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Research article

BRCA1 and *BRCA2* mutations in a population-based study of male breast cancer

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Abstract

Background: The contribution of *BRCA1* and *BRCA2* to the incidence of male breast cancer (MBC) in the United Kingdom is not known, and the importance of these genes in the increased risk of female breast cancer associated with a family history of breast cancer in a male first-degree relative is unclear.
Methods: We have carried out a population-based study of 94 MBC cases collected in the UK. We screened genomic DNA for mutations in *BRCA1* and *BRCA2* and used family history data from these cases to calculate the risk of breast cancer to female relatives of MBC cases. We also estimated the contribution of *BRCA1* and *BRCA2* to this risk.
Results: Nineteen cases (20%) reported a first-degree relative with breast cancer, of whom seven also had an affected second-degree relative. The breast cancer risk in female first-degree relatives was 2.4 times (95% confidence interval [CI] = 1.4–4.0) the risk in the general population. No *BRCA1* mutation carriers were identified and five cases were found to carry a mutation in *BRCA2*. Allowing for a mutation detection sensitivity frequency of 70%, the carrier frequency for *BRCA2* mutations was 8% (95% CI = 3–19). All the mutation carriers had a family history of breast, ovarian, prostate or pancreatic cancer. However, *BRCA2* accounted for only 15% of the excess familial risk of breast cancer in female first-degree relatives.
Conclusion: These data suggest that other genes that confer an increased risk for both female and male breast cancer have yet to be found.

Keywords: *BRCA1*, *BRCA2*, family history, male breast cancer

Introduction

Male breast cancer (MBC) is a rare disease and little is known about its aetiology. However, female first-degree relatives of MBC cases are at increased risk of breast cancer [1–6], which suggests that there is an inherited component to the disease. Several genes that are associated with a high lifetime risk of breast cancer in women have been identified during the past decade. One of these, *BRCA2*, has also been shown to confer a significant risk of breast cancer in men, and a recent study found the risk of breast cancer in male *BRCA2* mutation carriers from multiple case breast/ovarian cancer families to be 80-fold higher than in the general population [7]. This equates to a 7% risk of breast cancer by age 80. The

CI = confidence interval; MBC = male breast cancer; PCR = polymerase chain reaction; SSCA/HA = single strand conformation analysis/heteroduplex analysis.

prevalence of *BRCA2* mutations in MBC cases unselected for family history has been estimated in several studies from Europe and the United States [8–14]. The estimated mutation carrier frequencies varied from 4% to 40%, but were higher in the three studies carried out in populations in which founder mutations are known to occur [9,12,13]. Two other studies have screened for the *BRCA2* founder mutation 6174delT in male breast cancer cases of Jewish origin unselected for family history. Struewing *et al.* identified 15 mutation carriers in 100 cases [15] and Sverdlov *et al.* found no mutation carrier in 20 cases [16].

A putative association between *BRCA1* and MBC is less clear. MBC is known to occur in multiple breast/ovarian cancer case families with mutations in *BRCA1* [17], but a study of 22 MBC families found strong evidence against linkage to the *BRCA1* locus [18]. The two studies of the Jewish *BRCA2* founder mutation described above also screened for the *BRCA1* founder mutation 185delAG. Sverdlov *et al.* identified no mutation carrier in 20 cases [16] and Struewing *et al.* identified four mutation carriers out of 110 cases [15]. However, in two other studies of MBC in populations without common founder mutations, no mutation carriers were identified in a total of 72 cases [10,13]. Together, these data suggest that the MBC risk associated with *BRCA1* is, at best, very small.

The purpose of this study was to estimate the breast cancer risks in women with a family history of male breast cancer and to establish the prevalence of *BRCA1* and *BRCA2* mutations in a male breast cancer cases series from the UK.

Materials and methods

Cases were identified from an ongoing population-based study of male breast cancer. All men diagnosed with breast cancer in the areas served by the East Anglia, Trent and West Midlands cancer registries who were alive on 31 December 1998 were eligible to take part, excluding those whose general practitioner thought them unsuitable for the study because of severe concurrent illness. Participants were asked to complete a detailed epidemiological questionnaire, which includes details of a family history of cancer, and to provide a blood sample for genetic analysis. The study has approval from the Anglia and Oxford multi-centre research ethics committee. One hundred and sixty-five eligible patients were identified, of whom 137 have so far agreed to take part. The first 94 patients who enrolled in the study were included in the current analysis.

Pedigree data from the 94 index cases were used to estimate the risk of breast cancer associated with a family history of breast cancer in a male first-degree relative. The program PERSON YEARS [19] was used to calculate the expected number of breast cancers in female first-degree relatives based on age and period specific breast cancer incidence rates. Individuals entered the at risk cohort on 1 January 1960 and were censored on diagnosis of cancer, on death, or on the date the family history questionnaire was completed. Relatives born before 1890 and those who died or developed cancer before 1960 were excluded because reported data for these individuals was likely to be less reliable.

BRCA1 and *BRCA2* mutation analysis was performed as previously described [20]. The whole coding sequence, including intron–exon boundaries, was screened using a combination of single strand conformation analysis/heteroduplex analysis (SSCA/HA) and direct fluorescent sequencing. DNA from six patients could not be reliably amplified by PCR and these were excluded from the results.

Results

The mean age of diagnosis of the cases was 67.3 years (range 36–89 years). Forty-five cases reported a history of cancer in at least one first-degree relative. Of these, 18 reported a history of breast cancer in a female first-degree relative, and one reported a brother with breast cancer. In 8125 person years of observation of first-degree female relatives 16 breast cancers were observed compared to 6.63 expected, which equates to a relative risk of 2.4 (95% CI = 1.4–4.0).

We considered deleterious mutations to be those that are predicted to result in protein truncation (frameshift, splice site and nonsense mutations), or those missense mutations that have been previously shown to be disease associated on the basis of their co-segregation with disease in families. No disease-associated mutations were identified in *BRCA1*. Five disease-associated variants were identified in *BRCA2* (Table 1). These were all in the coding region and are predicted to lead to a truncated protein, and all but one (2192delC) has been previously reported on the Breast Information Core database [21]. The mean age at diagnosis in the mutation carriers was 58.8 years (range 48–69 years), which was 9 years younger than in non-mutation carriers (67.9 years, range 36–89 years). The five mutation carriers all reported a family history of cancer (Table 1). One had a single second-degree relative affected with breast cancer, and the other four had at least one first-degree relative affected with one of the cancers thought to be associated with *BRCA2* (breast, prostate or pancreas). We also identified rare missense mutations of unknown significance in five patients (Table 1) and several common polymorphisms in both *BRCA1* and *BRCA2*.

Discussion

Our estimate of the familial risk of breast cancer associated with a family history of male breast cancer is similar to other published estimates. However, our estimate is based on a reported family history of breast cancer that

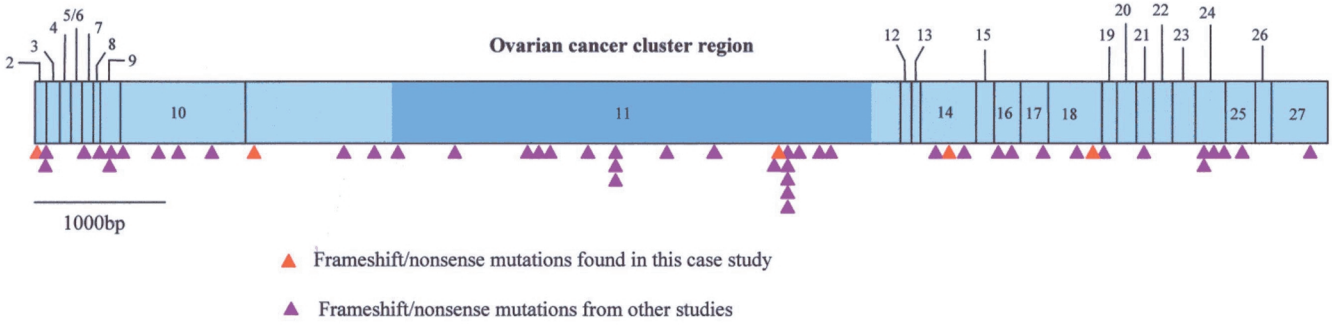
Table 1

Details of rare gene variants

Patient	Gene	Variant	Effect	Age*	Family history (age at diagnosis)	
					First-degree relatives	Second-degree relatives
Protein truncating mutations						
M0271	*BRCA2*	253delC	Frameshift	48	None	MGM Br(?age)
M0041	*BRCA2*	2192delC	Frameshift	55	F Pa(74)	PU Pa(42), PA Br(48), PA Pa(63)
M0040	*BRCA2*	5974delCT	Frameshift	60	F Pr(61)	None
M0293	*BRCA2*	7928delCT	Frameshift	69	M Br(67)	None
M0238	*BRCA2*	8474delAG	Frameshift	62	F Pr + Ki(75)	None
Missense variants of unknown significance						
M0383	*BRCA1*	2640C > T	R841W	63	None	None
M0016	*BRCA2*	5972T > C	T1915M	74	F HN(85), B Lu(50's)	None
M0021	*BRCA2*	5972T > C	T1915M	72	None	None
M0288	*BRCA2*	4486G > T	D1420Y	55	M Br(77)	None

*Age at diagnosis (years). F, father; M, mother; B, brother; MGM, maternal grandmother; PU, paternal uncle; PA, paternal aunt; Br, breast cancer; HN, head and neck; Ki, kidney cancer; Lu, lung cancer; Pa, pancreatic cancer; Pr, prostate cancer.

Figure 1



Summary of the location of published mutations in *BRCA2* identified in male breast cancer cases (red triangles, this study; purple triangles, other published studies).

was not independently confirmed. It is known that a family history of breast cancer is generally accurately reported by women [22], but it may be less accurate for men, for whom some degree of under-reporting is likely. This may result in an underestimate of the familial risk.

As with other studies, we found no *BRCA1* mutation carriers in our case series. The carrier frequency of *BRCA2* mutations was 6% (95% CI = 2–13%), which is broadly comparable with that of previous estimates, but likely to be an underestimate of the true carrier frequency. For example, some of the missense variants that we identified may disrupt the protein structure substantially, but in the absence of a good functional assay for *BRCA1* or *BRCA2* these variants were not classified as disease causing. In addition, the sensitivity of SSCP/HA is known to be less than 100%, and for nonsense mutations may be as low as 50%. Finally, large genomic rearrangements which will not be identified by PCR-based mutation detection methods are known to occur in *BRCA1* and *BRCA2*. A study combining linkage and mutation data in multiple case families has estimated the sensitivity of the most commonly used mutation detection techniques for these genes to be approximately 70% [23]. Based on this sensitivity, the frequency of *BRCA2* mutations in this series could be as high as 8%. Survival bias may also affect the estimate of carrier mutation frequency. The participants in this study were prevalent cases and carrier frequency may be either underestimated or overestimated if male breast cancer associated with a *BRCA1* or *BRCA2* mutation carries a different prognosis from breast cancer in men without a mutation. However, there are no published data

on outcome in *BRCA*-associated MBC, and in women the effect of *BRCA* status on outcome is not clear [24,25].

The risk of female breast and ovarian cancer is thought to vary depending on the location of mutations throughout the *BRCA2* gene [7]. Mutations in a central portion of the gene, referred to as the ovarian cancer cluster region, are associated with either an increase in the risk of ovarian cancer, a decrease in the risk of breast cancer, or a combination of both, compared with mutations elsewhere in the gene. Figure 1 shows the location of *BRCA2* mutations identified by this and other studies of male breast cancer. This illustrates that there appears to be no clustering of mutations that would indicate a genotype–phenotype correlation analogous to that observed for female breast and ovarian cancer.

Our data show that *BRCA2* accounts for only a small proportion of the excess familial risk of female breast cancer associated with MBC. There were 9.37 excess cases of breast cancer in female first-degree relatives (16 observed – 6.63 expected), of which only one was accounted for by *BRCA2*. Thus, allowing for a mutation detection sensitivity of 70%, *BRCA2* accounts for approximately 15% of the excess risk.

Conclusion

The majority of male breast cancer is not accounted for by *BRCA1* and *BRCA2*, and these genes account for only 15% of the excess familial risk of breast cancer in female first-degree relatives. This suggests that other genes that confer an increased risk for both female and male breast cancer have yet to be found.

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