RESEARCH ARTICLE

Association Between the XRCC3 Thr241Met Polymorphism and Cervical Cancer Risk: a Meta-analysis

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Abstract

Background: Numerous epidemiological studies have been conducted to evaluate the association between variants of the DNA repair gene XRCC3 and cancer risk. Here we focused on one XRCC3 polymorphism and development of cervical cancer, performing a meta-analysis. Methods: The pooled association between the XRCC3 Thr241Met polymorphism and cervical cancer risk was assessed by odds ratios (ORs) and their 95% confidence intervals (95%CIs). Results: A total of 5 case-control studies met the inclusion criteria. The pooled ORs for the total included studies showed no association among homozygotes TT vs. CC: OR=1.93, 95%CI=0.68-5.49, *P*=0.22; dominant model TT+TC vs. CC: OR=1.37, 95%CI=0.90-2.06, *P*=0.14; and recessive model TT vs. TC+CC: OR=1.76, 95%CI=0.68-4.55, *P*=0.25, but might be a slight risk factor for cervical cancer in heterozygote contrast TT vs. CT: OR= 1.33, 95%CI=1.04-1.71, *P*=0.02. In subgroup analysis, significant associations were found for Asians under all genetic models. Conclusions: Our meta-analysis suggested the XRCC3 Thr241Met polymorphism might not act as a cervical cancer risk factor overall. However, in subgroup analysis, a significant association was found in Asians under all genetic models. The association should be studied with a larger, stratified population, especially for Asians.

Keywords: Cervical cancer - XRCC3 - polymorphism - meta-analysis

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Introduction

The cancer type has been selected in our study because of its high incidence and mortality in women, ranking on the fifth place by incidence and on the seventh place by mortality [http://eco.iarc.fr/EUCAN/Country.aspx?ISOCountryCd=968]. Cervical cancer kills more than 288,000 women each year worldwide and disproportionately affects the poorest, most vulnerable women. At least 80 percent of cervical cancer deaths occur in developing countries, with most occurring in the poorest regions - South Asia, sub-Saharan Africa, and parts of Latin America [http://screening.iarc.fr/cervicalindex.php]. Although prophylactic vaccines against HPV have been developed, they are effective only to prevent new infections of certain types of HPV (HPV16 and HPV18), and the current hrHPV carriers or new carriers with hrHPV other than type 16 and 18 remain facing the risk. Since only a small part of hrHPV carriers can develop cervical cancer, it is indicated that the presence of other factors other than hrHPV infection is responsible for the development of cervical cancer. To prevent and reduce cervical cancer cases, it is thus important to unveil cryptic risks for cervical cancer (Settheetham-Ishida et al., 2011). The X-ray repair cross-complementing group 3 (XRCC3) belongs to a family of the DNA repair genes involved in repairing DNA double strand breaks (DSB). It is located on chromosome 14q32.33. The coding DNA repair protein of XRCC3 contains 346 amino acids (Lee et al., 2007). Current analyses show that replication of integrated HPV may induce local rearrangements within the integration locus, activating DNA damage checkpoints and recruiting proteins of homologous recombination (HR) (Kadaja et al., 2009).The findings made us to speculate that polymorphisms in HR genes may be directly involved in cervical cancer risk. The XRCC3 gene belongs to HR genes in repairing DNA double strand breaks (DSB) caused by normal metabolic processes and/or exposure to ionizing radiation (Tebbs et al., 1995). DNA-repair systems are essential for the maintenance of integrity of the genetic material and dysfunction of the systems thus plays critical roles in cancer development (Mathonnet et al., 2003).

The Thr241Met (XRCC3-18067C>T, rs861539) substitution, a C to T transition at codon241 in exon7, is the most common investigated polymorphism in XRCC3 (Matullo et al., 2001). The XRCC3 Thr241Met polymorphism includes three genotypes wild-type genotype (CC), the heterozygote (CT) and the homozygote (TT).Many studies have been performed to estimate the relationship of XRCC3 Thr241Met polymorphism with cervical cancer risk, but the findings of their publications are contradictory. To derive a more precise estimation of the association, we performed a meta-analysis.

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Materials and Methods

Search strategy

All case-control studies about the XRCC3 Thr241Met polymorphism and cervical cancer risk published up to August, 2013. Systematic searches were identified by the PubMed, Embase and CNKI (China National Knowledge Infrastructure), using the terms "XRCC3" or "X-ray repair cross-complementing group 3" in combination with "polymorphism" or "polymorphisms" or "variant" or "mutation" in combination with "cervical cancer". Concurrently, the reference lists of reviews and retrieved articles were searched manually. No language or country restrictions were applied. Review articles were also examined to find additional eligible studies. The literature retrieval was performed in duplication by two independent reviewers (Lingyan Qin and Xu Chen).

Inclusion and Exclusion Criteria

Studies included in the meta-analysis must meet the following criteria: they (a) evaluated the association between XRCC3 Thr241Met polymorphism and cervical cancer risk; (b) supplied the number of individual genotypes for the XRCC3 Thr241Met gene polymorphisms in cervical cancer cases and controls, respectively; and (c) were case-control studies. The exclusion criteria were as follows: they were (a) not case-control studies; (b) studies that were based on incomplete raw data and those with no usable data reported; (c) conference abstracts, case reports, reviews, letters and editorial articles; and (d) studies that contained overlapping data.

Data extraction

From each eligible study, the following information were extracted by two investigators independently with the standard protocol: first author's surname, year of publication, country of origin, ethnicity, source of control, method of genotyping, numbers of cases and controls, Hardy–Weinberg equilibrium (HWE) of controls, and the frequency of genotypes in both cases and controls. Ethnic backgrounds were categorized as Caucasians, Asians and African. If a study did not show the ethnic descendent or it was not possible to separate participants according to such phenotype, the group reported was termed as "mixed ethnicity". We did not contact the author of the primary study to request the information.

Statistical analysis

The strength of the association between XRCC3 polymorphisms and cervical cancer was measured by odds ratio (OR) with 95% confidence intervals (CI). The significance of the pooled OR was determined by Z test and a p value of less than 0.05 was considered significant. We calculated the pooled OR under homozygote (TT vs. CC), heterozygote (TC vs. CC), dominant (TT/TC vs. CC) and recessive (TT vs. TC/CC) genetic models for each polymorphism, respectively. Subgroup analysis was done by ethnicity.

When we assessed the between-study heterogeneity, the chi-square (based Q statistic test) was to test for heterogeneity and the I^2 statistic was to quantify the proportion of the total variation due to heterogeneity. If the results of the Q test was $P_Q \geq 0.1$ and $I^2 < 50\%$, the fixed-effects was performed to pool the results (the Mantel-Haenszel method) (Mantel and Haenseal,1959). Otherwise, random-effects model model was considered when the result of the Q test was $P_Q < 0.1$ or I^2 50% (DerSimonian and Laird method) (DerSimonian and Laird, 1986). If heterogeneity was observed, meta-regression analysis was applied to both general analyses and subgroup analyses to find the source of heterogeneity.

In order to confirm the stability of our pooled results in the meta-analysis, a sensitivity analysis was performed by sequential omission of a single study (Attia et al., 2003). The distribution of the genotypes in the control population was tested for HWE by a goodness-of-fit Chi-square test. The publication bias was tested by funnel plot, Begg's funnel plot and Egger's linear regression test ($P < 0.05$ was considered a statistically significant publication bias) (Egger et al., 1997; Stuck et al., 1998). All calculations were performed using STATA, version12.0 (Stata Corporation, College Station, TX), and all the *P* values were two-sided.

Results

Study characteristics

The literature search identified 30 related articles through PubMed, Embase, and CNKI. With the step of screening the title, abstract or content, 5 eligible studies were selected. Manual search of references cited in 1 additional article, but it was excluded for lack of data about XRCC3 Thr241Met gene polymorphism of cervical cancer (Wang et al., 2009). Finally, a total of 5 relevant studies including 4 English articles (He et al., 2008; Settheetham-Ishida et al., 2011; Djansugurova et al., 2013; Perez et al., 2013) and 1 Chinese paper (Xiao et al., 2010) met the inclusion criteria for the meta-analysis (including a total of 806 cervical cancer cases and 850 controls). There were 3 studies of Asians (He et al., 2008; Xiao et al., 2009; Settheetham-Ishida et al., 2011) and 2 studies of Caucasians (Djansugurova et al., 2013; Perez

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

First author	Year	Country	Ethnicity	Source of control	Method of Genotyping	Sample size case/control	HWE of Controls
He	2008	China	Asian	PB	ASPCR	200/200	0.39
Xiao	2010	China	Asian	PB	PCR-RFLP	158/164	0.1
Settheetham-Ishida	2011	Thailand	Asian	PB	PCR-RFLP	111/118	0.4
Pérez	2013	Argentina	Caucasian	PB	Sequencing	117/205	0.73
Djansugurova	2013	Kazakhstan	Caucasian	PB	ASPCR	217/160	0.28

HWE, Hardy-Weinberg equilibrium; HB, hospital based; PB, population based

Table 2. Meta-Analysis of XRCC3 Thr241Met Polymorphism with the Cervical Cancer Risk.

Comparison	Population	N	Test of association			Mode	Test of heterogeneity		Pegger
			OR	95% CI	*P*		*P*	I^2	
TT vs. CC	Overall	5	1.93	0.68-5.49	0.22	random	0.01	70.8	0.46
	Asian	3	2.88	1.32-6.30	0.01	fixed	0.74	0	-
	Caucasian	2	1.48	0.19-11.9	0.71	random	0	89.6	-
TC vs.CC	Overall	5	1.33	1.04-1.71	0.02	random	0.16	38.7	0.48
	Asian	3	1.49	1.04-2.13	0.03	fixed	0.18	42.3	-
	Caucasian	2	1.2	0.71-2.04	0.5	random	0.13	57	-
TT/TC vs.CC	Overall	5	1.37	0.90-2.06	0.14	random	0.02	65	0.57
	Asian	3	1.62	1.16-2.28	0.01	fixed	0.14	49.8	-
	Caucasian	2	1.25	0.55-2.83	0.6	random	0.01	84	-
TTvs.TC/CC	Overall	5	1.76	0.68-4.55	0.25	random	0.02	66.3	0.37
	Asian	3	2.4	1.11-5.18	0.03	fixed	0.76	0	-
	Caucasian	2	1.2	0.71-2.04	0.5	random	0.13	57	-

N, number of studies; PQ value used to test the heterogeneity; CI, confidence interval; OR, odds ratio

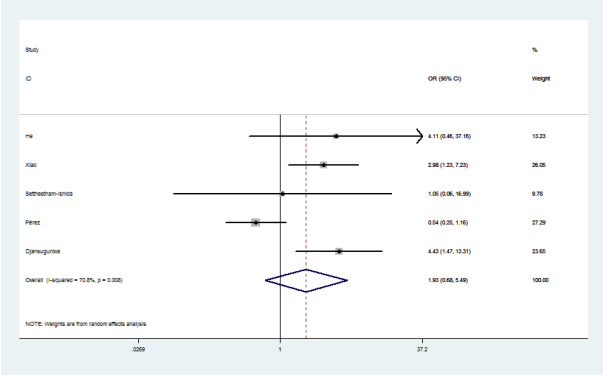

Figure 1. Forest Plot Showed That XRCC3 Thr241Met Polymorphism Was Not Associated with Cervical Cancer Susceptibility under Homozygote Contrast (TT vs. CC)

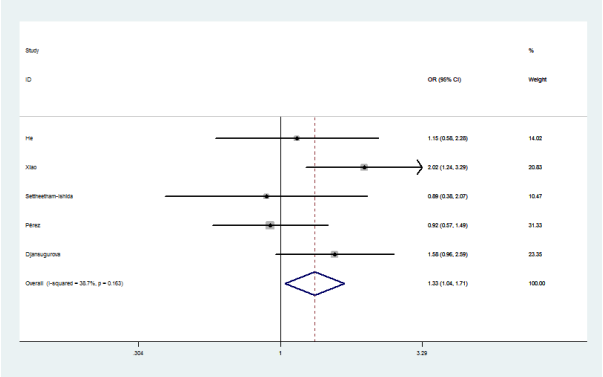

Figure 2. Forest Plot Showed That XRCC3 Thr241Met Polymorphism Might Be a Slight Risk Factor for Cervical Cancer in Heterozygote Contrast (CT vs. CC)

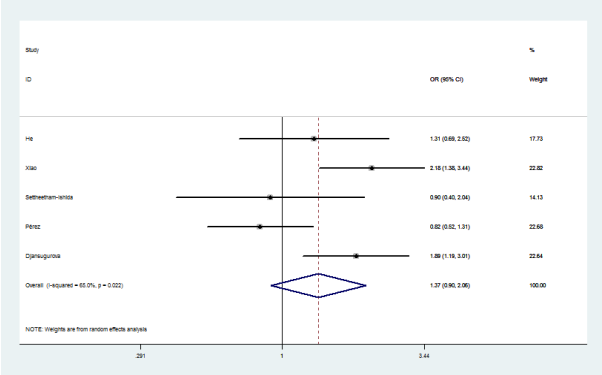

Figure 3. Forest Plot Showed That XRCC3 Thr241Met Polymorphism Was Not Associated with Cervical Cancer Susceptibility under Dominant Model (TT/CT vs. CC)

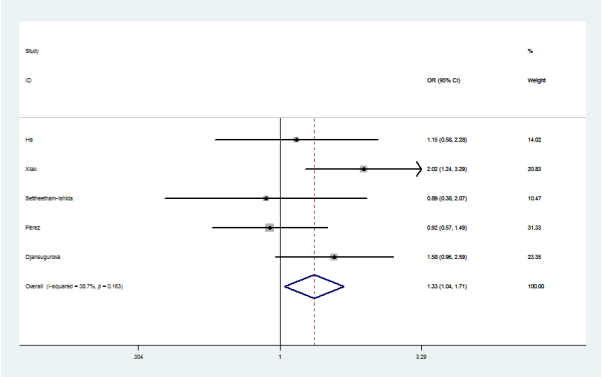

Figure 4. Forest Plot Showed That XRCC3 Thr241Met Polymorphism Was Not Associated with Cervical Cancer Susceptibility under Recessive Model (TT vs. CT/CC)

et al., 2013). The main characteristics of the studies were presented in Table 1.

Meta-analysis results

Table 2 lists the main results of this meta-analysis. Overall, significantly elevated cervical cancer risk was associated in heterozygote contrast (TC vs. CC: OR = 1.33, 95%CI = 1.04-1.71, *P* = 0.02, P_Q= 0.16) (Figure 2, Table 2), when all the eligible studies were pooled into the meta-analysis. However, there was no significant association in homozygote contrast (TT vs. CC: OR=1.93, 95%CI=0.68-5.49, *P*=0.22, P_Q=0.01), dominant model (TT/TC vs. CC: OR=1.37, 95%CI=0.90-2.06, *P*=0.14, P_Q=0.02), and recessive model (TT vs. TC/CC: OR=1.76, 95%CI=0.68-4.55, *P*=0.25, P_Q=0.02) (Figure 2, Figure 3, Figure 4, Table 2).

In the stratified analysis by ethnicity, significantly increased risks were found among Asians for all of models (homozygote contrast TT vs. CC: OR=2.88, 95%CI=1.32-6.30, *P*=0.01, P_Q=0.74; heterozygote contrast TC vs. CC: OR = 1.49, 95%CI = 1.04-2.13, *P* = 0.03, P_Q= 0.18; dominant model TT/TC vs. CC: OR= 1.62, 95%CI= 1.16-2.28, *P*= 0.01, P_Q= 0.14; recessive model TT vs. TC/CC:

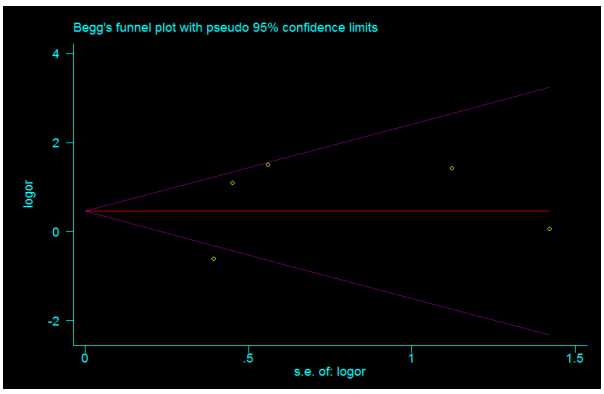

Figure 5. Funnel Plot Did Not Indicate Evidence of Obvious Asymmetry under Heterozygote Contrast (CT vs. CC)

OR= 2.40, 95%CI= 1.11-5.18, *P*= 0.03, P_Q= 0.76), while no significant association was found in both Caucasians for any genetic model (Table 2).

Heterogeneity analysis

In our current study, heterogeneity existed in homozygote contrast (P_Q=0.01), dominant model (P_Q=0.02), and recessive model (P_Q=0.02) (Table 2). To explore the sources of heterogeneity, we performed meta-regression and subgroup analysis. After assessing the source of heterogeneity for all genetic models by subgroup analysis on ethnicity, the heterogeneity was partly decreased.

Sensitivity analyses and publication bias

One study involved in the meta-analysis was deleted each time to reflect the influence of the individual study setting to the pooled OR. Our analyses indicated that the results of the overall population and subgroup were stable and reliable.

Begg's funnel plot and Egger's test were used to assess the publication bias of studies included in this meta-analysis. *P* values of Egger's tests were used to provide statistical evidence of funnel plots' symmetry. The shapes of Funnel plot did not reveal obvious evidence of asymmetry. All the *p* values of Egger's tests were more than 0.05 (homozygote contrast: Egger's Test for *P*= 0.46; heterozygote contrast: Egger's Test for *P*= 0.48; dominant model: Egger's test for *P*= 0.57; recessive model: Egger's test for *P*= 0.37) (Table 2, Figure 5) and further confirmed that publication bias did not exist among including studies.

Discussion

Approximately 30% of women with sexual experience are infected with high-risk HPV. However, only 1% of these women will develop CIN and cervical cancer (Giuliano et al., 2002). This indicates that HPV infection is not sufficient to develop CIN or cancer, and other cofactors, such as variations in DNA repair genes, should be considered. Therefore, it is reasonable to understand genetic variability in DNA repair genes and their influence on modifying an individual's susceptibility to cancer (Spitz et al., 2003).

Recently, genetic polymorphisms of the DNA repair genes in the etiology of several cancers have drawn increasing attention. X-ray cross-complementing group 3 (XRCC3) is one of protein components involved in homologous recombination repair (HRR) pathway responsible for DNA double-strand break (DSB) repair. Because of genetic variation, decreased capacity of DNA repair gene is associated with increased susceptibility to human tumors (Garcia-Closas et al., 2006). The association between the XRCC3 Thr241Met polymorphism and cervical cancer risk was not clear due to inconsistent data. Therefore we performed a meta-analysis to clarify the inconsistency and to establish its gene-disease association.

By pooling 5 studies with 803 cases and 847 controls, our meta-analysis showed a statistically significant association between the XRCC3 Thr241Met polymorphism and cervical cancer risk when applying a heterozygote contrast (TC vs. CC: OR = 1.33, 95%CI = 1.04-1.71, *P* = 0.02, P_Q= 0.163). Further subgroup analyses revealed that this association only existed in Asians (homozygote contrast TT vs. CC: OR=2.88, 95%CI=1.32-6.30, *P*=0.01, P_Q=0.74; heterozygote contrast TC vs. CC: OR = 1.49, 95%CI = 1.04-2.13, *P* = 0.03, P_Q= 0.18; dominant model TT/TC vs. CC: OR= 1.62, 95%CI= 1.16-2.28, *P*= 0.01, P_Q= 0.14; recessive model TT vs. TC/CC: OR= 2.40, 95%CI= 1.11-5.18, *P*= 0.03, P_Q= 0.76). Considerable heterogeneity was detected across studies, and the heterogeneity cannot be fully explained by ethnicity (heterogeneity still existed in Caucasians). The reason might be that the including studies might have sample selective bias, especially for the study from Kazakhstan. What is more, many factors could affect the genomic polymorphism spectrum in populations, such as ethnicity, habits, geographical location, type of diet etc. The ethno-genetic status, the radiation background, age, and bad habits strongly influence on mutagenic processes. According to the above phenomenon, it is necessary to further conduct large studies by using standardized unbiased methods, homogeneous cervical cancer patients and well matched controls. Moreover, gene-gene and gene-environment interactions should be considered in future analysis. Such studies taking these factors into account may ultimately conduct our better and more comprehensive understanding of the association between the XRCC3 Thr241Met polymorphism and cervical cancer.

The ORs showed there was a significant risk of disease related with XRCC3 Thr241Met polymorphism among Asians rather than Caucasians. In addition, our results were consistent with the conclusions of many previous studies, which showed no associations between the XRCC3 Thr241Met polymorphism and cancer risk in all of studies, but association was found in Asians, such as Lung cancer, colorectal cancer and glioma (Liang et al., 2013; Tian et al., 2013; Wang et al., 2013). These analyses confirmed the liability of the results.

In a word, XRCC3 is a popular gene in cancer study. However there are some points need to be further verified, such as whether it plays an important role in cancer or not, what its anticancer mechanism is, and whether it triggers cancer in the same way among diverse ethnicities.

There were still some limitations inherited from the

published studies. Firstly, the number of studies involved in the meta-analysis was relatively small, so the subgroup analysis was hard to perform. Secondly, the small number of studies and sample sizes limited the ability to draw a more completely reliable conclusion. Therefore, a larger number of available studies with a large sample size and adequate representation of ethnicities will help us to determine a more confident estimate of the effect of this polymorphism on cervical cancer. Thirdly, cervical cancer is a multi-factorial disease, and potential interactions between gene-gene and gene-environment should be considered. Though there were some limitations, our meta-analysis could still provided valuable information for studying the relationship between XRCC3 Thr241Met polymorphism and cervical cancer risk.

Our meta-analysis suggested the XRCC3 Thr241Met polymorphism might not act as a cervical cancer risk factor among all subjects. However, in subgroup analysis, the significant association was found in Asians under all genetic models. The association should be studied with a larger, stratified population, especially for Asians.

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