Ku80 POLYMORPHISM AND BREAST CANCER RISK - A REVIEW

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Abstract

The aim of this review work is to highlight the association between the genotypes of ku80 gene and the breast cancer risk. In a hospital based case- control study, 1272 breast cancer patients and 1272 healthy controls of the same age and sex were genotyped. A significantly different distribution was observed in the frequency of the ku80 G-1401T genotype only, between the breast cancer patients and control groups. The T allele of the ku80 G-1401T may be associated with the development of breast cancer. It also may be a very useful marker for the detection of breast cancer.

Keywords

Breast cancer, ku80 gene, polymorphism, DSB, HR, NHEJ

Introduction

Breast cancer is the most prevalent female cancer worldwide and the etiology of breast cancer is largely unknown. Epidemiological studies suggest that the etiology of breast cancer is multifactorial, such as: 1) exposure to ionizing radiation, 2) high fat dietary intake, 3) alcohol consumption, 4) the use of hormones or oral contraceptives *etc*. However, only a small proportion of women thus exposed develop breast cancer, suggesting that genetic susceptibility plays a role in the individual risk of breast cancer. The appropriate response of the cell to genetic injury and its ability to maintain genomic stability by means of a variety of DNA repair mechanisms are essential in preventing tumor initiation and progression. So, mutations or defects in the DNA repairing system are essential for tumor genesis. Among the types of DNA damage, double strand breaks (DSBs) may lead to dramatic genomic instability, which is closely related to carcinogenesis. There are two specific DNA repair pathways responsible for DSB repair: 1) Homologous Recombination (HR), 2) Non Homologous End Joining (NHEJ). The role of BRCA1 and the homologous recombination (HR) pathway in breast cancer has been extensively studied. But there is also an alternative repair pathway for DSBs — that is Non Homologous End Joining (NHEJ). Most DSBs repaired by NHEJ, involving several key components. Once DSBs occur in genomic DNA, they are recognized by a DNA binding heterodimer - KU. KU is formed from ku70 and ku80. In an earlier study, the expression and biological significance of the KU70/80 heterodimer in the different

nuclear classes of breast cancer was assessed [1]. The conclusion of the study was that the KU70/80 may play a role in DNA DSBs repair in HR- deficient tumors. In another study, Ku80 polymorphism has been reported to play a role in breast cancer development [2]. The ku80 gene is located on chromosome 2q35. This gene has 21 exons. Ku80 is encoded by the XRCC5 gene that contains a variable number tandem repeat (VNTR) insertion in its promoter region [3].

In a hospital based case control study, the association of ku80 G-1401T rs 828907, ku80 C-319T rs 11685387 and ku80 intron 19 rs 9288518 polymorphisms with breast cancer risk in a Taiwanese population was investigated by Hwei- Chung Wang *et al*., 2009, for the first time [2].

Patients and Methods

Study population and sample collection

The study population consisted of 1272 breast cancer patients and 1272 cancer free control volunteers. The equal number of non-cancer healthy volunteers as controls were selected by matching for age and gender. The exclusion criteria of the control group included any previous or current malignancy metastasized cancer from other or unknown origin (other cancers with previous diagnosis) and any familial or genetic diseases.

Genotyping assays

Genomic DNA was prepared from peripheral blood leukocytes. The following primers were used:

1) For ku80 G-1401T
   5' TAGCTGACAACCTCACAGAT 3'
   and 5' ATTCAGAGGTGCTCATAGAG 3'
2) For ku80 C-319T
   5' AACTCTGAGCATGCGCAGAT 3' and
   5' TCTAACTCCAGAGCTCTGAC 3'
3) For ku80 intron 19
   5' GGTGTGAAGACCTATCAATC 3' and
   5' TTACAGAACAAGCCTTGCAC 3'

The following cycling conditions were performed:

1) one cycle at 94 degrees C for 5 minutes
2) 35 cycles of 94 degrees C for 30 s, 55 degrees C for 30 s, and 72 degrees C for 30 s and a final extension at 72 degrees C for 10 minutes

The PCR products are studied after digestion with restriction enzymes.

Results

The frequency distribution of the selected characteristics of the breast cancer patients and controls are shown in Table 1 and were all well matched. There is not any significant difference between two groups. Table 1 shows the frequency distributions among breast cancer patients and controls.

Table 1:

Characteristics	Control	%	Patient	%
>50 years	517	40.6	475	37.3
<50 years	755	59.4	797	62.7
Breast feeding				
Yes	446	35.1	469	36.9
No	826	64.9	803	63.1
Smoker	315	24.8	329	25.9
Non-smoker	957	75.2	943	74.1

Here breast feeding means breast feeding for at least 10 days and smokers means those having smoking habit previously or currently for at least 1 year. The frequency distributions of the genotypes for the ku80 G- 1401T rs 828907, ku80 C-319T rs 11685387, and ku80 intron 19 in the controls and breast cancer patients are shown in table 2.

Table 2:

Genotype	Controls	%	Patients	%
G-1401T				
GG	956	75.2	833	64.5
GT	230	18.1	309	24.3
TT	86	6.7	130	10.2
C-319T				
CC	170	13.4	155	12.2
CT	303	23.8	322	25.3
TT	799	62.8	795	62.5
Intron 19				
AA	132	10.4	141	11.1
AG	402	31.6	373	29.3
GG	738	58.0	758	59.6

This table shows the distribution of the ku80 genotypes among breast cancer patients and controls. The genotype distribution of the genetic polymorphisms of ku80 G-1401T was significantly different between the breast cancer and control groups. But those for C-319T and intron 19 were not significantly different. To sum up, the ku80 G-1401T and homozygous TT genotypes were significantly associated with breast cancer susceptibility. The frequency distributions of the alleles for ku80 G-1401T, C-319T and intron 19 in the controls and breast cancer patients are shown in table 3.

Table 3:

Alleles	Controls	%	Patients	%
G-1401T				
Allele G	2142	84.2	1975	77.6
Allele T	402	15.8	569	22.4
C-319T				
Allele C	643	25.3	632	24.8
Allele T	1901	74.7	1912	75.2
Intron 19				
Allele A	666	26.2	655	25.8
Allele G	1878	73.8	1889	74.2

The T allele of the ku80 G-1401T polymorphism was significantly associated with breast cancer. The genotype distribution of the various polymorphisms of ku80 G-1401T was significantly different between breast cancer and control groups who had a smoking habit, while those for C-319T and intron19 were not (Table 4). The T allele frequency was significantly higher in the breast cancer patients who smoked, than in non-cancer controls and the patients who did not smoke. The table 4 represents the ku80 G-1401T genotype and breast cancer after stratification by cigarette smoking.

Table 4:

Variable	Ku80 G-1401T genotype GG	Ku80 G-1401T genotype GT+TT
Smokers		
Controls	170	145
Patients	89	240

Nonsmokers		
Controls	786	171
Patients	744	199

Discussion

In a population based breast cancer case control study, polymorphisms in genes involved in HR (NBS1, RAD52, RAD51, XRCC2 and XRCC3) and NHEJ (Ku70/80 and LIG4) was analysed. These DNA double stranded break repair genes are candidates for breast cancer susceptibility [4]. In another study [5], the following hypotheses were made....1) two single nucleotide polymorphism (SNPs) in Ku70 and XRCC4 were associated with breast cancer risk, 2) A tread toward increased risk of developing breast cancer was found in women harboring a greater number of putative high risk genotypes of NHEJ genes, 3) The association between risk and number of putative high risk genotypes was stronger and more significant in women thought to be more susceptible to estrogen, *i.e.* those with no history of full time pregnancy, and 4) the protective effect conferred by a history of full term pregnancy was only significant in women with a lower number of putative high risk genotypes of NHEJ genes.

However, there was no evidence till the publication of a paper by Hwei-Chung Wang *et al*., in 2009, that there is a significant association between ku80 poymorphism and breast cancer [2]. This was the first study which focused on the association between the polymorphisms of ku80 and breast cancer susceptibility. Only ku80 G-1401T was found to have much significance in association with increased breast cancer, while the ku80 C-319T and ku80 intron 19 genotypes had no effect. In the population with a smoking habit, the genetic effect of the ku80 G-1401T on breast cancer risk was much more significant. In the smoking groups, the T allele clearly raised the breast cancer risk. Accordingly it is proposed that the T allele of ku80 G-1401T polymorphism may play a role in carcinogenesis. Non-smokers carrying the T allele may have similar efficiency compared to non T allele carriers in removing double strand breaks, but in smokers, the DNA damage increases significantly and people with the T allele may not have sufficient capacity to remove all the DSBs properly and efficiently, thus increasing their breast cancer risk.

[Note: The results from another study on colorectal cancer provide the first evidence that the T allele of the ku80 G-1401T may be associated with the development of colorectal cancer (6)].

Inference

The T allele of ku80 G-1401T may be associated with the development of breast cancer and may be a novel useful marker for breast cancer detection and prediction.

References

1. Alaa, T. Alshareeda *et al.*, 2013, Clinico-pathological significance of KU70/KU80, a key DNA damage repair protein in breast cancer, Breast Cancer Research and Treatment, 139(2), pp. 301-310.

2. Wang, Hwei-Chung *et al.*, 2009, Significant Association of DNA Repair Gene ku80 Genotypes with Breast Cancer Susceptibility in Taiwan, Anticancer Research, 29(12), pp. 5251-5254.

3. Journal.frontiersin.org>fonc.2016.00092

4. Kuschel, Bettina *et al.*, 2002, Variants in DNA double strand break repair genes and breast cancer susceptibility, Hum. Mol. Genet.,11(12) pp.1399-1407.

5. Yi- Ping Fu *et al*., 2003, Breast Cancer Risk Associated with Genotypic Polymorphism of the Non homologous End- Joining Genes

6. Yang, Mei - Due *et al.*,2009, Significant Association of ku80 single Nucleotide Polymorphisms with Colorectal Cancer Susceptibility in Central Taiwan; Anticancer Research, 29(6), pp. 2239- 2242.