

The ERCC2 K751 polymorphism is associated with breast cancer risk

Mojgan Hosseini¹, Masoud Houshmand^{2,3}, Ahmad Ebrahimi²

¹Islamshahr Branch, Islamic Azad University, Islamshahr, Tehran, Iran

²National Institute for Genetic Engineering and Biotechnology (NIGEB), Tehran, Iran

³Special Medical Center, Tehran, Iran

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Corresponding author:

Mojgan Hosseini, PhD
Department of Science
Islamshahr Branch
Islamic Azad University
Sayad Shirazi St. Islamshahr
Tehran, Iran
Phone/fax: +98 21 66936779
E-mail: Mojgan-Hosseini@iaau.ac.ir,
moj.hosseini@gmail.com

Abstract

Introduction: Numerous studies are addressing associations of polymorphisms in DNA repair genes and cancer risks, because accurate and efficient DNA repair is crucial to genomic integrity and fidelity. ERCC2 are important in DNA nucleotide excision repair and lie on chromosome 19q13.3. We genotyped constitutive variants ERCC2 K751Q and R156R in approximately 400 adults with breast adenocarcinoma and 160 controls of Iranian women.

Material and methods: Totally 560 Iranian sporadic breast cancer affected women compare to control group were studied by PCR-RFLP for ERCC2 (K751Q and R156R).

Results: Our results showed that heterozygote genotype ERCC2 (751) has the highest frequency in both groups (21.8 in patients and 8.7 in control group). The genotype ERCC2 (751) GT were most risk factor in our population [GG/TT odds ratio, 5.90 (95% confidence interval, CI 11.45-12.15) $p = 0.001$, GG/GT odds ratio, 4.737 (95% CI, 9.03-9.92) $p = 0.029$, TT/GT odds ratio, 5.465 (95% CI, 10.41-11.45) $p = 0.002$].

Conclusions: We conclude that not only G/G and T/T in our patients was not associated with breast cancer risk but also there is a relation between presence of G/T and increasing of breast cancer risk.

Key words: ERCC2 gene, polymorphism, breast cancer, PCR-RFLP, susceptibility factor.

Introduction

Breast cancer is the second common cancer in the world and the first common cancer in Iranian women in rate [1].

The DNA repair system is complex, governed by more than 125 genes, many of which are polymorphic [2]. Two DNA repair genes, ERCC2 K751Q and R156R, whose products are important in nucleotide excision repair lie near each other on chromosome 19q13.3.

Variation in efficiency of these processes might influence either cancer development (defective or inefficient repair could lead to accumulation of deleterious mutations in the absence of apoptotic destruction of DNA-damaged cells) or cancer progression (by any of the previously mentioned mechanisms or by more efficient repair reducing effectiveness of chemotherapy aimed at DNA damage and resultant reduction of cancer cell killing).

Recently demonstrated that down regulation of ERCC2, induce up regulation of cdk-activating kinases and mitotic progression [3].

There were some previous studies on the frequency of these polymorphisms and the allele frequencies of the two silent nucleotide substitutions for ERCC2 in another Caucasian study [4-6].

We reported here the allele frequencies of ERCC2 K751 and R156R in breast cancer patients in Iran country.

Material and methods

Patients data

Analyses were conducted for the 400 patients and 160 controls genotyped for ERCC2, including 182 patients, 76 control of premenopausal women and 218 patients, 84 control of postmenopausal women and ages were 35-55 years.

This study ethically was approved by the local Ethical Committee of Islamic Azad University from the point of view of patients' and also control group samples rights.

All patients participated in the Special Medical Centre, part of chemotherapy, Tehran, Iran. A questionnaire including questions on breast cancer risk factors were completed and Each patient filled a consent form. The blood samples were collected from patients and controls prior to start of treatment. Subjects were genotyped for the ERCC2 SNPs using genomic DNA Extracted from peripheral blood lymphocytes. DNA was isolated from peripheral blood using FelxiGene DNA extraction kit (Qiagen Germany).

Genotyping

The polymorphisms were detected using a modified PCR-RFLP method [7, 8]. The PCR primers were synthesized by TAG Copenhagen A/S Primers were for each polymorphism are as follows.

ERCC2-R156R (rs238406) (245 bp), Forward: ACCCTGTCTGGGTGCTAAGA.

Reverse: AATTCTGGGACAAGAGTGC [8], the cycling conditions were 94°C, 30 min, 56°C, 30 min (35 cycle), 72°C, 60 min. The PCR products were digested with 1 unit of HinfI.

ERCC2-K751Q (rs13181)(184 bp), Forward: CCCCTCTCCCTTCTCTG.

Reverse: AACGAGGCCAGGCAAGAC, the cycling conditions were 94°C, 30 min, 55.5°C, 30 min (35 cycle), 72°C, 60 min. The PCR products were digested with 1 unit of MboII, and separated on a 6% acrylamid gel.

This method is able to detect all three possible genotypes for the polymorphism: homozygous wild type, heterozygous variant type and homozygous variant type.

The genotypes and allelic frequencies of ERCC2 K751Q and R156R polymorphisms in patient and

control groups were analyses by χ^2 and Fisher's exact tests.

Results

Analyses of affected and controls show that heterozygote genotype ERCC2 (751) has the highest frequency in both groups (21.8 in patients and 8.7 in control group).

The homozygote genotype in ERCC2 (156), has an increase in patients group (13.1) compared with controls (4.9) also (Table I).

Comparison between genotypes, odds ratio and *p* value. Showed that *p* value of our results showed that the genotype ERCC2 (751) GT were most risk factor in our population: GG/TT odds ratio, 5.90 (95% confidence interval, CI 11.45-12.15) *p* = 0.001, GG/GT odds ratio, 4.737 (95% CI 9.03-9.92) *p* = 0.029, TT/GT odds ratio, 5.465 (95% CI 10.41-11.45) *p* = 0.002.

Discussion

There have been several studies of ERCC2 variants with other cancers, most notably with head and neck cancers, lung cancer, and skin cancer [2, 9-15].

ERCC2 are potentially relevant to cancer because of their involvement in the process of nucleotide excision repair (NER) [10].

In addition to the direct role of ERCC2 in DNA repair find, the complexes it forms with the transcription factor TFIIH and other molecules are involved in transcription activation or with cdk-activating kinases that lead to involvement with apoptosis and phosphorylation of nuclear receptors [9].

But some ideas was agreement to our about suggested K751, Butkiewicz *et al.*, who is that

Table I. ERCC2 K751Q and R156R genotype frequencies (*n* [%]) for cases and control. Analyses of 400 affected women and 160 controls for ERCC2 K751Q and R156R genotype frequency shows that G/T ERCC2 K751Q genotype has the highest frequency in both group (21.8 in cases and 8.7 in control group). The G homozygote genotype R156R has an increase in case group (13.1) compared with controls (4.9) and TT ERCC2 K751Q genotype has a decrease in case group (5.1) compared to control group (1.6)

Genotype	Case <i>n</i> [%]	Control <i>n</i> [%]	Total
ERCC2 (156)			
GG	72 (13.1)	27 (4.9)	99 (18.0)
GT	72 (13.1)	26 (4.7)	98 (17.8)
TT	56 (10.2)	22 (4.0)	78 (14.2)
ERCC2 (751)			
GG	52 (9.5)	18 (3.3)	70 (12.7)
GT	120 (21.8)	48 (8.7)	168 (30.5)
TT	28 (5.1)	9 (1.6)	37 (6.7)

suggested linkage disequilibrium for codons 312 and 751 [16].

Thus, in other study, observed increase in breast cancer risk may be fully attributed to the influence of the polymorphic *ERCC2* gene [17].

To our knowledge, no studies have published the associations of *ERCC2* K751Q and R156R polymorphisms and breast cancer in Iranian population-based studies yet and this is the first study in Iran.

In the other study, one of breast cancer and 15 at high risk of breast cancer based on family history [18], Benhamou *et al.* find associations between XPD/*ERCC2* single nucleotide polymorphisms and breast cancers [9].

Justenhoven *et al.* find an increased breast cancer risk for female carriers of the *ERCC2*_6540_GG genotype in a German population [17].

On the other hands, Jing Shen was found to be statistically significantly associated with elevated breast cancer risk [4].

Furthermore, Kuschel *et al.* such as no find significantly associated in three single nucleotide polymorphisms rs1799783 in intron 4 (IVS4 a > g), rs1799793 in exon 10 (D312N), and rs1052559 in exon 23 with the incidence of breast cancer [19]. However we identified an increased breast cancer risk for woman carriers of the *ERCC2* K751_GT genotype in an Iranian population.

We in this study, conclude that there is a relation between presence of G/T of *ERCC2* K751 genotype and increasing of breast cancer risk.

In conclusion, we conclude that not only G/G and T/T in our patients was not associated with breast cancer risk but also there is a relation between presence of G/T and increasing of breast cancer risk.

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