62–0.93)	0.009	0.78 (0.56–1.09)	0.14	60	0.03
	East Asian	0.70 (0.55–0.89)	0.004	0.70 (0.47–1.05)	0.08	62	0.05
	Chinese	0.61 (0.46–0.82)	0.0008	0.61 (0.39–0.96)	0.03	58	0.09
	EAC	0.72 (0.55–0.94)	0.02	0.74 (0.37–1.47)	0.03	82	0.0002
	ESCC	0.94 (0.77–1.15)	0.54	0.94 (0.76–1.15)	0.54	0	0.60
CC+CT To TT	All	0.72 (0.58–0.90)	0.003	0.74 (0.50–1.09)	0.12	60	0.005
	Caucasian	0.84 (0.54–1.30)	0.43	0.82 (0.34–2.00)	0.67	67	0.02
	Asian	0.69 (0.54–0.88)	0.003	0.69 (0.46–1.05)	0.08	59	0.03
	East Asian	0.74 (0.56–0.97)	0.03	0.75 (0.46–1.22)	0.25	66	0.03
	Chinese	0.60 (0.44–0.82)	0.001	0.60 (0.44–0.82)	0.001	0	0.57
	EAC	0.73 (0.41–1.28)	0.27	0.79 (0.29–2.16)	0.64	59	0.04
	ESCC	0.62 (0.44–0.85)	0.003	0.62 (0.35–1.09)	0.10	60	0.04

95 % CI=0.59–0.84). No heterogeneity (I^2=0 %, P=0.88) was detected among the three studies. The contrast of homozygotes and the recessive and dominant models produced the same pattern of results as the allele contrast. Our data indicated null association of the polymorphism with EC with cancer cases stratified by tumor histopathology.

Begg's funnel plot and Egger's test were performed to assess the publication bias in the literature. The evaluation of publication bias for the C allele versus the T allele showed that the Egger test was not significant (p =0.68), which did not indicate a potential for publication bias.

Discussion

The genetic susceptibility to cancer has been the focus of research in the scientific community. The homozygous TT and the heterozygote CT genotype had a decrease in enzyme activity, and the lack of NQO1 activity might result in reduced detoxification exogenous carcinogens, which might be linked to the risk of esophageal cancer. This meta-analysis summarized all of the available data on the association between the NQO1 C609T polymorphism and EC, including a total of 1, 619 cases and 2,101 controls. In this study, we noticed that the NQO1 C609T polymorphism could play divergent roles across different ethnic populations. In Chinese population, a significant association was found for the genetic models examined. In addition, heterogeneity disappeared when the Chinese population was viewed as separate groups, which suggested that the effect of the NQO1 C609T polymorphism on the risk of EC might differ based on ethnicity. A wide variation of the allele frequency has been observed across ethnic groups. The NQO1 C609T variant genotypes in white populations were different from Asian populations [27]. There were striking differences in terms of mutant 609 T allele frequency in controls between Asians and Caucasians, suggesting that different genetic backgrounds may account for this discrepancy or that different populations may have different linkage disequilibrium patterns.

A previous meta-analysis addressing the association between NQO1 C609T polymorphism and EC had several errors [11]. First, some studies were not included in that analysis. Second, the study of Zhou et al. was included in that analysis, whose distributions of genotypes in the control groups were not in HWE. Third, ethnicity of the study of Marjani et al. is Asian but not Caucasian. These errors resulted in the wrong conclusion.

Several limitations should be considered when interpreting the results of our analysis. First, our results were based on unadjusted estimates, whereas a more precise analysis could have been conducted if the individual data were available, which would allow researchers to adjust for covariates, including age, family history, lifestyle, and environmental factors. Second, publication bias is another major concern in all meta-analyses because studies reporting positive or significant findings are more likely to be published than those reporting nonsignificant results. Third, the heterogeneity among the trials could be another limitation of our meta-analysis, although we applied both a random effects model and a fixed effects model to combine the data. The absence of a statistically significant heterogeneity in Chinese population might reveal that the studies should be separated based on ethnicity. However, as the number of trials was limited, careful interpretation of the heterogeneity is necessary. Lastly, only three Chinese studies might be insufficient for getting conclusive result or for evaluating heterogeneity or publication bias.

In conclusion, our pooled data suggest a significant association exists between NQO1 C609T polymorphism and EC among Chinese.

Conflicts of interest None

References

1. Enzinger PC, Mayer RJ. Esophageal cancer. N Engl J Med. 2003;349:2241–52.
2. Ferlay J, Shin HR, Bray F, Forman D, Mathers C, Parkin DM. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. Int J Cancer. 2010;127:2893–917.
3. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin. 2011;61(2):69–90.
4. Zhong S, Zhao W, Lu C, Li B, Yuan Y, Guo D, et al. Glutathione S-transferase M1 null genotype contributes to increased risk of esophageal carcinoma in Chinese population. Tumour Biol. 2013;34(4): 2403–7.
5. Zhang J, Zhou J, Zhang P, Wang W, Tao S, Wang M. A meta-analysis of the association between the hOGG1 Ser326Cys polymorphism and the risk of esophageal squamous cell carcinoma. PLoS One. 2013;8(6):e65742.
6. Jaiswal AK. Regulation of genes encoding NAD(P)H:quinone oxidoreductases. Free Radic Biol Med. 2000;29(3–4):254–62.
7. Traver RD, Siegel D, Beall HD, Phillips RM, Gibson NW, Franklin WA, et al. Characterization of a polymorphism in NAD(P)H: quinone oxidoreductase (DT-diaphorase). Br J Cancer. 1997;75:69–75.
8. Kuehl BL, Paterson JW, Peacock JW, Paterson MC, Rauth AM. Presence of a heterozygous substitution and its relationship to DT-diaphorase activity. Br J Cancer. 1995;72:555–61.
9. Sarbia M, Bitzer M, Siegel D, Ross D, Schulz WA, Zotz RB, et al. Association between NAD(P)H: quinone oxidoreductase 1 (NQ01) inactivating C609T polymorphism and adenocarcinoma of the upper gastrointestinal tract. Int J Cancer. 2003;107(3):381–6.
10. Umar M, Upadhyay R, Kumar S, Ghoshal UC, Mittal B. Null association of NQO1 609C>T and NQO2–3423G>A polymorphisms with susceptibility and prognosis of Esophageal cancer in north Indian population and meta-analysis. Cancer Epidemiol. 2012;36(6):e373–9.
11. Yanling H, Yuhong Z, Wenwu H, Lei X, Mingwu C. NQO1 C609T polymorphism and esophageal cancer risk: a HuGE review and meta-analysis. BMC Medical Genetics. 2013;14:31.

12. Berlin JA, Laird NM, Sacks HS, Chalmers TC. A comparison of statistical methods for combining event rates from clinical trials. Stat Med. 1989;8(2):141–51.
13. Poole C, Greenland S. Random-effects meta-analyses are not always conservative. Am J Epidemiol. 1999;150:469–75.
14. Mantel N, Haenszel W. Statistical aspects of the analysis of data from retrospective studies of disease. J Natl Cancer Inst. 1959;22:719–48.
15. DerSimonian R, Laird N. Meta-analysis in clinical trials. Control Clin Trials. 1986;7(3):177–88.
16. Cochran WG. The combination of estimates from different experiments. Biometrics. 1954;10:101–29.
17. di Martino E, Hardie LJ, Wild CP, Gong YY, Olliver JR, Gough MD, et al. The NAD(P)H:quinone oxidoreductase I C609T polymorphism modifies the risk of Barrett esophagus and esophageal adenocarcinoma. Genet Med. 2007;9(6):341–7.
18. Feng XX, Li ZF, Wang LB, Zhang JB, Lu ZX. Study on the relationship between polymorphisms of NQO1 gene and susceptibility to esophageal cancer. Chin J Dis Control Prev. 2008;12(2):112–4.
19. Hamajima N, Matsuo K, Iwata H, Shinoda M, Yamamura Y, Kato T, et al. NAD(P)H: quinone oxidoreductase 1 (NQO1) C609T polymorphism and the risk of eight cancers for Japanese. Int J Clin Oncol. 2002;7(2):103–8.
20. Malik MA, Zargar SA, Mittal B. Role of NQO1 609C>T and NQO2–3423G>A gene polymorphisms in esophageal cancer risk in Kashmir valley and meta analysis. Mol Biol Rep. 2012;39(9):9095–104.
21. Marjani HA, Biramijamal F, Rakhshani N, Hossein-Nezhad A, Malekzadeh R. Investigation of NQO1 genetic polymorphism, NQO1 gene expression and PAH-DNA adducts in ESCC. A case–control study from Iran. Genet Mol Res. 2010;9(1):239–49.
22. von Rahden BH, Stein HJ, Langer R, von Weyhern CW, Schenk E, Döring C, et al. C609T polymorphism of the NAD(P)H:quinone oxidoreductase I gene does not significantly affect susceptibility for esophageal adenocarcinoma. Int J Cancer. 2005;113(3):506–8.
23. Zhang J, Schulz WA, Li Y, Wang R, Zotz R, Wen D, et al. Association of NAD(P)H: quinone oxidoreductase 1 (NQO1) C609T polymorphism with esophageal squamous cell carcinoma in a German Caucasian and a northern Chinese population. Carcinogenesis. 2003;24(5):905–9.
24. Zhang JH, Li Y, Wang R, Geddert H, Guo W, Wen DG, et al. NQO1 C609T polymorphism associated with esophageal cancer and gastric cardiac carcinoma in North China. World J Gastroenterol. 2003;9(7):1390–3.
25. Zhang WC, Yin LH, Pu YP, Liang GY, Hu X, Liu YZ, et al. Relationship between quinone oxidoreductase 1 gene ns-SNP and genetic susceptibility of esophageal cancer. Chin J Prev Med. 2006;40:324–7.
26. Zhou YL, Chen HF, Shi XS, Zhou ZJ, Li GL, Pan PC, et al. A case–control study on the polymorphisms of NQO1 and susceptibility of esophageal cancer. Chin Cancer. 2006;15:659–63.
27. Kelsey KT, Ross D, Traver RD, Christiani DC, Zuo ZF, Spitz MR, et al. Ethnic variation in the prevalence of a common NAD(P)H quinone oxidoreductase polymorphism and its implications for anticancer chemotherapy. Br J Cancer. 1997;76:852–4.