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# Founder BRCA1 and BRCA2 Mutations in Ashkenazi Jews in Israel: Frequency and Differential Penetrance in Ovarian Cancer and in Breast-Ovarian Cancer Families

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## Summary

**Germ-line BRCA1 and BRCA2 mutations account for most of familial breast-ovarian cancer. In Ashkenazi Jews, there is a high population frequency (~2%) of three founder mutations: BRCA1 185delAG, BRCA1 5382insC, and BRCA2 6174delT. This study examined the frequency of these mutations in a series of Ashkenazi women with ovarian cancer unselected for family history, compared with the frequency of these mutations in families ascertained on the basis of family history of at least two affected women. Penetrance was compared, both according to the method of family ascertainment (i.e., on the basis of an unselected ovarian cancer proband vs. on the basis of family history) and for the BRCA1 founder mutations compared with the BRCA2 6174delT mutation. There was a high frequency (10/22; [45%]) of germ-line mutations in Ashkenazi women with ovarian cancer, even in those with minimal or no family history (7/18 [39%]). In high-risk Ashkenazi families, a founder mutation was found in 59% (25/42). Families with any case of ovarian cancer were significantly more likely to segregate a founder mutation than were families with site-specific breast cancer. Penetrance was higher in families ascertained on the basis of family history than in families ascertained on the basis of an unselected proband, but this difference was not significant. Penetrance of BRCA1 185delAG and BRCA1 5382insC was significantly higher than penetrance of BRCA2 6174delT (hazard ratio 2.1 [95% CI 1.2–3.8]; two-tailed $P = .01$). Thus, the high rate of germ-line BRCA1/BRCA2 mutations in Ashkenazi women and families with ovarian cancer is coupled with penetrance that is lower than previously estimated. This has been shown specifically for the BRCA2 6174delT mutation, but, because of ascertainment bias, it also may be true for BRCA1 mutations.**

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## Introduction

Germ-line mutations in the BRCA1 and BRCA2 genes are thought to account for most of familial breast-ovarian and breast cancer (Easton et al. 1993; Wooster et al. 1994). Since 5%–10% of all breast and ovarian cancer are considered to be due to an inherited predisposition, carrier frequencies for BRCA1 and BRCA2 in the population should not be uncommon, and they have been estimated to be on the order of 1/300 (Claus et al. 1991). Identification of these genes has allowed for direct mutation detection, but large-scale screening has been hampered both by the large size of BRCA1 and BRCA2, which have 5.592-kb and 10.254-kb coding sequences, respectively (Miki et al. 1994; Wooster et al. 1995; Tavtigian et al. 1996), and by the occurrence of multiple private mutations (Couch et al. 1996). However, within defined ethnic groups, specific mutations are likely to recur. For example, in Iceland, the BRCA2 999del5 founder mutation occurs in most breast cancer families and in 0.4% of Icelandic controls (Johannesdottir et al. 1996). In Ashkenazi Jews, (Jews of eastern European ancestry), three founder mutations have been described in breast cancer families: 185delAG and 5382insC in BRCA1 and 6174delT in BRCA2 (Simard et al. 1994; Friedman et al. 1995; Neuhausen et al. 1996). One of these mutations was found in 31% of 80 Ashkenazi women with early-onset (<42 years of age) breast cancer and in 41% of 27 Ashkenazi women with a positive family history who were diagnosed with breast cancer at 42–50 years of age (Neuhausen et al. 1996; Oddoux et al. 1996; Offit et al. 1996). In a series of 220 North American Ashkenazi breast cancer families, one of the founder mutations accounted for 45% of all families and for a significantly higher percentage (73%) of families with a history of ovarian cancer (Tonin et al. 1996). The combined population frequency of these mutations in a number of large series of young Ashkenazi controls approaches 2.5%: BRCA1 185delAG, 1.0%; BRCA1 5382insC, 0.1%; and BRCA2 6174delT, 1.4% (Streuwing et al. 1995; Oddoux et al. 1996; Roa et al. 1996). Current estimates of penetrance of these muta-

tions are based on the high-risk pedigrees used in the original linkage studies, where lifetime penetrance of BRCA1 mutations was found to be >80% for breast cancer and approached 50% for ovarian cancer (Easton et al. 1993; Ford et al. 1994; Friedman et al. 1995). Penetrance of BRCA2 mutations was estimated to be similar for breast cancer and lower for ovarian cancer (Wooster et al. 1994). Families used in BRCA1 and BRCA2 linkage studies were selected for high penetrance at an early age (e.g., the presence of at least four affected living relatives), so it is unclear whether these penetrance estimates are reasonably applied either to families with fewer affected individuals or to asymptomatic carriers regardless of family history. Indeed, the BRCA2 6174delT mutation has been inferred to have lower penetrance than the BRCA1 185delAG mutation, because, although their frequencies in the Ashkenazi population are similar, BRCA2 6174delT is significantly less common in high-risk Ashkenazi breast-ovarian cancer families. Comparisons of these frequencies have led to the conclusion that BRCA2 6174delT penetrance is 25%–30% that of 185delAG (Oddoux et al. 1996; Roa et al. 1996).

Genetic testing is technically more feasible in the setting of a limited number of mutations in a defined ethnic group. However, before such testing is implemented, either in affected individuals or in the population, it is important to define the penetrance and attributable risk of these mutations for breast and ovarian cancer in the population studied. In this study we examined the frequency of the founder BRCA1/BRCA2 mutations, both in a series of Ashkenazi women with ovarian cancer unselected for family history and in a group of high-risk families with breast and/or ovarian cancer. In order to assess the effect of ascertainment on penetrance, we compared families ascertained on the basis of an unselected ovarian cancer proband to families ascertained on the basis of positive family history. We also compared the penetrance of the BRCA1 and BRCA2 founder mutations.

## Subjects and Methods

### *Subjects*

Ovarian cancer cases are consecutive patients with ovarian cancer diagnosed after January 1994 and seen at the Division of Gynecologic Oncology at Shaare Zedek Medical Center (SZMC) in Jerusalem during September 1995–May 1996. Familial cancer cases are self- and physician-referred families that were seen at the SZMC cancer genetics clinic since its establishment in February 1996 and that fulfilled the following criteria: (1) A minimum of two women affected with breast or ovarian cancer. (2) At least one affected woman available for testing; included are four women with both breast and ovarian cancer referred by the gynecologic oncologist (U.B.).

For both ovarian cancer and familial cancer cases, family pedigrees were drawn as far back and as far laterally as possible. Recorded information included age, either age at death or age at which status was last known, cancer status, cancer type, age at diagnosis, and age at which oophorectomies were performed for reasons other than ovarian cancer. Cancer diagnoses in cases were based on pathology reports. Cancer diagnoses in relatives were based on hospital records, when the latter were available, and on family report, which has moderately high accuracy, especially for breast and ovarian cancer (Novakovic et al. 1996). Women found to be carriers were informed which living relatives were at risk and were offered the opportunity to refer relatives for counseling. At-risk relatives were not approached directly by the clinic staff. All women were given genetic counseling and gave informed consent for testing.

### *Non-Ashkenazi Carriers of the BRCA1 185delAG Mutation*

Two carriers were identified at SZMC: one (SM-1) was an Iranian Jewish woman in our ovarian-cases series, and the second (BH-1) was an Iraqi Jewish woman with bilateral breast cancer diagnosed at age 35 years by the SZMC Institute of Oncology. Two women of Iraqi origin were identified at Rabin Medical Center (by M.S.): one woman (MS-1) diagnosed with unilateral breast cancer at age 29 years, whose mother was diagnosed with breast cancer at age 38 years (Sher et al. 1996), and a second woman (MS-2) diagnosed with both ovarian and breast cancer (at ages 54 and 58 years, respectively), whose family history includes a sister with breast cancer (diagnosed at age 55 years) and three nieces with early-onset breast cancer.

### *Mutation Analysis*

Genomic DNA was extracted from peripheral blood samples by use of standard methods and was amplified by PCR with the following primer sets:

1. BRCA1 185delAG (exon 2)—forward, 5′GAA GTT GTC ATT TTA TAA ACC TTT-3′; and re- verse—5′-TGT CTT TTC TTC CCT AGT ATG T-3′;
2. BRCA1 5382insC (exon 20)—forward, 5′-ATA TGA CGT GTC TGC TCC AC-3′; and reverse, 5′-GGG AAT CCA AAT TAC ACA GC-3′;
3. BRCA2 6174delT (exon 11)—forward, 5′-AAC GAA AAT TAT GGC AGG TTG TTA C-3′; and reverse, 5′-GCT TTC CAC TTG CTG TAC TAA ATC C-3′.

PCR conditions for both BRCA1 mutations were 95°C for 5 min (94°C for 1 min, 58°C for 1 min, and 72°C for 1 min), for 33 cycles, and 72°C for 10 min. PCR conditions for the BRCA2 mutation were identical, except that the annealing was at 55°C. Amplification products were analyzed by conformation-sensitive gel electrophoresis (Ganguly et al. 1993), which separates

heteroduplexes on a mildly denaturing acrylamide gel. PCR products were denatured at 95°C for 5 min, reannealed at 68°C for 30 min, and run on 10% polyacrylamide gel (99:1 acrylamide:1,4-bis[acroyl]piperazine), 10% ethylene glycol, 15% formamide, 0.5 × TTE (44.4 mM Tris, 14.25 mM Taurine, and 0.1 mM EDTA, pH 9.0). Electrophoresis was performed overnight at 200 V in 0.5 × TTE, and gels were stained with ethidium bromide and were photographed (Couch et al. 1996). Each sample was analyzed in duplicate.

### *Genotyping Markers and Haplotype Analysis*

Markers D17S855, D17S1322, and D17S1326 were typed as described elsewhere (Couch et al. 1996), for all four non-Ashkenazi carriers, a noncarrier daughter (SM-2) of SM-1, seven Ashkenazi 185delAG carriers, and a previously characterized Ashkenazi control (data kindly provided by M.-C. King). Haplotypes could be constructed for the mother-daughter pair, and alelle sharing was demonstrated in the other cases.

### *Statistical Methods*

Penetrance analysis in families segregating a founder mutation excluded all probands and included all other female family members. The age used for affected subjects was age at cancer diagnosis, and that used for unaffected subjects was either age at death or age at which status was last known. For five deceased subjects affected with breast cancer, for whom only age at death was known, we assumed a mean survival of 5 years (Harris et al. 1996) and set age at onset at 5 years prior to death. Either carrier status was tested directly or the risk of carrier status was assigned according to pedigree structure. All subjects with ovarian cancer or with breast cancer diagnosed <60 years of age were assumed to be carriers. For subjects with other types of cancer the analysis was done twice: once under the assumption of pedigree-derived risk and once under the assumption that they were carriers. For previous, deceased generations, in which the mutation-bearing lineage could not be determined on the basis of either carrier status or family history, equal probability for each lineage was assumed. Results shown are for an age-corrected model that includes the following corrections: (1) For women with breast cancer diagnosis at <60 years of age who were not obligate or tested carriers, the background probability for breast cancer was adjusted according to the age-specific population risk (per decade) (Easton et al. 1993). (2) For women who remained unaffected at ≥60 years of age and who were not obligate or tested carriers, posterior probability of carrier status is smaller than the pedigree-derived risk. This was adjusted for use of the computed gene-specific penetrance by 60, 70, and 80 years of age in our study population, controlled for ascertainment. In the families used in the original BRCA1 linkage studies, carrier rate was only 2/32 (6%) in older (≥60 years of age) unaffected first-degree relatives of breast/ovarian cancer cases (Easton et al. 1993). Subsequent penetrance analyses in BRCA1-linked pedigrees (Ford et al. 1994) took this figure into account and adjusted for posterior probability of carrier status according to age. Since this adjustment assumes the high penetrance observed in the original families studied, it was not used in this study; rather, the same method was applied with penetrance data derived from our families. Penetrance was analyzed separately for BRCA1 and BRCA2 mutations and for families ascertained as consecutive ovarian cancer cases versus families ascertained by the cancer genetics clinic. We analyzed penetrance for total malignancy, breast and ovarian cancer combined, breast cancer only, and ovarian cancer only. Penetrance analysis was done by Kaplan Meier survival weighted for carrier risk, and penetrance comparisons were performed by Cox regression weighted for carrier risk.

Comparisons of mean ages at onset, family history, and mutation status were done by *t*-test. All statistical analyses were performed with the SPSS for Windows 6.1 software package (Norusis 1994).

**Table 1**

**Age at Onset in Consecutive Ashkenazi Women with Ovarian Cancer**

| Mutation Status (*n*) | Mean Age at Onset (±SD) [Range] |
|---|---|
| Noncarriers (12)[a] | 54.4 (±11.3) [43–80] |
| Carriers (10):[a] | 55.6 (±14.0) [34–79] |
| BRCA1 (7)[b] | 50.1 (±12.0) [34–72] |
| BRCA2 (3)[b] | 68.3 (±10.1) [59–79] |

[a] Nonsignificant difference for comparison of carriers versus noncarriers.

[b] $P = .05$ for comparison of BRCA1 carriers versus BRCA2 carriers.

## Results

### *Frequency of the Founder BRCA1/BRCA2 Mutations in the Study Population*

*1. Ovarian cancer cases.*—Thirty three cases were ascertained; 22 were Ashkenazi Jewish, and 11 were non-Ashkenazi Jewish. One of the three founder BRCA1/BRCA2 mutations was found in 10/22 (45%) Ashkenazi cases and in 1/11 (9%) non-Ashkenazi cases. Mean age at onset was not different in those with identified mutations versus those without identified mutations. However, the mean age at onset in BRCA1 carriers was significantly younger than that in BRCA2 carriers (table 1). The majority of Ashkenazi women with ovarian cancer who had *any* family history of breast or ovarian cancer (table 2) were found to be carriers of a founder mutation. Two of 12 women with no family history also were found to be carriers: the 185delAG carrier has a

## Table 2

**Family History and Founder BRCA1/BRCA2 Mutations in Consecutive Ashkenazi Women with Ovarian Cancer**

| | No. of Cases | | | | |
|---|---|---|---|---|---|
| | | | BRCA1 | | |
| Family-History Status | | Mutation Carriers | 185delAG | 5382insC | BRCA2 6174delT |
| Suggestive of familial breast/ovarian cancer[a] | 4 | 3 (75%) | 2 | 0 | 1 |
| Minimal family history[b] | 6 | 5 (83%) | 2 | 2 | 1 |
| No family history[c] | 12 | 2 (17%) | 1 | 0 | 1 |
| Total | 22 | 10 (45%) | 5 | 2 | 3 |

[a] Three or more breast or ovarian cancer cases (including proband) in the same lineage, in a pattern consistent with autosomal dominant inheritance.

[b] Two breast or ovarian cancer cases (including proband), either first- or second-degree relatives.

[c] No relatives affected with breast or ovarian cancer, up to (and including) third-degree relatives.

truncated pedigree (five at-risk relatives >40 years of age in both lineages), but the 6174delT carrier has eight at-risk relatives in her generation and the preceding generation. Founder mutations were not found in either four women with a highly suggestive family history or in one of six cases with minimal family history. Among 11 non-Ashkenazi Jewish cases tested, one woman of Iranian descent was found to be a 185delAG carrier. She has no family history of breast or ovarian cancer, and disease onset was at age 79 years.

*2. Familial cancer cases.*—Of 44 families ascertained, 42 were Ashkenazi Jewish, and 2 were non-Ashkenazi Jewish (one each of Iraqi and Bokharan [southern Russian] origin). Although age was not a selection criterion, all breast cancer probands were diagnosed at <60 years of age. One of the founder mutations was found in 25 (59%) of 42 Ashkenazi families and in neither of the non-Ashkenazi families. The proportion of cancer accounted for by one of the founder mutations varied according to family history (table 3). Families with at least one case of ovarian cancer were significantly more likely to segregate one of the founder mutations than were families with only breast cancer ($P = .01$). One of the founder mutations was identified in all six families in which a subject had both breast and ovarian cancer. In subjects with both breast and ovarian cancer, mean age at onset was 45.5 years for breast cancer and 58.2 years for ovarian cancer, with a median interval of 16 years between diagnoses (range −2 to 22).

None of the founder mutations were identified in the two families who were non-Ashkenazi Jewish or in 10/11 non-Ashkenazi ovarian cancer cases. The BRCA1 185delAG mutation was identified in one ovarian cancer case of Iranian Jewish origin. Analysis of three intragenic and flanking markers spanning ~190 kb in four Iranian/Iraqi 185delAG carriers revealed the same haplotype that previous reports have described in Ashkenazi Jews (Simard et al. 1994; Friedman et al. 1995) (table 4). These results suggest a common origin for the 185delAG mutation in Ashkenazi and Iraqi/Iranian Jews.

## Table 3

**Frequency of Founder BRCA1/BRCA2 Mutations in Ashkenazi Breast and/or Ovarian Cancer Families**

| | No. of Families with | | | | |
|---|---|---|---|---|---|
| | | | BRCA1 | | |
| Family History | No Mutation[a] | Any Founder Mutation | 185delAG | 5382insC | BRCA2 6174delT |
| Two breast cancers | 7 | 1 (12%) | 0 | 0 | 1 |
| Three or more breast cancers | 2 | 2 (50%) | 0 | 2 | 0 |
| Breast cancer and one or more ovarian cancers | 7 | 12 (63%) | 4 | 3 | 5 |
| Ovarian cancer only | 1 | 4 (80%) | 2 | 1 | 1 |
| One case with both breast and ovarian cancer | 0 | 6 (100%) | 2 | 2 | 2 |
| Total | 17 | 25 (59%) | 8 | 8 | 9 |

[a] Families were tested only for the three founder mutations.

**Table 4**

**Penetrance of Breast and Ovarian Cancer Combined by Method of Family Ascertainment**

| Mutation and Method of Ascertainment | Penetrance (Standard Error) (%) | | Hazard Ratio[a] |
|---|---|---|---|
| | At Age 70 Years | At Age 80 Years | |
| BRCA1: | | | |
| Ovarian series | 51 (15) | ... | 1.6 (NS) |
| Cancer genetics clinic | 57 (6) | 64 (7) | |
| BRCA2: | | | |
| Ovarian series | 43 (13) | ... | 1.3 (NS) |
| Cancer genetics clinic | 49 (13) | ... | |

[a] For penetrance of clinical ascertainment compared with ascertainment on the basis of ovarian cancer–cases series. For BRCA1 and BRCA2 combined, two-tailed $P = .2$. NS = not significant.

*Penetrance of the Founder Mutations*

A total of 301 subjects were included in the analysis (35 probands with founder mutations and 21 at-risk women tested and found not to have a founder mutation were excluded). For BRCA1, there were 208 subjects (64 deceased), including 41 breast cancer cases, 15 ovarian cancer cases, and 14 other cancer events. When probands were excluded, 14 women were found to be carriers on the basis of molecular testing, and 22 were obligate carriers. For BRCA2, there were 93 subjects (41 deceased), including 13 breast cancer cases, 6 ovarian cancer cases , and 11 other cancer events. When probands were excluded, 7 women were found to be carriers on the basis of molecular testing, and 11 were obligate carriers. Subjects who were not tested directly or who were not obligate carriers were assigned a statistical risk for carrier status (see Subjects and Methods). Results of analysis with the age-corrected model were only slightly different from those of analysis without the age-corrected model; therefore, only the results of the age-corrected model are shown (table 4 and fig. 1). These results are represented as curves of breast and ovarian cancer-free survival in carriers (fig. 1), which correspond to the proportion of carriers remaining nonpenetrant.

*Penetrance and Method of Ascertainment*

Penetrance of breast and ovarian cancer combined was not significantly lower in families ascertained on the basis of consecutive ovarian cancer cases than in families ascertained by the cancer genetics clinic (table 4 and fig. 1). This was true for both BRCA1 mutations and for the BRCA2 mutation. The proportional hazard ratio for clinic ascertainment versus ovarian-cases ascertainment was 1.6 and 1.3 for BRCA1 and BRCA2, respectively, but these differences were not significant. Similar results were obtained for penetrance of all malignancies and for both estimates of risk in subjects with non–breast/ovarian cancer (see Subjects and Methods).

*Reduced Penetrance of the BRCA2 6174delT Mutation (table 4 and fig. 1)*

After method of ascertainment was controlled for, both penetrance of breast and ovarian cancer combined and penetrance of breast cancer alone were significantly higher for the 185delAG and 5382insC BRCA1 mutations than for the BRCA2 6174delT mutation: the hazard ratio for breast and ovarian cancer combined was 2.1 (95% CI 1.2–3.8; two-tailed $P = .01$), and that for breast cancer alone was 2.1 (95% CI 1.03–4.1; two-tailed $P = .04$). A similar difference in penetrance was observed for ovarian cancer alone (hazard ratio 2.2 [95% CI 0.7–6.4]), but it did not reach statistical significance. Because, compared with 5′ mutations, mutations in the 3′ end of BRCA1 have been reported to be associated with increased ovarian cancer risk (Gayther et al. 1995), we compared ovarian cancer penetrance for 185delAG-mutation (5′) carriers versus 5382insC-mutation carriers (3′). Carriers of 185delAG had a small, nonsignificantly higher risk for ovarian cancer than did carriers of 5382insC, an effect opposite to that reported elsewhere (Gayther et al. 1995).

## Discussion

*Frequency of Founder BRCA1/BRCA2 Mutations in Ovarian Cancer and Familial Breast-Ovarian Cancer in Ashkenazi Women*

We found a surprisingly high frequency (45%) of germ-line BRCA1/BRCA2 mutations in a series of women with ovarian cancer who were unselected for family history. In general, only 5%–10% of ovarian cancer is expected to result from germ-line mutations. In our series 4/22 (18%) cases had a suggestive family history, and 3 of these segregated one of the founder mutations (table 2). Most of the cases found to be carriers would not have been considered to be familial (i.e., they had none or one family member with breast or ovarian cancer). The carrier rate for all nonfamilial cases

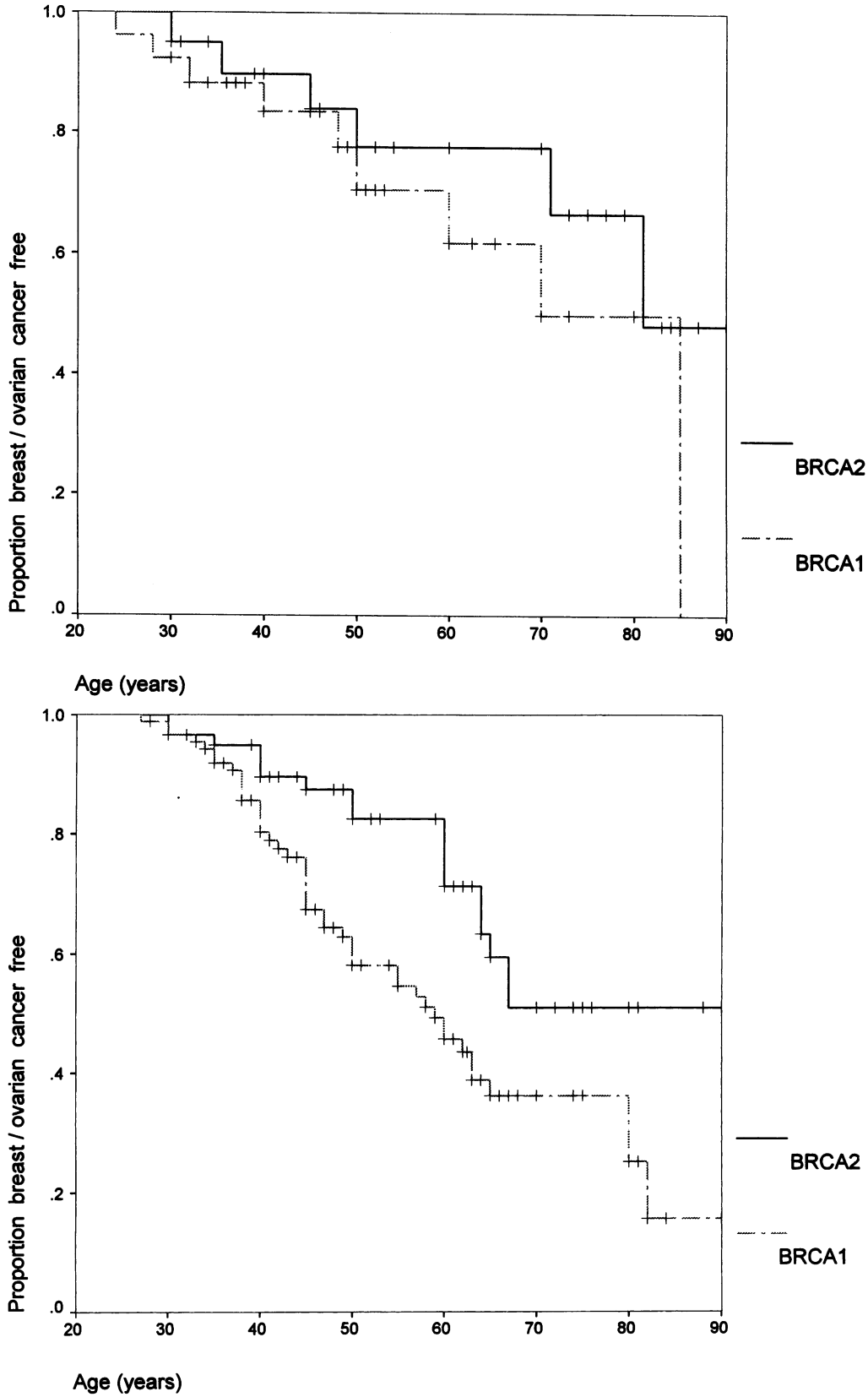


**Figure 1** Proportion of carriers remaining breast and ovarian cancer free: BRCA1 versus BRCA2. This proportion is the complement of the penetrance of these mutations (i.e., penetrance equals 1 minus the proportion remaining disease free). *Top,* Families ascertained on the basis of the ovarian-cases series. *Bottom,* Families ascertained by the SZMC cancer genetics clinic.

was 7/18 (39%) (it was 5/6 [83%] for cases with a minimal family history and was 2/12 [17%] for cases with no family history). Similar results were found in a selected series of ovarian cancer patients in Israel (Modan et al. 1996). Even though these cases were tested only for the 185delAG mutation (which accounted for only half of carriers in our cases), 39% (7/18) of cases with family history and 13% (8/61) of cases with no family history were found to be 185delAG carriers.

Although larger studies are necessary in order to determine the proportion of ovarian cancer in Ashkenazi women accounted for by these mutations, it is clearly much higher than originally estimated. The combined frequency of the founder BRCA1/BRCA2 mutations in the general Ashkenazi population has been estimated at 2% (Oddoux et al. 1996; Roa et al. 1996), compared with 1/300-1/800 in Caucasians (Claus et al. 1991). Despite the high carrier rate in the population, the incidence of ovarian cancer in Ashkenazi women in Israel is similar to that observed in other Western countries (Knapp and Berkowitz 1993). This suggests that penetrance of one or more of the founder Ashkenazi mutations is lower than has been estimated on the basis of published pedigrees. Lower penetrance also would explain the absence of significant family history in most carriers. Pedigree truncation because of the Holocaust is a common cause of lack of family history in Ashkenazi Jews, but, if it were the only cause, the incidence of ovarian cancer in current generations of Ashkenazi women in Israel would be expected to rise significantly—and it has not. As also was true of a previous study in Israel, we did not find a significant difference, in age at onset of ovarian cancer, between carriers of a founder mutation and those who did not carry such mutations (a result that is consistent with lower penetrance). This could be the result of combining all founder mutations together, because age at onset of ovarian cancer in BRCA2 carriers is significantly higher than that in BRCA1 carriers (68.3 years vs. 50.1 years; table 1).

Of 42 Ashkenazi families ascertained by the cancer genetics clinic, 25 (59%) segregated one of the founder mutations. These results confirm previous reports (Butler et al. 1996; Tonin et al. 1996) in which the three founder mutations accounted for approximately half of high-risk breast and ovarian cancer families. Interestingly, in families with a history of ovarian cancer, the frequency of BRCA2 6174delT families in this study is significantly higher than that observed in a multicenter study of 220 North American Ashkenazi pedigrees (Tonin et al. 1996) (8/22 vs. 8/82; $P = .02$). If BRCA2 6174delT families have lower penetrance, they perhaps were less likely to have been included in familial cancer series previously collected by multiple tertiary centers. In the clinic-ascertained families, the 5382insC mutation was as frequent as the other two founder mutations, even though its frequency in the population is approximately eightfold lower than that of either 185delAG or 6174delT. This phenomenon has been reported elsewhere (Tonin et al. 1996) and has been thought to suggest either increased penetrance of this mutation or underestimation of its true frequency in the population. Comparison of 5382insC penetrance versus 185delAG penetrance in our study population did not reveal any difference, but sample size is probably insufficient to detect a difference if the latter exists.

We found that families with any case of ovarian cancer were significantly more likely to segregate one of the founder mutations than were families with site-specific breast cancer. In a previous study (Tonin et al. 1996), ovarian cancer history was found to be a strong risk factor for presence of one of the BRCA1 mutations, but founder mutations accounted for only one-third of site-specific breast cancer families. Combined with the high frequency of carriers observed in our ovarian-cases series, we conclude that the three founder mutations account for a majority of breast-ovarian cancer families but for only a sizable minority of site-specific familial breast cancer in Ashkenazi Jews. It remains to be seen whether these families segregate other BRCA1/BRCA2 mutations, or whether other loci are involved.

### *Effects of Ascertainment and Specific Mutations on Penetrance*

We found that families ascertained on the basis of an ovarian-cases proband, regardless of family history, had lower penetrance than did families ascertained by a familial cancer clinic, although this difference was not statistically significant. For BRCA1 mutations, penetrance estimates for breast and ovarian cancer combined in this study's familial cases were statistically significantly lower than estimates in families used for linkage analysis. By 70 years of age BRCA1 penetrance in this study was 64% (95% CI 51%–77%), compared with 82% (95% CI 64%–91%) (Easton et al. 1993) and 87% (95% CI 72%–95%, breast cancer only) (Ford et al. 1994) in the Breast Cancer Linkage Consortium studies. These values are not directly comparable, both because families in this study are not as extensively characterized and because analysis included data based on family reports and statistical risk assignment. However, taken together with the observed difference, in penetrance, between ascertainment on the basis of an ovarian cancer case versus ascertainment on the basis of family history, our results suggest that current penetrance estimates may be inflated because of an ascertainment bias.

Penetrance of BRCA2 mutations for breast cancer has been estimated to be similar to that of BRCA1 mutations (Wooster et al. 1994). In this study, penetrance of the BRCA2 6174delT mutation was significantly lower than that of the BRCA1 185delAG and BRCA1 5382insC mutations. This was true for both breast and ovarian cancer combined and for breast cancer alone, with an

approximately twofold hazard ratio. Reduced BRCA2 6174delT penetrance has been inferred indirectly by comparison of its frequency in the population versus its frequency in breast cancer families (Oddoux et al. 1996; Roa et al. 1996). To the best of our knowledge, this is the first direct demonstration of differential penetrance of these mutations. Since data collection and analysis were done in an identical manner for all families studied, if there is a systematic bias it would be expected to be of similar magnitude in both BRCA1 families and BRCA2 families and should not have influenced comparisons between these groups. Although Ashkenazi Jews have a relatively high rate of breast cancer (Newell 1961; Steinitz et al. 1989) and may prove to have a truly high rate of pathogenic BRCA1/BRCA2 mutations (as has been observed in other diseases, such as Tay-Sachs), it is also possible that the observed high carrier rate is, at least in part, a reflection of the current ease of mutation detection in a genetically homogeneous population.

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