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Risk of breast cancer recurrence and contralateral breast cancer in relation to *BRCA1* and *BRCA2* mutation status following breast-conserving surgery and radiotherapy

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Abstract

BRCA1 and *BRCA2* germline mutations are associated with a strong risk of breast cancer, which may preclude breast-conserving treatment in carriers. This study examined whether mutation status influenced the rate of breast cancer recurrence following breast-conserving treatment. *BRCA1* and *BRCA2* genes were screened for germline mutations in 131 patients with a family history of breast and/or ovarian cancer, who had been treated with breast-conserving surgery and radiotherapy. The 131 patients with familial history were matched to 261 patients without, according to age at diagnosis and year of treatment. The follow-up of controls was at least equal to the time-interval between diagnosis and genetic testing in familial cases. Matched cohorts were compared according to rates of breast cancer recurrence as first event and contralateral breast cancer using log-rank tests. *BRCA1/2* mutations were found in 20.6% patients with a family history. Nineteen patients had a *BRCA1* mutation and 8 had a *BRCA2* mutation. Breast cancers in mutation carriers were more often grade III ($p < 10\text{-}4$) and oestrogen receptor negative ($p = 0.005$) than tumours in both non-carriers and controls. Median follow-up for all 392 patients was 8.75 years. No significant differences in breast cancer recurrence as first event were seen between *BRCA1/2* tumours and controls ($p = 0.47$), carriers and non-carriers with a family history ($p = 0.96$), or non-carriers and controls ($p = 0.10$). On multivariate analysis, age was the most important factor significantly predicting for breast cancer recurrence. The rate of contralateral breast cancer was significantly increased in all patients with a family history: *BRCA1/2* carriers *versus* controls ($p = 0.0003$), non-carriers *versus* controls ($p = 0.0034$) and carriers *versus* non-carriers ($p = 0.02$). At a 9-year median follow-up, the rate of ipsilateral breast cancer recurrence was not higher in *BRCA1* and *BRCA2* mutation carriers than in non-carriers with a family history or sporadic cases. These results support the hypothesis that breast tumours in *BRCA* carriers are more sensitive to radiation. Therefore, breast-conserving treatment can be offered to these patients. However, longer follow-up is needed to ensure that the rate of new primary cancer in the treated breast does not increase in the long-term.

Keywords: *BRCA1/2* mutations; Breast-conserving treatment; Radiotherapy; Recurrence

1. Introduction

Breast conservative surgery and radiation therapy is a standard treatment for early stage breast cancer.

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Numerous randomised trials have proven the equivalence in survival between breast-conserving surgery with radiotherapy and mastectomy in early stage breast cancer [1–8].

BRCA1 and *BRCA2* mutations are found in approximately 5% of all breast cancers, and in up to 20–25% in the case of a family history of breast and/or ovarian cancer [9]. It has been shown previously that *BRCA1* mutation carriers develop tumours with higher grade and proliferation index, and lower oestrogen receptor levels than those who do not have a mutation [10,11]. On the other hand, *BRCA2* mutation carriers present tumours with pathological features similar to sporadic cases [10–16]. It has also been shown that *BRCA1* mutation carriers tend to have a worse outcome [13].

BRCA1 and BRCA2 proteins are involved in DNA repair in response to ionising radiation, through various mechanisms that are not yet fully understood, such as DNA double-strand break repair, apoptosis and cell cycle checkpoint control [17,18]. The safety of breast conservation with radiotherapy in *BRCA* mutation carriers is controversial, because of the potential of ionising radiation to induce new primaries in the treated breast.

The issue of breast cancer recurrence after breast-conserving treatment in *BRCA1* and *BRCA2* mutation carriers was addressed in a small number of studies, using different methodologies to compare breast cancer outcome in carriers and in patients with sporadic cancers, with several biases related to the retrospective nature of these studies [19–25]. A matched cohorts study was conducted at the Institut Curie to assess the rate of recurrences and contralateral breast cancers in *BRCA1/2* mutation carriers compared with sporadic control cases.

2. Patients and methods

2.1. Patients

A cohort of women with small breast cancers treated at the Institut Curie with breast-conserving surgery and radiotherapy from 1981 to 2000, were analysed retrospectively. These patients were invited to attend the family cancer clinic of the institute between 1990 and 2001 if they presented a familial history of breast or ovarian cancer. Selection criteria and the procedure used for molecular testing, as well as the method of obtaining information such as family cancers, age at cancer diagnosis of relatives, and age at death or current age have been reported previously [26,27].

Molecular testing was proposed to women who presented with one of the following family criteria: (a) two first-degree relatives affected with cancer, with at least either one with invasive breast cancer before 41 years or one with ovarian cancer at any age; and (b) at least three first- or second-degree relatives from the same lineage affected with invasive breast or ovarian cancer at any age. The index case was one of the affected family members.

The probability of being carrier of a breast cancer predisposing allele mutation was estimated by taking into account the segregation parameters of Claus modified by Easton and by using the MLINK program [28,29].

After informing the patients about the aims and the limits of breast cancer genetic testing, a blood sample was collected with their written consent.

A total of 131 patients (with 136 breast cancers) was tested. All of them underwent conservative surgery and radiotherapy in our hospital. They were matched to 261 control breast cancer patients (with 271 tumours) without family history (sporadic cases), randomly taken from a prospectively registered population of 9179 patients in the Institut Curie's breast cancer database [30]. All of them had been treated conservatively from 1981 to 2000 at the Institut Curie. Matching was performed on an individual basis: for each case two controls were randomly selected. Matching factors included the age at diagnosis, year of treatment, and period of follow-up between cases and controls. *BRCA* status was unknown in all patients but one at the time of diagnosis and treatment. Clinical, pathological and outcome data were recorded. One control tumour has been excluded from the group because it did not conform with selection criteria.

2.2. Patient treatments

All patients underwent breast-conserving treatment: wide surgical excision of the primary tumour and axillary lymph node dissection in most cases followed by breast irradiation and regional node irradiation when nodes were involved. The total dose delivered to the whole breast was 50–55 Gy in 25–27 fractions and to regional lymph node areas 45 Gy in 23 fractions. If indicated, a boost was delivered to the tumour site.

2.3. BRCA1 and BRCA2 genetic testing

Screening for *BRCA1*/*BRCA2* point mutations or small rearrangements was performed through analysis of genomic DNA from patients' lymphocytes, using either the denaturing gradient gel electrophoresis method (DGGE) or DNA high-performance liquid chromatography (DHPLC) according to the conditions described previously [26,27]. In brief, *BRCA1* and *BRCA2* screening was performed on the 22 and 27 coding exons and their flanking intron–exon junctions. Genes were investigated by use of a total of 85 different amplifications.

Polymerase chain reaction (PCR) products showing an electrophoretic or an elution variant pattern were purified and sequenced in both directions using the

Rhodamine or BigDye Terminator Cycle Sequencing V1.1 Ready Reaction kit (ABI). Electrophoresis was performed with an ABI 377 DNA segment or ABI PRISM 3100 Genetic Analyser.

Only *BRCA* mutations leading to premature termination codon–putative truncated protein were taken into account as causal mutations.

2.4. Statistical analysis

χ^2 or Fisher's exact test for qualitative variables and ANOVA analysis (mean's comparisons) or Kruskal–Wallis test (median's comparisons) for quantitative variables were used to compare patients' and tumours' characteristics.

Survival was determined from the date of diagnosis to the date of death or last follow-up among the 392 patients.

Local recurrence-free interval was defined as the period from the date of diagnosis of breast cancer to the date of first local recurrence among the 407 tumours; time to local recurrence was censored at the time of any other event occurring before the local recurrence (death, lymph node recurrence, distant recurrence, contralateral tumours or second cancer) or at the time of last follow-up.

Contralateral-free interval was defined as the period from the date of diagnosis to the date of contralateral breast cancer whenever it occurred among the 392 patients. Five patients had a bilateral breast cancer at diagnosis. One of these tumours was considered as a contralateral occurring at the time of diagnosis (time to event equals zero).

Kaplan–Meier estimates were calculated to assess overall survival, breast local recurrence-free and contralateral-free rates. Event-free survival times of patients with sporadic disease, familial *BRCA* mutated cases and familial *BRCA* non-mutated cases were compared using log-rank tests [31,32].

The influence of *BRCA* mutation, adjusted for other prognostic factors, was assessed in a multivariate analysis by the Cox proportional hazards model, in a forward stepwise regression procedure [33]. Age, histological nodal status, oestrogen and progesterone receptor status, and Scarff–Bloom–Richardson grading were entered in the model.

Categorical variables were transformed into dummy variables to avoid any assumption concerning the estimation of the relative risks between the various subgroups. Missing values were coded as separate variables when necessary.

The analyses were realised with Splus 2000 software (MathSoft Inc., Seattle, WA, USA).

3. Results

Nineteen patients had a *BRCA1* mutation, and 8 had a *BRCA2* mutation. This represents 20.6% of all patients with a family history (21.3% tumours). Patients' characteristics are presented in Table 1. As shown in this table, the familial and sporadic cohorts were well-matched with regard to age at diagnosis. All cases were treated the same year as their controls, using the same treatment modalities. The follow-up of controls was at least equal to the time-interval between diagnosis and genetic testing in familial cases. The median follow-up for all patients was 105 months (range 31–230): 105 months (35–230) for familial cases and 108 months (31–230) for the control group. As expected, the median probability of being a *BRCA* carrier was significantly higher in *BRCA* carriers than in non-carriers familial cases (Table 1). Two *BRCA1* carriers and 3 *BRCA1/2* non-mutation carriers had synchronous bilateral breast cancers.

Table 2 shows the tumour characteristics according to the *BRCA1*/*BRCA2* mutation status in the three groups. There were 29 tumours in the group of 27

Table 1
Patients' characteristics in relation to *BRCA1/2* mutation status

Characteristics	*BRCA1/2* mutation carriers ($n = 27$)	Non-carriers ($n = 104$)	Sporadic cases ($n = 261$)	p
Age at diagnosis (years)				
Median	43	43.5	43	0.92
Range	(26–60)	(24–78)	(23–79)	
Menopausal status (%)				
Pre-	85.2	70.2	75.9	0.24
Post-	14.8	29.8	24.1	
Time between diagnosis and genetic test (months)				
Mean	39.5	38		
Range	(−17–158)	(2–195)	–	–
Probability of being BRCA-carrier				
Median	90	55		
Range	(73–98)	(6–98)	–	0.002
ND	22	61		

ND, not defined.

Table 2
Tumour characteristics according to *BRCA1/2* status

Characteristic	*BRCA1/2*-mutated tumours ($n = 29$)	Non-mutated tumours ($n = 107$)	Sporadic tumours ($n = 271$)	p
T stage UICC (n (%))				
No palpable tumour	3 (10.3)	17 (15.9)	49 (18.1)	
T1–2	26 (89.7)	85 (79.4)	212 (78.2)	
T3	0	0	1 (0.4)	0.85
T*x*	0	5 (4.7)	9 (3.3)	
Clinical tumour size (mm)				
Median	20	15	20	0.49
Range	(0–35)	(0–35)	(0–70)	
N stage (*n* (%))				
N0	26 (89.7)	89 (84)	243 (70.5)	0.22
N1	3 (10.3)	17 (16)	26 (29.5)	
N*x*				
Pathological nodal status (n (%))				
Negative	21 (72.4)	49 (45.8)	133 (49.1)	
Positive	3 (10.3)	20 (18.7)	41 (15.1)	
No lymph node dissection	5 (17.2)	38 (35.5)	97 (35.8)	0.13
Pathology (n (%))				
Ductal invasive	17 (65.4)	77 (84.6)	216 (82.1)	
Lobular invasive	3 (11.5)	10 (11.0)	16 (6.1)	<10-3[a]
Medullary	3 (11.5)	1 (1.1)	2 (0.8)	
Other	2 (7.7)	1 (1.1)	12 (4.5)	
DCIS	1 (3.9)	2 (2.2)	17 (6.5)	
ND	3	16	8	
Histological grade (n (%))				
I, II	931.1	6776.1	16681.0	
III	1468.9	2123.9	3919.0	<10-4
"Non-gradable" + ND	6	19	66	
Oestrogen receptors (n (%))				
−ve	11 (47.8)	19 (27.5)	33 (20.9)	
+ve	12 (52.2)	50 (72.5)	125 (79.1)	0.018
ND	6	38	113	
Progesterone receptors (n (%))				
−ve	11 (47.8)	15 (21.7)	34 (21.7)	
+ve	12 (52.2)	54 (78.3)	123 (78.3)	
ND	6	38	114	0.02

ND, not defined.
[a] Medullary subtype was more frequent in *BRCA* carriers than in other groups (11.5% *versus* 0.85%, $p = 0.005$).

BRCA1/*BRCA2* mutation carriers. Breast tumours in mutation carriers were more often grade III ($p < 10\text{-}4$) and receptor negative ($p < 0.02$) than tumours in both non-carriers and controls. Medullary subtype was more frequent in mutation carriers than in the two other groups. Moreover, all medullary carcinomas in the genetic cohort were found in patients with *BRCA1* mutations.

Treatment modalities and total dose of radiation therapy are shown in Table 3. There were no significant differences in the treatment modalities between the three groups. The boost to the tumour bed was delivered in 72% of cases of *BRCA1/2* mutation carriers, 61% of cases of familial non-mutated tumours and 66% of sporadic tumours. There was no significant difference between these 3 groups.

There were no significant differences in ipsilateral breast cancer recurrence as first event between *BRCA1/2* tumours and controls ($p = 0.47$), tumours in *BRCA1* and *BRCA2* carriers and non-carriers with a family history ($p = 0.96$), or non-carriers and controls ($p = 0.10$) (Table 4, Fig. 1). The median time-interval before ipsilateral breast cancer recurrences in *BRCA* carriers, non-carriers and controls were as follows: 80 months (35–96), 39 months (22–131) and 46 months (11–150). The site of ipsilateral breast cancer recurrences is shown in Table 4. Two of the 6 patients with medullary carcinomas (Table 2) had an ipsilateral breast cancer recurrence: one recurrence at 79 months in the control group and one at 91 months in the *BRCA1* carriers group.

On multivariate analysis, age as a continuous variable was the most important factor significantly predicting for breast cancer recurrence. The relative risk (RR) of breast cancer recurrence was 1.06 (95% confidence interval (CI) 1.03–1.08, $p < 10\text{-}4$) for each

Table 3
Treatment modalities

	BRCA1/2 mutated tumours ($n = 29$)		Non-mutated tumours ($n = 107$)		Sporadic tumours ($n = 271$)		p
	n	(%)	n	(%)	n	(%)	
Node dissection							
No	5	17.2	38	35.5	97	35.8	
Yes	24	82.7	69	64.5	174	64.2	0.14
Node irradition							
No	15	51.7	40	37.4	108	39.9	
Yes	14	48.3	67	62.6	163	60.1	0.40
Whole breast dose (Gy)							
Median	52		52		52		0.87
Range	(45–62)		(43–62)		(45–66)		
Tumour dose (Gy)							
Median	65		64		65		0.75
Range	(50–75)		(50–78)		(45–82)		
Chemotherapy (%)	38		28		25		0.29
Hormonal therapy (%)	7		13		6		0.045

Table 4
Crude rates of BR as first event

	BRCA1/2-mutated tumours ($n = 29$)	Non-mutated tumours ($n = 107$)	Sporadic tumours ($n = 271$)
Breast cancer recurrences (BR) (n (%))	7 (24)	23 (22)	52 (19)
Site of BR (n (%))			
Same quadrant	6 (85.7)	15 (65.2)	39 (75)
Other quadrant	1 (14.3)	8 (34.8)	13 (25)
Time to BR (months)			
Median	80	39	46
Range	(35–96)	(22–131)	(11–150)

decreasing year of age. Lymph node status was the second significant predictor of ipsilateral breast tumour recurrence (IBTR). The relative risk of IBTR in node-positive tumours in comparison with node negative tumours was 2.11 (95% CI 1.15–3.89, $p = 0.016$). This relative risk in tumours treated with axillary radiotherapy without axillary surgery was 1.99 (95% CI, 1.01–3.9, $p = 0.045$). *BRCA* mutation status, as well as hormonal receptor status and tumour grade, were not significant predictors for local relapse in the analysis.

The risk of contralateral breast cancer was increased significantly in all patients with a family history. The crude rates were 37% (10/27) in the group of *BRCA1/2* mutation carriers, 18.3% (19/104) in the non-mutated group and 7.3% (19/261) in the sporadic group, respectively. Fig. 2(a) shows the rate of contralateral breast cancer in *BRCA1/2* mutation carriers compared with non-carriers and sporadic tumours ($p < 0.0001$), Fig. 2(b) compares mutation carriers with their own sporadic controls ($p = 0.0003$). The median time intervals to contralateral breast cancer were 29.5 months (range 0–140 months), 35 months (range 0–153 months) and 50 months (range 10–146 months), respectively.

4. Discussion

This retrospective case-control study showed that at a 9-year median follow-up, the rate of breast cancer recurrence was not higher in *BRCA1* and *BRCA2* mutation carriers than in non-carriers with family history, or sporadic cases, despite more aggressive features associated with *BRCA1* mutations.

These results are in accordance with the findings of a multi-institute case-control study of Pierce and colleagues [24], where patients in the genetic cohort were matched by age and date of diagnosis with sporadic cases. These authors reported no significant difference in ipsilateral breast cancer recurrences between *BRCA1/2* mutation carriers ($n = 170$) and their controls ($n = 469$) after a median follow-up of 8.3 years. Seynaeve and colleagues [34] conducted a matched cohorts study of hereditary cases matched to 174 sporadic cases according to age and year of diagnosis with a median follow-up of 6 years. After adjustment for age, they reported an increase in IBTR in the hereditary group after 5 years. However, no significant increase was observed in 26 *BRCA1/2* mutation carriers, compared with sporadic cases.

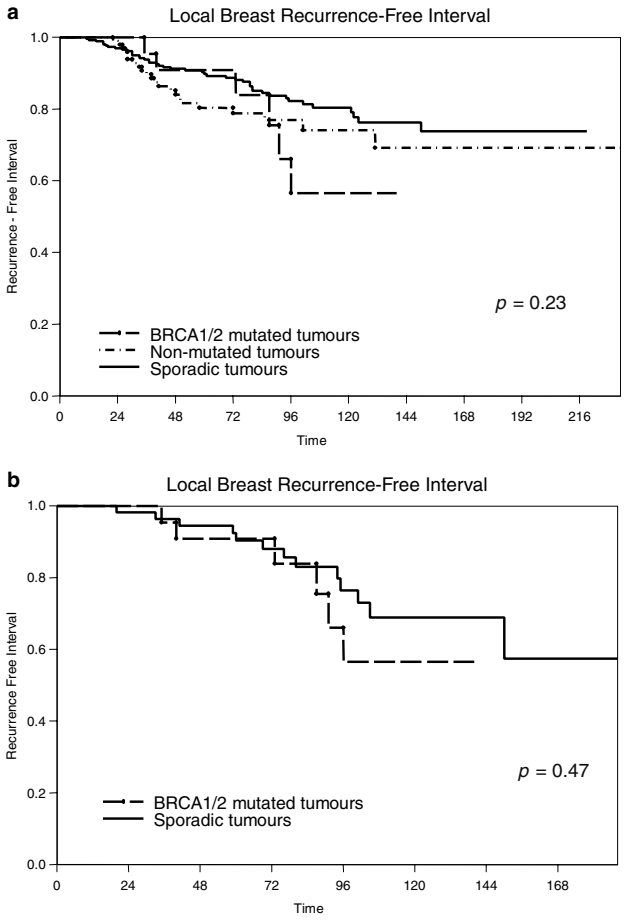

Fig. 1. Local recurrence rate. (a) *BRCA1/2* mutation carriers *versus* non-carriers *versus* controls. (b) Breast cancer recurrence: *BRCA1/2* mutation carriers *versus* their controls.

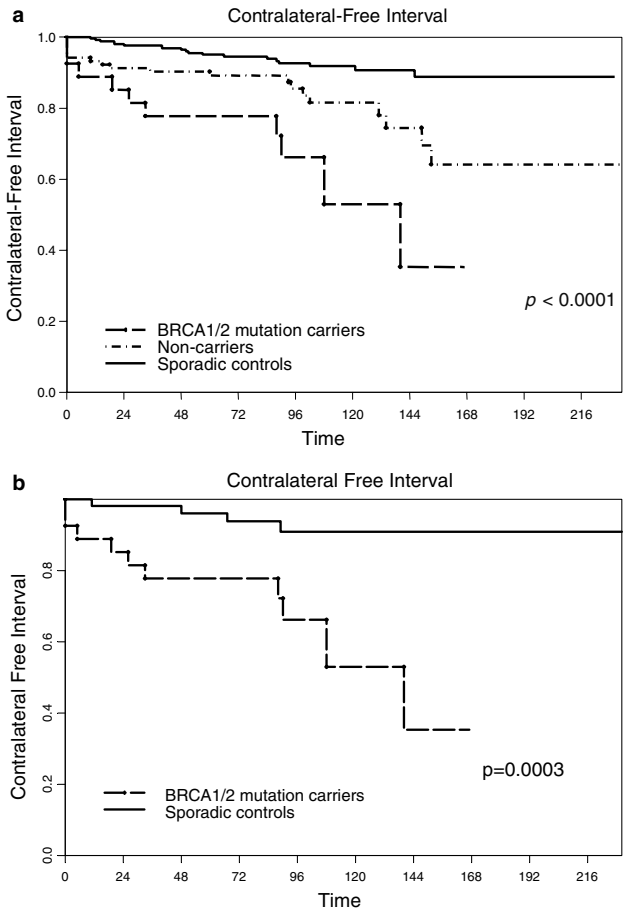

Fig. 2. Contralateral breast cancer. (a) *BRCA1/2* mutations carriers *versus* non-carriers *versus* controls. (b) Contralateral breast cancer: *BRCA1/2* mutation carriers *versus* their controls.

To avoid any possible bias, the follow-up of controls in our study was at least equal to the time-interval between diagnosis and genetic testing in cases. Two non-matched retrospective studies showed contradictory results [15,21]. In the study by Robson and colleagues [15], 56 *BRCA* mutation carriers were compared with 440 sporadic cases. At a median follow-up of 9.7 years, no significant difference in IBTR rate was observed between the two groups. In the study by Haffty and colleagues [21], a subgroup of 127 patients under the age of 42 years was tested for genetic mutations. At a median follow-up of 13 years, the rate of IBTR was significantly higher in *BRCA1/2* mutation carriers ($n = 22$) than in sporadic cases ($n = 105$), suggesting a probable increased rate of second primary cancer after 10 years. Conversely, in our study, age remained a significant predictor of breast cancer recurrence. Our results are consistent with the first conclusions of Delaloge and colleagues [19] who reported that young age, more than *BRCA* status, is a strong predictive factor for local relapse among hereditary breast cancer patients.

The increased risk of breast cancer associated with a *BRCA1/2* mutation is reflected in the risk of contralateral breast cancer. All authors who published their series of *BRCA1* and/or *BRCA2* mutation carriers have found increased incidence of contralateral breast cancer [13,15,20,21,23,24,34]. The present series showed a significantly higher incidence of contralateral breast cancer in *BRCA1/2* carriers (37%) compared with non-carriers (18.3%) and sporadic cases (7.3%) (Fig. 2). Pierce and colleagues [20] reported similar rates of contralateral risk in the collaborative series, with 5-year actuarial estimates of 20% and 2% for the carriers and sporadic groups, respectively ($p < 0.0001$). These results were recently confirmed by the last updated analysis of their series [24]. With 10 years of follow-up, Robson and colleagues [15] reported a 27% risk of contralateral breast cancer for germline carriers compared with 8% for women without mutations ($p = 0.002$). In the series of Haffty and colleagues [21], rates of contralateral breast cancer at 12 years were 42% *versus* 9% for carriers and non-carriers, respectively ($p = 0.001$). Thus, these studies and ours demonstrate that the risk of contralateral breast cancer in *BRCA1*/*BRCA2* mutation carriers is very high and must be taken into consideration when discussing treatment strategies. If a breast-conserving

therapeutic option is chosen, other strategies, such as prophylactic oophorectomy and tamoxifen with close radiological surveillance, should be strongly considered after discussion with individual patients [35–47].

In the present study, the median time to ipsilateral breast cancer recurrence was almost doubled in *BRCA1/2* mutation carriers in comparison with non-carriers and sporadic cases. Due to the small number of patients, this difference is not significant. On the other hand, the median time to contralateral breast cancer was shorter in carriers than in non-carriers or sporadic cases. This fact also supports that radiotherapy is an efficient treatment in reducing the rate of early in-breast cancer recurrences in mutation carriers.

It was shown that tumours in *BRCA1*/*BRCA2* carriers are more sensitive to ionising radiation [48,49]. A probable mechanism might be related to the loss of *bcl2* expression in tumours of *BRCA1* mutation carriers, thus increasing apoptosis in response to treatment [50]. A gene-expression profile study suggests that *BRCA* mutation-associated tumours display increased expression of genes associated with inducing apoptosis, and decreased expression of genes associated with suppressing apoptosis [50]. Finally, the BRCA1 and BRCA2 proteins are thought to be strongly involved in the repair of DNA double-strand breaks induced by ionising radiation, such as those involving ATM [51], *rad51* [52] and *CHK2* [17]. It has been shown that knockout murine embryonic homozygotes *BRCA1* cells were extremely radiosensitive [53,54]. Brodie and colleagues [55] showed that breast cancer cell lines obtained from *BRCA1* tumours grown in mammary conditional knockout mice, were more sensitive to doxorubicin and irradiation than other breast cancer cell lines derived from non-*BRCA1* mutated mice. All these studies support the hypothesis that the mutation of *BRCA* genes could impair the repair capacity of breast cancer in response to radiation therapy.

In conclusion, our study demonstrates that, with a median 9-year follow-up after breast cancer treatment, the rate of breast cancer recurrence was not higher in *BRCA1* and *BRCA2* mutation carriers than in either non-carriers or in patients without family history, despite more aggressive tumour features and a higher risk of contralateral breast cancer. Risk reduction strategies are needed for the contralateral breast. Tumours in *BRCA* carriers may be more sensitive to radiation; therefore, *BRCA* mutation carriers can be offered breast-conserving treatments. However, longer follow-up is needed to ensure that the rate of new breast cancer in the treated breast does not increase in the long-term.

Conflict of interest statement

None declared.

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References

1. Fisher B, Anderson S, Bryant J, *et al*. Twenty-year follow-up of a randomised trial comparing total mastectomy, lumpectomy and lumpectomy plus irradiation for the treatment of invasive breast cancer. *N Engl J Med* 2002, **347**, 1233–1241.
2. Clark R, McCullock P, Levine M, *et al*. Randomised clinical trial to assess the effectiveness of breast irradiation following lumpectomy and axillary dissection for node-negative breast cancer. *J Natl Cancer Inst* 1992, **84**, 683–689.
3. VanDongen JA, Voogd AC, Fentiman IS, *et al*. Long-term results of a randomized trial comparing breast-conserving therapy with mastectomy: EORTC 10801 trial. *J Natl Cancer Inst* 2000, **92**, 1143–1150.
4. Veronesi U, Cascinelli N, Mariani L, *et al*. Twenty-year follow-up of a randomised study comparing breast-conserving surgery with radical mastectomy for early breast cancer. *N Engl J Med* 2002, **347**, 1227–1232.
5. Jacobson JA, Danforth DN, Cowan KH, *et al*. Ten-year results of a comparison of conservation with mastectomy in the treatment of stage I and II breast cancer. *N Engl J Med* 1995, **332**, 907–911.
6. Blichert-Toft M, Roce C, Anderson JA, *et al*. Danish randomised trial comparing breast conservation therapy with mastectomy: six years of life table analysis. Danish Breast Cancer Cooperative group. *J Natl Cancer Inst Monograms* 1992, **11**, 19–25.
7. Arriagada R, Le MG, Guinebretière JM, *et al*. Late local recurrences in a randomised trial comparing conservative treatment with total mastectomy in early breast cancer patients. *Ann Oncol* 2003, **14**, 1617–1622.
8. Early Breast Cancer Trialists' Collaborative Group. Favourable and unfavourable effects on long-term survival of radiotherapy for early breast cancer: an overview of the randomised trials. *Lancet* 2000, **355**, 1757–1770.
9. Robson ME. Clinical considerations in the management of individuals at risk for hereditary breast and ovarian cancer. *Cancer Control* 2002, **9**, 457–465.
10. Adem C, Reynolds C, Soderberg CL, *et al*. Pathologic characteristics of breast parenchyma in patients with hereditary breast carcinoma, including BRCA1 and BRCA2 mutation carriers. *Cancer* 2003, **97**, 1–11.
11. Chappuis PO, Nethercot V, Foules WD, *et al*. Clinico-pathological characteristics of BRCA1 and BRCA2-related breast cancer. *Semin Surg Oncol* 2000, **18**, 287–295.
12. Breast Cancer Linkage Consortium. Pathology of familial breast cancer: differences between breast cancers in carriers of BRCA1 or BRCA2 mutations and sporadic cases. *Lancet* 1997, **349**, 1505–1510.
13. Stoppa-Lyonnet D, Ansquer Y, Dreyfus H, *et al*. Familial invasive breast cancers: worse outcome related to BRCA1 mutations. *J Clin Oncol* 2000, **18**, 4053–4059.
14. Lakhani SR, Jacquemier J, Sloane JP, *et al*. Multifactorial analysis of differences between sporadic breast cancers and cancers involving BRCA1 and BRCA2 mutations. *J Natl Cancer Inst* 1998, **90**, 1138–1145.
15. Robson M, Chappuis PO, Satagopan J, *et al*. A combined analysis of outcome following breast cancer: differences in survival based on BRCA1/BRCA2 mutation status and admin-

istration of adjuvant treatment. *Breast Cancer Res* 2004, **6**, R8–R17.
16. Foulkes WD, Chappuis PO, Wong N, *et al*. Primary node negative breast cancer in BRCA1 mutation carriers has a poor outcome. *Ann Oncol* 2000, **11**, 307–313.
17. Venkitaraman A. Cancer susceptibility and the functions of BRCA1 and BRCA2. *Cell* 2002, **108**, 171–182.
18. Powel SN, Kachnic LA. Roles of BRCA1 and BRCA2 in homologous recombination, DNA replication fidelity and the cellular response to ionising radiation. *Oncogene* 2003, **22**, 5784–5791.
19. Delaloge S, Kloos I, Ariane D, *et al*. Young age is the major predictor of local relapse among conservatively treated BRCA1-, BRCA2-, or non BRCA-linked hereditary breast cancer. *Proc ASCO* 2003, **22**(41), 11. (abstract).
20. Pierce LJ, Strawderman M, Narod SA, *et al*. Effect of radiotherapy after breast-conserving treatment in women with breast cancer and germline BRCA1/2 mutations. *J Clin Oncol* 2000, **18**, 3360–3369.
21. Haffty BG, Harrold E, Khan AJ, *et al*. Outcome of conservatively managed early-onset breast cancer by BRCA1/BRCA2 status. *Lancet* 2002, **359**, 1471–1477.
22. Bremer M, Doerk T, Sohn C, Karstens JH. Local relapse after postoperative radiotherapy in patients with bilateral breast cancer by BRCA1/2 status. *Proc ASCO* 2003, **22**(42), 11. (abstract).
23. Turner BC, Harrold E, Matloff E, *et al*. BRCA1/BRCA2 germline mutations in locally recurrent breast cancer patients after lumpectomy and radiation therapy: implications for breast-conserving management in patients with BRCA1/BRCA2 mutations. *J Clin Oncol* 1999, **17**, 3017–3024.
24. Pierce L, Levin A, Rebbeck T, *et al*. Ten-year outcome of breast-conserving surgery and radiotherapy in women with breast cancer and germline BRCA1/2 mutations: results from an international collaboration. *Breast Cancer Res Treat* 2003, **82**(#5), S7. (abstract).
25. Verhoog LC, Brekelmans CTM, Seynaeve C, *et al*. Survival and tumour characteristics of breast cancer patients with germline mutations of BRCA1. *Lancet* 1998, **351**, 316–321.
26. Wagner T, Stoppa-Lyonnet D, Fleischmann E, *et al*. Denaturing high-performance liquid chromatography detects reliably BRCA1 and BRCA2 mutations. *Genomics* 1999, **62**, 369–376.
27. Stoppa-Lyonnet D, Laurent-Puig P, Essioux L, *et al*. BRCA1 sequence variations in 160 individuals referred to a breast/ovarian family cancer clinic. Institut Curie Breast Cancer Group. *Am J Hum Genet* 1997, **60**, 1021–1030.
28. Claus EB, Risch N, Thompson WD. Genetic analysis of breast cancer in the cancer and steroid hormone study. *Am J Hum Genet* 1991, **48**(2), 232–242.
29. Easton DF, Bishop DT, Ford D, *et al*. Genetic linkage analysis in familial breast and ovarian cancer: results from 214 families. The Breast Cancer Linkage Consortium. *Am J Hum Genet* 1993, **52**(4), 678–701.
30. Salmon RJ, Asselain B, Le Gal M, *et al*. Twelve years experience of breast cancer at the Institut Curie: improvement of survival and value of screening mammographics. *The Breast* 1997, **6**, 202–205.
31. Kaplan EL, Meier P. Non-parametric estimation from incomplete observations. *J Am Stat Assoc* 1958, **53**, 457–481.
32. Mantel N. Evaluation of survival data and two new rank order statistics arising in its consideration. *Cancer Chemother Rep* 1966, **50**, 163–170.
33. Cox DR. Regression models and life tables. *J R Stat Soc* 1972, **34**, 187–220.
34. Seynaeve C, Verhoog LC, Van De Bosch LM, *et al*. Ipsilateral breast tumour recurrence in hereditary breast cancer following breast-conserving therapy. *Eur J Cancer* 2004, **40**, 1150–1158.
35. Pierce L. Radiotherapy for breast cancer in BRCA1/BRCA2 carriers: clinical issues and management dilemmas. *Semin Radiat Oncol* 2002, **12**, 352–361.
36. Meijers-Heijboer EJ, Verhoog LC, Brekelmans CTM, *et al*. Presymptomatic DNA testing and prophylactic surgery in families with a BRCA1 or BRCA2 mutation. *Lancet* 2000, **355**, 2015–2020.
37. Bresser PJC, Seynaeve C, Van Gool AR, *et al*. Long-term satisfaction with bilateral prophylactic mastectomy and immediate breast reconstruction in genetically predisposed women. *Breast Cancer Res Treat* 2003, **82**, S16., #32 (abstract).
38. Klijn JGM, Janin N, Cortes-Funes, *et al*. Should prophylactic surgery be used in women at high risk of breast cancer? *Eur J Cancer* 1997, **33**, 2149–2159.
39. Shrag D, Kuntz KM, Garber JE, *et al*. Decision analysis-effects of prophylactic mastectomy and oophorectomy on life expectancy among women with BRCA1 or BRCA2 mutations. *N Engl J Med* 1997, **336**, 1465–1471.
40. Blamey RW, Mackay J, Macmillan D. EUSOMA guidelines on the management of familial breast cancer risk. *Eur J Cancer* 2004, **2**, 155. (abstract).
41. Klijn JGM, Erasmus MC. Prophylactic mastectomy in high risk women. *Eur J Cancer* 2004, **2**, 156. (abstract).
42. Narod SA, Brunet JS, Ghadirian P, *et al*. Tamoxifen and risk of contralateral breast cancer in BRCA1 and BRCA2 mutation carriers: a case-control study. *Lancet* 2000, **356**, 1876–1881.
43. Early Breast Cancer Trialists' Collaborative Group. Tamoxifen for early breast cancer: an overview of the randomised trials. *Lancet* 1998, **351**, 1451–1467.
44. Fisher B, Costantino JP, Wickerham DL, *et al*. Tamoxifen for prevention of breast cancer: Report of the National Surgical Adjuvant Breast and Bowel Project P-1 study. *J Natl Cancer Inst* 1998, **90**, 1371–1388.
45. Kauff ND, Satagopan JM, Robson ME, *et al*. Risk reducing salpingo-oophorectomy in women with a BRCA1 or BRCA2 mutation. *N Engl J Med* 2002, **346**, 1609–1615.
46. Rebbeck TR, Lynch HT, Neuhausen SL, *et al*. Prophylactic oophorectomy in carriers of BRCA1 or BRCA2 mutations. *N Engl J Med* 2002, **346**, 1616–1622.
47. Haber D. Prophylactic oophorectomy to reduce the risk of ovarian and breast cancer in carriers of BRCA mutations. *N Engl J Med* 2002, **346**, 1660–1662.
48. Baeyens A, Thierens H, Claes K, *et al*. Chromosomal radiosensitivity in breast cancer patients with a known or putative genetic predisposition. *Br J Cancer* 2002, **87**, 1379–1385.
49. Fourquet A, Stoppa-Lyonnet D, Sigal-Zafrani B, et al. Familial invasive breast cancer: clinical response to induction chemotherapy or radiotherapy related to BRCA1 and BRCA2 mutations, in press.
50. Freneaux P, Stoppa-Lyonnet D, Mouret E, *et al*. Low expression of bcl-2 in Brca1-associated breast cancers. *Br J Cancer* 2000, **83**, 1318–1322.
51. Hedenfalk I, Duggan D, Chen Y, *et al*. Gene-expression profiles in hereditary breast cancer. *N Engl J Med* 2001, **344**, 539–548.
52. Cortez D, Wang Y, Qin J, *et al*. Requirement of ATM-dependent phosphorylation of Brca1 in the DNA damage response to double-strand breaks. *Science* 1999, **286**, 1162–1166.
53. Scully R, Chen J, Ochs RL, *et al*. Dynamic changes of BRCA1 subnuclear location and phosphorylation state are initiated by DNA damage. *Cell* 1997, **90**, 425–435.
54. Xu X, Weaver Z, Linke SP, *et al*. Centrosome amplification and a defective G2-M cell cycle checkpoint induce genetic instability in BRCA1 exon 11 isoform-deficient cells. *Mol Cell* 1999, **3**, 389–395.
55. Brodie SG, Xu X, Qiao W, *et al*. Multiple genetic changes are associated with mammary tumorigenesis in BRCA1 conditional knockout mice. *Oncogene* 2000, **20**, 7514–7523.