Int J Clin Exp Pathol 2014;7(9):6262-6269
www.ijcep.com /ISSN:1936-2625/IJCEP0001511

Original Article

Prevalence of BRCA1 gene mutation in breast cancer patients in Guangxi, China

Liping Sun*, Junjie Liu*, Sida Wang, Yuanyuan Chen, Zhixian Li

*Department of Diagnostic Ultrasound, The First Affiliated Hospital of Guangxi Medical University, Nanning, Guangxi, China. *Equal contributors.*

Received July 22, 2014; Accepted August 23, 2014; Epub August 15, 2014; Published September 1, 2014

Abstract: Objective: The prevalence of breast cancer susceptibility gene 1 mutation in breast cancer patients of south China has not been well revealed. This study was to invest the prevalence of BRCA1 gene mutation in breast cancer patients in Guangxi, China, and to try reflecting its relevance in genetic counseling of breast cancer. Methods: In this study, 463 breast cancer patients and 30 healthy women (control group) were involved. Entire sequence and splicing sites of BRCA1 genes were detected by PCR-DNA sequencing. Results: About 8.9% (41/463) patients were with 22 BRCA1 mutations (all in exon 10). The average hospitalized age of BRCA1-associated breast cancer cases was significantly younger (t = -2.965, P = 0.003). The nuclear grade (U = 2321.0, P = 0.030), ER (U = 4343.5, P = 0.041) and CerbB-2 (U = 3894.0, P = 0.038) expression levels, and triple negative breast cancer diagnosing rate (χ^2 = 4.719, P = 0.03) were disclosed more in BRCA1-associated patients. Conclusions: The four most frequent BRCA1 mutation (2798 T > C, 3971 G > A, 3971 G > A and 624 C > T) found in female breast cancer cases in Guangxi are all located in exon 10. BRCA1-associated breast cancer cases have earlier onset age, higher nuclear grade and negative ER and CerbB-2 expression.

Keywords: Breast cancer, BRCA1 gene mutation, pathology, histopathology

Introduction

Worldwide, breast cancer is the most common invasive cancer in women. The incidence of breast cancer is very high in western countries (especially in western countries) [1-3]. The characteristics of the cancer decide the treatment of choice, including surgery, medications (hormonal therapy and chemotherapy), radiation and immunotherapy [4]. Prognosis and survival rates for breast cancer vary greatly according to the cancer type, stage, treatment, and geographical location of the patient.

Breast cancer susceptibility gene 1 (BRCA1, GeneBank: AY273801.1) is one of the most important tumor suppressor genes associated with breast cancer in recent years [5]. It has been shown that abnormality in BRCA1 gene structure and function was likely to cause defect in the mechanism of cell apoptosis [6], thus it would play a role in the development process in breast cancer and ovarian cancer [7]. When mutations happened in the BRCA1 gene, the encoded protein led to the reduction or loss of function, which make the blockage in cell differentiation, lose the role of the normal differentiation to proliferation, and mutations in the BRCA1 may also make the possibility of cell malignant transformation and tumor increased greatly by blocking the normal physiological function of wild-type BRCA1. The Western and a few Asian countries reported the prevalence of BRCA gene mutation [3, 8-12], but this was rarely reported in China is fewer; only one relevant report had been released about Guangzhou area [13].

Estrogen receptor (ER) and progesterone receptor (PR), proto-oncogenes C-erbB-2, P53, and P16, etc are the molecular markers closely related with breast cancer occurrence and development [14, 15]. But there are few reports about the P53 and P16 associated with breast cancer imaging characteristics; and the molecular biology indexes has not been clear.

To investigate the BRCA1 mutation prevalence in breast cancer patients in Guangxi region, China, this study was aimed to detect the "hot-

Table 1. Pathological type of 463 breast cancer patients, 465 lesions

Malignant lesions	Number of cases
Infiltrating ductal carcinoma	387
Intraductal carcinoma	22
Intraductal carcinoma with local infiltrating ductal carcinoma	13
Invasive lobular carcinoma	8
Lobular carcinoma in situ	1
Medullary carcinoma	1
Colloid carcinoma	4
Myoepithelial carcinoma	2
Paget disease (or with Intraductal carcinoma)	7
Intraductal papillary carcinoma	5
Low-grade myofibroblastic sarcoma	1
Malignant phyllodes tumor	4
Invasive cribriform carcinoma	3
Invasive tubular carcinoma	4
Small cell neuroendocrine carcinoma	1
Infiltrating neuroendocrine carcinoma	1
Mixed type of breast cancer (invasive lobular carcinoma complicated with invasive ductal carcinoma)	1
Total	465

spot" of BRCA1 gene mutation and to give guidance in gene detection, diagnosis and future treatment in Guangxi region, China.

Materials and methods

Subjects

All participants were given written informed consent about this study, after the researchers explained the use of blood samples and study content, the participants signed for consent. Ethical approval was granted from ethics committee of Guangxi Medical University.

A total of 987 blood samples were collected before surgery or biopsy, between May, 2010 and December 2012, from patients with breast lumps hospitalized in the Gastrointestinal and gland surgery in the first affricated hospital of Guangxi medical university. After excluding patients with benign lesions from postoperative pathological results, a total of 463 patients with breast cancer were enrolled for DNA and immunohistochemical detection. Breast cancer patients were with an average age of 47.7 ± 10.9 (18 to 81 years), 57 patients ≤ 35 years old.

Inclusion criteria: all patients had complete clinical and pathological data (including pathologic type, histological grade and ER, PR, CerbB2, P53, and P16 immunohistochemical examination results, etc.).

Exclusion criteria: 1) accepted radiotherapy or chemotherapy before surgery; 2) did not receive surgery or biopsy; 3) postoperative pathological results confirmed to be benign lesions.

The control group was consisted of DNA extracted from blood samples of 30 randomly chosen healthy women for the same period.

BRCA1 mutation detection

The study was performed in genomic DNA obtained from whole blood. All blood examples of breast cancer DNA extraction were applied by improved phenol/chloroform extraction.

Primer design

Primer design is extremely important in the PCR reaction. We amplified all the coding sequence of BRCA1 gene exons and introns splice zone of the 463 breast cancer patients according to the BRCA1 DNA sequence (NG_005905. 2) and mRNA sequence (NM_007300. 3) reported on the national center for biotechnology information web site *http://www.ncbi.nlm.nih.gov/gene*. Software Primer premier 5 was

Table 2. 41 gene mutations detected in BRCA1 exon 10

Base change	Codon encoded amino acid sites	Protein change	Mutation type*	N cases	Incidence % (N = 463)
1227th G > A	332	Arg→Gln	M	2	0.4
1268th C > T	346	Pro→Ser	M	1	0.2
1624th C > T	464	Thr→Thr	S	4	0.8
1633th G > A	467	Lys→Lys	S	1	0.2
2029th T > C	599	Asn→Asn	S	1	0.2
2238th T > C	669	Met→Thr	M	4	0.8
2309th G > A	693	Asp→Asn	M	1	0.2
2798th T > C	856	Tyr→His	M	8	1.7
2844th C > T	871	Pro→Leu	M	3	0.6
3236th C > T	1002	Asn→Asp	M	1	0.2
452th the beginning AGAAACA > TT	1083	Termination codon	N	1	0.2
3589th T > A	1119	Thr→Thr	S	1	0.2
3678th C > G	1149	Thr→Arg	M	1	0.2
3709th A > G	1159	Ile→Met	M	1	0.2
3971th G > A	1247	Val→Ile	M	5	1.1
3986th C > G	1252	Leu→Val	M	3	0.6
4030th C > G	1421	Ser→Arg	M	1	0.2
4200th started missing AAAT	1334	Termination codon	F	1	0.2

Footnotes: *Mutation type: M missense mutation; S samesense mutation; N nonsense mutation; F frame-shifting mutation.

Table 3. Pathological type of 43 lesions in 41 cases of BRCA1 mutation-associated breast cancer

Pathological type	BRCA1 mutation (N)	Without BRCA1 mutation (N)
Infitrating ductal carcinoma	33	354
Intraductal carcinoma	3	19
Intraductal carcinoma with local infiltrating	2	11
Intraductal papillary carcinoma	1	4
Malignant phyllodes tumor	1	3
Invasive cribriform carcinoma	2	1
Mixed type of breast cancer	1	-
Infiltrating lobular carcinoma	-	8
Lobular carcinoma in situ	-	1
Medullary carcinoma	-	1
Colloid carcinoma	-	4
Myoepithelial carcinoma	-	2
Paget disease (or with Intraductal carcinoma)	-	7
Low-grade myofibroblastic sarcoma	-	1
Invasive tubular carcinoma	-	4
Small cell neuroendocrine carcinoma	-	1
Infiltrating neuroendocrine carcinoma	-	1
Total	43	422

used for primers design and software Oligo 7 was used for primers evaluation. The mutation analysis of augmented products was directly conducted by DNA sequence. The synthesis of PCR primers was completed by Shanghai Shenggong biological engineering technology service co., LTD.

Direct sequencing of polymerase chain reaction (PCR) products

Purification and sequencing of PCR products were assisted by Shanghai Shenggong biological engineering technology service co., LTD. Unidirectional sequencing was firstly undergone, when BRCA1 gene mutations found out, mutations were all reconfirmed by different primers. Compared the amplification of PCR sequences with the standard sequence in Gene bank (NM_007300. 3) using DNAstar software; if base changes were discovered, mutations could be judged after eliminated SNPs. And Breast Cancer Information core (BIC) *http://www.acro-*

Table 4. Histological grading results of BRCA1 mutation-associated breast cancer

Group	Cases	Histological grading results			Mann-Whitney U	*P*
		I	II	III		
Experimental group	28	2	12	14	2321.0	0.030
Control group	214	30	121	63		

Table 5. Immunohistochemical results of the BRCA1 mutation-associated breast cancer

Immunohistochemical results		Group		Mann-Whitney U	*P*
		Experimental group (31 cases)	Control group (321 cases)		
ER	-	15	102	4343.5	0.041
	+	8	83		
	++	5	61		
	+++	3	75		
PR	-	13	111	4693.0	0.584
	+	8	109		
	++	5	39		
	+++	5	62		
CerbB-2	-	17	108	3752.0	0.019
	+	16	68		
	++	4	71		
	+++	4	74		
P53	-	19	161	4562.0	0.405
	+	5	91		
	++	3	28		
	+++	4	41		
P16	-	11	127	4757.0	0.665
	+	13	132		
	++	4	23		
	+++	3	39		

nymfinder.com/Breast-Cancer-Information-Core-(BIC).html was used for comparison, and determine whether it was the new mutation.

Statistical analysis

All the statistical analyses were performed using the statistical program for social sciences (SPSS) software package version 18.0 (Chicago, IL, USA). The mean ± standard deviation ($\bar{x}$ ± S) was used to represent measurement data, and two independent sample t tests were applied in comparison between groups. Enumeration data was expressed as cases or percentage. Chi-square test was used in comparison between groups. The group comparison between ranked data was applied by Mann-Whitney U test; with $P < 0.05$ represented for the difference was statistically significant.

Results

Pathological type of 463 breast cancer patients

The diagnosis was confirmed by histopathology (**Table 1**) obtained postoperatively, in which one participant was detected with 2 invasive breast cancer lumps in the left breast, and one participant was detected with bilateral breast malignant lesions (infiltrating ductal carcinoma and intraductal papillary carcinoma both in the left breast), and all the other patients were with single lesion.

BRCA1 gene direct sequencing results of 463 cases breast cancer patients

BRCA1 gene mutation was not found in 30 healthy women. In the cohort of 463 patients, 41 mutations (all located in exon 10) were found. And the prevalence of BRCA1 mutation was 8.9%. There were 7 synonymous mutations, 1 nonsense mutation, 1 frame-shifting mutation, and 32 missense mutations (**Table 2**).

General condition of BRCA1 mutation-associated breast cancer patients

Hospitalized BRCA1 mutation-associated breast cancer patients aged 18 to 60 years old, the average age was 42.8 ± 8.8 years old, hospitalized patients without BRCA1 mutations aged 24 to 81 years old, mean age 48.2 ± 11.0 years, the former average hospitalization age was significantly younger ($t = -2.965$, $P = 0.003$).

Table 6. The detection rate of TNBC in BRCA1 mutation-associated breast cancer

Group	Cases	TNBC		c^2	*P*
		Yes	No		
Experimental group	31	11	20	4.719	0.03
Control group	321	61	260		

Pathological type of BRCA1 mutations related breast cancer patients

A total number of 41 cases of BRCA1 mutation breast cancer patients with one case of two invasive breast cancer lumps in the left breast and one case of bilateral breast malignant lesions (infiltrating ductal carcinoma in the right breast, intraductal papillary carcinoma in the left breast), and all the remainings are single lesions; Lesions without BRCA1 mutation are all single. Infiltrating ductal carcinoma is the most common in BRCA1 mutations related breast cancer (80.5%) (**Table 3**).

Histological classification of BRCA1 mutations related breast cancer patients

After excluding 7 cases of BRCA1 samesense mutation patients, 28 cases in the rest 34 cases had histological grading results, and been included in the experimental group, 214 patients without BRCA1 mutations and with histological grading results were classified into control group. The histological grading results of 28 patients in experimental group were higher than 214 patients in control group (U = 2321.0, *P* = 0.030) (**Table 4**).

Immunohistochemical of BRCA1 mutations related breast cancer

After excluding 7 cases of BRCA1 samesense mutation patients, 31 cases in the rest 34 cases had immunohistochemical results, and been included in the experimental group, 321 patients without BRCA1 mutations and with immunohistochemical results were classified into control group (**Table 5**).

35.5% of BRCA1 mutation-associated breast cancers were tripple negative breast cancer (TNBC), 19.0% of group without BRCA1 mutations were TNBC (**Table 6**), and the difference of two groups was statistically significant (χ^2 = 4.719, *P* = 0.03).

Discussion

The occurrence and development of breast cancer are a complex biological process consisting of multi-factor and multi-gene interaction and influence through many steps and phases. The BRCA1 gene closely related to breast cancer development was firstly reported by Hall et al. in 1990 [3], and firstly successfully cloned and separated by positional cloning technology by Miki and coworkers in 1994 [5]. The BRCA1 gene has high penetrations, and passes to the offspring in autosomal dominant inheritance. BRCA1 gene mutation is closely related to the familial early-onset breast cancer occurrence. The gene mutation makes the corresponding gene product change and causes the tumor inhibitory effect to reduce or disappear. The cumulative risk of breast cancer before the age of 70 to BRCA1 gene mutation carriers is as high as 65% [16, 17], not only the onset age of breast cancer is early, bilateral breast cancer risk increases obviously, but also with poor prognosis. Thus, for people at high risk of breast cancer, the BRCA1 gene mutation screening will contribute to breast cancer risk assessment, early diagnosis and gene therapy in the further.

BRCA gene mutation frequency and type have obvious geographical and racial differences. A total of 38 female BRCA mutation carriers were identified out of 487 referrals and 93 BRCA mutation carriers were disclosed in 6179 Jewish women group were found by Metcalfe KA et al. [8] from 2008 to 2012. Spanish scholars [9] extracted genome DNA from 495 sporadic breast cancer patients blood samples, and detected 52 cases of BRCA1/2 mutation, and of which 8 cases (1.6%) were the pathogenic mutations (6 cases of frameshift mutations, 1 case of missense mutations, and 1 case of nonsense mutation). The Hall et al. [10] reported that from 1996 to 2006, the BRCA1/2 pathogenic mutation rate was 12.5% in 46276 cases of female, and also found that the BRCA1/2 pathogenic mutation rate of African-American and Hispanic women is significantly higher than that of western European women. Han [11] found 79 cases of BRCA1/2 mutation in 793 cases of South Korea sporadic breast cancer patients, and 20 cases (2.5%) were pathogenic mutations. The Hartwig et al. [12] investigated 252 female breast cancer patients

in north-central Poland, and found that one out of 252 patients (0.4%) carried the BRCA1 c.68_69delAG gene mutation, and in the group of families with breast cancer history, the frequency of BRCA1 c.68_69delAG was high to 1.4%. Zhou et al. [13] reported that the incidence of familial and early-onset breast cancer BRCA1 gene mutation of Guangdong province, China, were obviously lower than western countries. In this study, we found the relative "hot spots" of BRCA1 gene mutations in female breast cancer patients in Guangxi region, China: (1) 19.5% (8/41) in exon 10 2798th base (T > C); (2) 12.2% (5/41) in exon 10 of 3971 bases (G > A); (3), 9.8% (5/41) in exon 10 of 3971 bases (G > A); (4) 9.8% (5/41) in exon 10 1624th base (C > T). The BRCA gene mutation detection is helpful for early diagnosis and prevention of breast cancer, but it cannot be popularized in clinical because of the expenses. The discovery of the "hot spots" of BRCA1 gene mutation can help reduce costs, improve the efficiency of detection in BRCA gene mutation screening in the future counseling work.

The minimum hospitalized age of BRCA1 mutation-associated breast cancer patients in this study was 18 years old (pathologically proven malignant phyllodes tumor), the average hospitalized age was 42.8 ± 8.8 years old, which was significantly lower than that in group without BRCA1 mutation (mean age, 48.2 ± 11.0 years old). 216 cases with the age of 45 years old or younger, 19.0% (41/216) breast cancer patients were detected with BRCA1 mutation; 70.0% (25/41) of the 41 cases with BRCA1 mutations related to breast cancer had the age of 45 years old or less, so we think it is very necessary for female breast cancer patients with the age of 45 years old or less to take the BRCA1 gene mutation detection, thus to provide reference of postoperative follow-up and regular review plans for breast cancer patients. 41 cases of BRCA1 mutation related breast cancer (including 7 cases of BRCA1 samesense mutations), of which 80.5% (33/41) were infiltrating ductal carcinoma, 7.3% (3/41) were intraductal carcinoma, the rest pathological types were 2 cases of intraductal carcinoma with cell infiltration and invasive cribriform carcinoma respectively, 1 case of malignant phyllodes tumor and mixed type of breast cancer respectively. There was no medullary carcinoma in 41 cases of BRCA1 mutation-associated breast cancer. It may be because the sample of medullary carcinoma in this study is not very rich (only 1 case).

This study also showed that the histological grade of 28 cases with BRCA1 mutation were significantly higher than that of 214 cases without BRCA1 mutation, and with the rise of histological grade, the detection rate of BRCA1 mutation rises, and this result is consistent with Atchley et al. [18] reported.

ER, PR, c-erbB-2, P53, and P16 expression level play important roles in clinical treatment and prognosis judgment. At present most of scholars believe that BRCA1 mutation is closely related to negative ER, PR expression breast cancer [1, 19, 20]. In our study, the ER and CerbB-2 expression intensity in 31 cases of BRCA1 mutation-associated breast cancer was significantly lower than those without BRCA1 mutation, usually presenting as ER and c-erb-B-2 expression negative in BRCA1 mutation-associated breast cancer cases, and with ER, CerbB-2 expression intensity decreased, the detection rate of BRCA1 mutation increased. But there was no statistical significance in PR expression intensity between 31 cases of patients with BRCA1 mutations and those without BRCA1 mutation.

There are other studies showing the relationship between BRCA mutation and TNBC, that 50%-88% BRCA1 mutation-associated breast cancer patients were diagnosed as TNBC, rather than only 14.6%~14.6% were detected as TNBC in patients without BRCA1 mutation breast cancer [19, 21, 22]. This study also further confirmed that the case rate was significant different between BRCA1 mutation patients and those without BRCA1 mutation: the former was 35.5% (11/31) were diagnosed as TNBC, while the latter was 19.0% (61/321) as TNBC patients.

The mutant P53 relates to some adverse factors such as poor tumor differentiation, easily relapse and metastasis, etc. And when missing or mutation of P16, P16 protein synthesis disorder leads cell proliferation to get out of control, and easily lead to the occurrence of tumor. Lagos-Jaramillo et al [22] reported that besides ER and PR negative expression rate in BRCA1 mutation-associated breast cancer is higher than patients without BRCA1 mutation in

Hispanic women, P53 status for high expression. But in this study, the 31 cases of BRCA1 mutations-associated breast cancer (except BRCA1 samesense mutation), the P53, and P16 expression intensity had no significant difference with patients without BRCA1 mutation.

To sum up, in this study, there were 4 relative mutation "hot spots" found in female breast cancer patients in Guangxi region, all of them were located in exon 10, and they were 2798th base (T > C), 3971 bases (G > A), 3971 bases (G > A) and 1624 bases (C > T) , respectively. Compared with patients without BRCA1 mutation, BRCA1 mutation breast cancer patients were with early onset age, high histological grade, the high negative expression rate of ER and CerbB-2.

Acknowledgements

This work was supported by the National Natural Science Foundation of China (NSFC 81160280). We thank Prof. Zhigang Yuan in the Genetics Department of Guangxi Medical University; Prof. Jian Zeng and coworkers in the Gastrointestinal & Gland Surgery of the First Affiliated Hospital of Guangxi Medical University, and pathologists in the Department of Pathology of the First Affiliated Hospital of Guangxi Medical University for their contributions to the research.

Disclosure of conflict of interest

None.

Address correspondence to: Dr. Zhixian Li, Department of Diagnostic Ultrasound, The First Affiliated Hospital of Guangxi Medical University, 22 Shuangyong Road, Nanning 530021, Guangxi, China. Tel: 86-771-5356706; E-mail: lizhixiandoc@163.com

References

[1] Dufloth RM, Carvalho S, Heinrich JK, Shinzato JY, dos Santos CC, Zeferino LC and Schmitt F. Analysis of BRCA1 and BRCA2 mutations in Brazilian breast cancer patients with positive family history. Sao Paulo Med J 2005; 123: 192-197.

[2] Jemal A, Center MM, DeSantis C and Ward EM. Global patterns of cancer incidence and mortality rates and trends. Cancer Epidemiol Biomarkers Prev 2010; 19: 1893-1907.

[3] Hall JM, Lee MK, Newman B, Morrow JE, Anderson LA, Huey B and King MC. Linkage of early-onset familial breast cancer to chromosome 17q21. Science 1990; 250: 1684-1689.

[4] Florescu A, Amir E, Bouganim N and Clemons M. Immune therapy for breast cancer in 2010-hype or hope? Curr Oncol 2011; 18: e9-e18.

[5] Miki Y, Swensen J, Shattuck-Eidens D, Futreal PA, Harshman K, Tavtigian S, Liu Q, Cochran C, Bennett LM, Ding W, et al. A strong candidate for the breast and ovarian cancer susceptibility gene BRCA1. Science 1994; 266: 66-71.

[6] Paterson JW. BRCA1: a review of structure and putative functions. Dis Markers 1998; 13: 261-274.

[7] Catteau A, Harris WH, Xu CF and Solomon E. Methylation of the BRCA1 promoter region in sporadic breast and ovarian cancer: correlation with disease characteristics. Oncogene 1999; 18: 1957-1965.

[8] Metcalfe KA, Poll A, Royer R, Nanda S, Llacuachaqui M, Sun P and Narod SA. A comparison of the detection of BRCA mutation carriers through the provision of Jewish population-based genetic testing compared with clinic-based genetic testing. Br J Cancer 2013; 109: 777-779.

[9] de Juan Jimenez I, Esteban Cardenosa E, Palanca Suela S, Barragan Gonzalez E, Aznar Carretero I, Munarriz Gandia B, Santaballa Bertran A, Torregrosa Maicas MD, Guillen Ponce C, Sanchez Heras AB, Bayon Lara A, Fuster Lluch O and Bolufer Gilabert P. Low prevalence of BRCA1 and BRCA2 mutations in the sporadic breast cancer of Spanish population. Fam Cancer 2012; 11: 49-56.

[10] Hall MJ, Reid JE, Burbidge LA, Pruss D, Deffenbaugh AM, Frye C, Wenstrup RJ, Ward BE, Scholl TA and Noll WW. BRCA1 and BRCA2 mutations in women of different ethnicities undergoing testing for hereditary breast-ovarian cancer. Cancer 2009; 115: 2222-2233.

[11] Han SH, Lee KR, Lee DG, Kim BY, Lee KE and Chung WS. Mutation analysis of BRCA1 and BRCA2 from 793 Korean patients with sporadic breast cancer. Clin Genet 2006; 70: 496-501.

[12] Hartwig M, Janiszewska H, Bak A, Pilarska M, Heise M, Junkiert-Czarnecka A, Laskowski R and Haus O. Prevalence of the BRCA1 c.68_69delAG (BIC: 185delAG) mutation in women with breast cancer from north-central Poland and a review of the literature on other regions of the country. Contemp Oncol (Pozn) 2013; 17: 34-37.

[13] Zhou J, Rao NY, Li SR, Jin L, Jia WJ, Gong C, Yu FY, Su FX, Song EW and Shao ZM. [Analysis of BRCA1 gene mutations in patients with early-onset breast cancer and their affected relatives in Guangdong province]. Nan Fang Yi Ke Da Xue Xue Bao 2009; 29: 213-216.

[14] Chen JH, Baek HM, Nalcioglu O and Su MY. Estrogen receptor and breast MR imaging features: a correlation study. J Magn Reson Imaging 2008; 27: 825-833.
[15] Jiang L, Ma T, Moran MS, Kong X, Li X, Haffty BG and Yang Q. Mammographic features are associated with clinicopathological characteristics in invasive breast cancer. Anticancer Res 2011; 31: 2327-2334.
[16] Loizidou M, Marcou Y, Anastasiadou V, Newbold R, Hadjisavvas A and Kyriacou K. Contribution of BRCA1 and BRCA2 germline mutations to the incidence of early-onset breast cancer in Cyprus. Clin Genet 2007; 71: 165-170.
[17] Antoniou AC, Cunningham AP, Peto J, Evans DG, Lalloo F, Narod SA, Risch HA, Eyfjord JE, Hopper JL, Southey MC, Olsson H, Johannsson O, Borg A, Pasini B, Radice P, Manoukian S, Eccles DM, Tang N, Olah E, Anton-Culver H, Warner E, Lubinski J, Gronwald J, Gorski B, Tryggvadottir L, Syrjakoski K, Kallioniemi OP, Eerola H, Nevanlinna H, Pharoah PD and Easton DF. The BOADICEA model of genetic susceptibility to breast and ovarian cancers: updates and extensions. Br J Cancer 2008; 98: 1457-1466.
[18] Atchley DP, Albarracin CT, Lopez A, Valero V, Amos CI, Gonzalez-Angulo AM, Hortobagyi GN and Arun BK. Clinical and pathologic characteristics of patients with BRCA-positive and BRCA-negative breast cancer. J Clin Oncol 2008; 26: 4282-4288.
[19] Lee E, McKean-Cowdin R, Ma H, Spicer DV, Van Den Berg D, Bernstein L and Ursin G. Characteristics of triple-negative breast cancer in patients with a BRCA1 mutation: results from a population-based study of young women. J Clin Oncol 2011; 29: 4373-4380.
[20] Zakhartseva LM, Gorovenko NG, Podolskaya SV, Anikusko NF, Lobanova OE, Pekur KA, Kropelnytskyi VA and Shurygina OV. Breast cancer immunohistochemical features in young women with BRCA 1/2 mutations. Exp Oncol 2009; 31: 174-178.
[21] Musolino A, Bella MA, Bortesi B, Michiara M, Naldi N, Zanelli P, Capelletti M, Pezzuolo D, Camisa R, Savi M, Neri TM and Ardizzoni A. BRCA mutations, molecular markers, and clinical variables in early-onset breast cancer: a population-based study. Breast 2007; 16: 280-292.
[22] Lagos-Jaramillo VI, Press MF, Ricker CN, Dubeau L, Mai PL and Weitzel JN. Pathological characteristics of BRCA-associated breast cancers in Hispanics. Breast Cancer Res Treat 2011; 130: 281-289.