Genetics and Molecular Research

Lack of association between *ERCC5* gene polymorphisms and gastric cancer risk in a Chinese population

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Genet. Mol. Res. 15 (2): gmr.15027779
Received October 5, 2015
Accepted February 11, 2016
Published June 10, 2016
DOI http://dx.doi.org/10.4238/gmr.15027779

ABSTRACT. We conducted a case-control study to assess the association between single nucleotide polymorphisms in the *ERCC5* promoter (rs2094258 and rs751402) and development of gastric cancer in a Chinese population. This investigation included 184 patients with pathologically diagnosed gastric cancer and 206 healthy subjects recruited between October 2012 and December 2014. The genotyping of *ERCC5* rs2094258 and rs751402 variants was performed by polymerase chain reaction coupled with restriction fragment length polymorphism. Genotype distributions of these polymorphisms conformed to Hardy-Weinberg equilibrium in both patient ($P = 0.25$ for rs2094258 and $P = 0.61$ for rs751402) and control groups ($P = 0.48$ for rs2094258 and $P = 0.42$ for rs751402). Using unconditional logistic regression analysis, we found that neither of these *ERCC5* variants was associated with increased risk of gastric cancer under co-dominant, dominant, or recessive models ($P < 0.05$). In conclusion, we suggest that the rs2094258 and rs751402 polymorphisms are not connected to the development of this disease under codominant, dominant, and recessive models.

Key words: *ERCC5*; rs2094258; rs751402; Polymorphism; Gastric cancer

INTRODUCTION

Gastric cancer is one of the most common malignant tumors worldwide. Individuals infected with *Helicobacter pylori* are known to be at higher risk of developing gastric ulcers and cancer. In addition, previous epidemiological studies have indicated that consumption of alcohol, lack of activity, obesity, and high sodium intake may play critical roles in susceptibility to gastric cancer (van den Brandt and Goldbohm, 2006). However, not all individuals exposed to such risk factors develop this disease, suggesting that genetic factors may contribute to its development. Indeed, many previous studies have reported that certain genes are involved in gastric cancer risk, including*miR-146a*, *IL-17A*, *LMP2*, *LMP7*, *IGF1*, and *GSTM1* (Farahani et al., 2015; Hou and Yang, 2015; Ma et al., 2015; Wei et al., 2015; Zhang and Cui, 2015).

Excision repair cross-complementing rodent repair deficiency, complementation group 5 (ERCC5) is an important member of a family of enzymes that includes the DNaseIV/flap structure-specific endonuclease 1 group of structure-specific nucleases, and functions in nucleotide excision repair (Hohl et al., 2007). To date, a small number of studies have examined the association between *ERCC5* gene polymorphisms and gastric cancer risk, but their results have been inconclusive (Duan et al., 2012; Xue et al., 2015). Therefore, we conducted a case-control study to assess the association between *ERCC5* promoter single nucleotide polymorphisms (SNPs; rs2094258 and rs751402) and development of gastric cancer in a Chinese population.

MATERIAL AND METHODS

Patients

This case-control study included 184 patients with pathologically diagnosed gastric cancer having attended Gansu Provincial Hospital between October 2012 and December 2014. Patients with other malignant neoplasms, recurrent tumors, or serious liver or kidney diseases were excluded from this study. The control group consisted of 206 subjects randomly selected while attending the health center of Gansu Provincial Hospital for checkups during the same period. Control subjects with a history of malignant cancer.

Patients and controls were interviewed using a standardized questionnaire concerning sociodemographic traits, including age and lifestyle habits. Details of tobacco and alcohol consumption were ascertained from all participants with a self-designed questionnaire. Tobacco smoking status was defined using two groups, namely, smokers and non-smokers. Likewise, alcohol drinking status was classified using two groups, drinkers and non-drinkers.

All individuals voluntarily took part in the study and gave their informed consent prior to participation. This project was approved by the Ethics Committee of Gansu Provincial Hospital.

DNA extraction and SNP genotyping

Following the manufacturer protocol, a TIANamp Blood DNA Kit (Tiangen, Beijing, China) was used to extract DNA from peripheral blood samples collected from patients and controls. Genotyping of *ERCC5* rs2094258 and rs751402 was performed by polymerase chain reaction (PCR) coupled with restriction fragment length polymorphism. The PCR fragments

were subsequently digested with restriction enzymes specific to the sequences of interest. The PCR began with an initial denaturation at 95°C for 5 min, followed by 30 cycles of denaturation at 94°C for 60 s, annealing at 60°C for 60 s, and extension at 72°C for 60 s, before a final extension at 72°C for 10 min.

Statistical analysis

Differences between gastric cancer patients and control subjects in terms of sociodemographic characteristics were calculated using the chi-square test or Student *t*-test. Hardy-Weinberg equilibrium (HWE) of *ERCC5* rs2094258 and rs751402 genotype distributions was assessed using a chi-square test. Univariate logistic regression analysis was employed to analyze the relationship between sociodemographic characteristics and gastric cancer risk. Multiple logistic regression models were established to estimate the relative gastric cancer risk associated with the two SNPs under investigation. Odds ratios (ORs) and their 95%CIs were calculated, and the results were adjusted for confounding factors.

RESULTS

The sociodemographic characteristics of the gastric cancer patients are summarized in Table 1. The mean ages of patients and control subjects were 57.27 ± 12.63 and 46.24 ± 11.42 years, respectively. There were 68 (36.96%) women and 116 (63.04%) men in the gastric cancer group, and 94 (45.63%) women and 112 (54.37%) men in the control group. Compared to control subjects, gastric cancer patients were more likely to be older (OR = 2.23, 95%CI = 1.46-3.42; P < 0.001) and drink alcohol (OR = 1.50, 95%CI = 0.98-2.28; P = 0.04).

Table 1. Sociodemographic and clinical characteristics of patients with gastric cancer and control subjects.

Variable	Patients	%	Controls	%	Chi-square	OR (95%CI)	P value
Age, years							
<50	77	41.85	127	61.65		1.0 (Ref.)	-
≥50	107	58.15	79	38.35	15.28	2.23 (1.46-3.42)	<0.001
Gender							
Female	68	36.96	94	45.63		1.0 (Ref.)	-
Male	116	63.04	112	54.37	3.01	1.43 (0.94-2.19)	0.08
Cancer history in first-degree relatives							
No	170	92.39	198	96.12		1.0 (Ref.)	-
Yes	14	7.61	8	3.88	2.53	2.04 (0.78-5.74)	0.11
Alcohol drinking							
Never	97	52.72	129	62.62		1.0 (Ref.)	-
Ever	87	47.28	77	37.38	3.91	1.50 (0.98-2.28)	0.04
Tobacco smoking							
Never	98	53.26	121	58.74		1.0 (Ref.)	-
Ever	86	46.74	85	41.26	1.18	1.25 (0.82-1.90)	0.28
TNM stage at diagnosis							
I-II	69	37.50					
III-IV	115	62.50					
Tumor size, cm							
<5	78	42.39					
≥5	106	57.61					
Lauren classification							
Intestinal	74	40.22					
Diffuse	110	59.78					

TNM = tumor, node, metastasis; Ref. = reference.

The genotype distributions of *ERCC5* rs2094258 and rs751402 polymorphisms are shown in Table 2. The frequencies of AA, AG, and GG rs2094258 genotypes were 100 (54.35%), 67 (36.4%), and 17 (9.24%), respectively, in the patient group, and 121 (58.74%), 72 (34.95%), and 13 (6.31%), respectively, in the control group. The frequencies of CC, CT, and TT rs751402 genotypes were 69 (37.50%), 91 (49.46%), and 24 (13.04%), respectively, in the patient group, and 87 (42.23%), 97 (47.09%), and 21 (10.68%), respectively, among the controls. A chi-square test revealed no significant differences in rs2094258 (chi-square = 1.47, P = 0.48) and rs751402 (chi-square = 1.34, P = 0.51) genotype distributions between patients and controls, which were found to be consistent with HWE in both groups (patients: P = 0.25 and 0.61 for rs2094258 and rs751402, respectively; controls: P = 0.48 and 0.42 for rs2094258 and rs751402, respectively).

Table 2. *ERCC5* rs2094258 and rs751402 polymorphism genotype distributions in patients with gastric cancer and control groups.

Genotype	Cases N = 184	%	Controls N = 206	%	P for HWE		Chi-square	P
					Cases	Controls		
rs2094258								
AA	100	54.35	121	58.74				
AG	67	36.41	72	34.95				
GG	17	9.24	13	6.31	0.25	0.61	1.47	0.48
rs751402								
CC	69	37.50	87	42.23				
CT	91	49.46	97	47.09				
TT	24	13.04	22	10.68	0.48	0.42	1.34	0.51

HWE = Hardy-Weinberg equilibrium.

The association between *ERCC5*rs2094258 and rs751402 polymorphisms and gastric cancer risk is shown in Table 3.Using unconditional logistic regression analysis, we found that these SNPs were not associated with an increased risk of gastric cancer under co-dominant, dominant, or recessive models (P < 0.05).

Table 3. Association between *ERCC5* rs2094258 and rs751402 polymorphisms and gastric cancer risk.

Genotype	Patients N = 184	%	Controls N = 206	%	OR (95%CI)	P value
Co-dominant						
rs2094258						
AA	100	54.35	121	58.74	1.0 (Ref.)	
AG	67	36.41	72	34.95	1.13 (0.72-1.76)	0.58
GG	17	9.24	13	6.31	1.58 (0.68-3.72)	0.24
Dominant						
AA	100	54.35	121	58.74	1.0 (Ref.)	
AG+GG	84	45.65	85	41.26	1.20 (0.78-1.82)	0.38
Recessive						
AA+AG	167	90.76	193	93.69	1.0 (Ref.)	
GG	17	9.24	13	6.31	1.51 (0.67-3.49)	0.28
rs751402						
Co-dominant						
CC	69	37.50	87	42.23	1.0 (Ref.)	
CT	91	49.46	97	47.09	1.18 (0.76-1.85)	0.44
TT	24	13.04	22	10.68	1.44 (0.70-2.96)	0.28
Dominant						
CC	69	37.50	87	42.23	1.0 (Ref.)	
CT+TT	115	62.50	119	57.77	1.23 (0.80-1.89)	0.32
Recessive						
CC+CT	160	86.96	184	89.32	1.0 (Ref.)	
TT	24	13.04	22	10.68	1.31 (0.67-2.58)	0.39

[1]Adjusted for gender, age, and alcohol consumption status. Ref. = reference.

DISCUSSION

In the present investigation, we conducted a case-control study to assess the association between *ERCC5* gene polymorphisms and gastric cancer risk in a Chinese population.

Previous studies have examined the association between *ERCC5*rs2094258 and rs751402 polymorphisms and the development of certain malignancies, including breast and gastric cancers, salivary gland tumors, and oral squamous cell carcinoma (Duan et al., 2012; Yang et al., 2012; Zavras et al., 2012; Meng et al., 2013; Na et al., 2015; Wang et al., 2015). For instance, Duan et al. (2012) and Yang et al. (2012) both reported that these polymorphisms contribute to the development of gastric cancer. Zavras et al. (2012) carried out a study of 575 oral squamous cell carcinoma patients and 575 controls, finding the CC genotype of the *ERCC5* rs751402 variant to be associated with decreased risk of this cancer. Meng et al. (2013) performed a case-control study including 133 patients and 142 control subjects, from which they established that this same polymorphism may correlate with susceptibility to salivary gland tumors. In another case-control investigation, Na et al. (2015) assessed 324 breast cancer patients and 325 controls, and revealed that the *ERCC5*rs2094258 polymorphism may contribute to the development of this disease. However, in a Chinese study, Wang et al. (2015) reported that *ERCC5* genetic polymorphisms do not affect outcome of treatment for osteosarcoma. Thus, previous studies tend to suggest that *ERCC5*rs2094258 and rs751402 polymorphisms contribute to cancer risk.

Only two studies have examined the association between these genetic variants and development of gastric cancer (Duan et al., 2012; Yang et al., 2012). Duan et al. (2012) conducted an investigation into the association between these two *ERCC5* promoter SNPs and gastric cancer in a Chinese population, concluding that rs751402 does affect the risk of developing this malignancy. In another Chinese study of 337 newly diagnosed gastric cancer cases and 347 healthy controls, Yang et al. (2012) found that rs2094258 is associated with increased risk of this disease. However, in the present work, we failed to establish an association between *ERCC5* rs2094258 and rs751402 polymorphisms and development of gastric cancer among Chinese patients and controls. The discrepancies between the abovementioned results may be explained by differences in the selection of cases and controls, study design, and sample size, as well as by chance.

Two limitations to our study should be considered. First, gastric cancer patients and control subjects were selected from only one location, and thus selection bias may have influenced our findings. However, the genotype distributions of the SNPs under investigation were consistent with HWE, suggesting that the study group was representative of the general population. Second, the sample size of our study was small, which may have reduced its statistical power, and could explain our failure to find a significant association between *ERCC5* rs2094258 and rs751402 polymorphisms and susceptibility to gastric cancer. Thus, further studies involving larger sample sizes are greatly required to confirm our findings.

In conclusion, we suggest that the *ERCC5* rs2094258 and rs751402 polymorphisms are not associated with development of gastric cancer under codominant, dominant, or recessive models. Further studies with large sample sizes are greatly needed to verify the results of our work.

Conflicts of interest

The authors declare no conflict of interest.

ACKNOWLEDGMENTS

We thanks for the supports from nurses in Gansu Provincial Hospital, and these nurses help use to collect blood samples from included subjects.

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