identified in this screen, including the Met1775Arg mutation, which had been detected in two unrelated African American families (Futreal et al. 1994; Miki et al. 1994), and the Cys64Gly mutation, which was identified once in an African American kindred (Castilla et al. 1994). This initial description of a spectrum of *BRCA1* mutations, two of which are recurrent in unrelated individuals from high-risk African American kindreds, should provide initial data for more-intensive genetic epidemiology studies in blacks of African descent.

QING GAO,[1] SUSAN NEUHAUSEN,[2] SHELLY CUMMINGS,[1] MICHAEL LUCE,[3] AND OLUFUNMILAYO I. OLOPADE[1]

[1]*Section of Hematology/Oncology, Department of Medicine, Committee on Cancer Biology, and Committee on Genetics, The University of Chicago Cancer Research Center, The University of Chicago, Chicago; and* [2]*Genetic Epidemiology Group, Department of Medical Informatics, University of Utah School of Medicine, and* [3]*Myriad Genetics, Inc., Salt Lake City*

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References

Castilla LH, Couch FJ, Erdos MR, Hoskins KF, Calzone KA, Garber JE, Boyd J, et al (1994) Mutations in the *BRCA1* gene in families with early-onset breast and ovarian cancer. Nat Genet 8:387–391

Eley JW, Hill HA, Chen VW, Austin DF, Wesley MN, Muss HB, Greenberg, RS, et al (1994) Racial differences in survival from breast cancer: results of the National Cancer Institute Black/White Cancer Survival Study. JAMA 272: 947–954

Futreal PA, Liu Q, Shattuck-Eidens D, Cochran C, Harshman K, Tasvtigian S, Bennett LM, et al (1994) *BRCA1* mutations in primary breast and ovarian carcinomas. Science 266:120–122

Gayther SA, Warren W, Mazoyer S, Russell P, Harrington PA, Chiano M, Seal S, et al (1995) Germline mutations of the *BRCA1* gene in breast and ovarian cancer families provide evidence for a genotype-phenotype correlation. Nat Genet 11:428–433

Goldgar DE, Reilly PR (1995) A common *BRCA1* mutation in the Ashenazim. Nat Genet 11:113–114

Hankey BF, Miller B, Curtis R, Kosary C (1994) Trends in breast cancer in younger women in contrast to older women. Monogr Natl Caner Inst 16:7–14

Kelsey JL, Gammon MD (1991) The epidemiology of breast cancer. CA Cancer J Clin 41:147–165

Miki Y, Swensen J, Shattuck-Eidens D, Futreal PA, Harshman K, Tavtigian S, Liu Q, et al (1994) A strong candidate for the breast and ovarian cancer susceptibility gene *BRCA1*. Science 266:66–71

Neuhausen SL, Mazoyer S, Friedman L, Stratton M, Offit K, Caligo A, Tomlinson G, et al (1996) Haplotype and phenotype analysis of six recurrent BRCA1 mutations in 61 families: results of an international study. Am J Hum Genet 58: 271–280

Parkin DM (1994) Cancer in developing countries. Cancer Surv 20:519–561

Shattuck-Eidens D, McClure M, Simard J, Labrie F, Narod S, Couch F, Hoskins K, et al (1995) A collaborative survey of 80 mutations in the *BRCA1* breast and ovarian cancer susceptibility gene. JAMA 273:535–541

Shiao YH, Chen VW, Scheer WD, Wu XC, Correa P (1995) Racial disparity in the association of p53 gene alterations with breast cancer survival. Cancer Res 55:1485–1490

Skolnick MH, Cannon-Albright LA (1992) Genetic predisposition to breast cancer. Cancer Suppl 70:1747–1753

Struewing JP, Abeliovich D, Peretz T, Avishai N, Kaback M, Collins FS, Brody LC (1995) The frequency of the *BRCA1* 185delAG mutation is approximately 1% in Ashkenzi Jewish individuals. Nat Genet 11:198–200

Tavtigian SV, Simard J, Rommens F, Couch D, Shattuck-Eidens S, Neuhausen S, Merajver S, et al (1996) The complete *BRCA2* gene and mutations in chromosome 13q-linked kindreds. Nat Genet 12:333–337

Wooster R, Bignell G, Lancaster J, Swift S, Seal S, Mangion J, Collins N, et al (1995) Identification of the breast cancer susceptibility gene *BRCA2*. Nature 378:789–792

Address for correspondence and reprints: Dr. Olufunmilayo I. Olopade, Department of Medicine, Section of Hematology/Oncology, University of Chicago, 5841 South Maryland Avenue, MC 2115, Chicago, IL 60637-1470. E-mail: fiolopade@mcis.bsd.uchicago.edu

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BRCA2 Mutations in Hereditary Breast and Ovarian Cancer in France

To the Editor:

BRCA2, a major gene responsible for susceptibility to breast cancer, located on chromosome 13q12-13, recently has been cloned (Wooster et al. 1994, 1995; Tavtigian et al. 1996). *BRCA2* germ-line mutations also have been shown to confer an elevated risk of ovarian and male breast cancer (Wooster et al. 1994; D. F. Eas-

ton, L. Steele, P. Fields, P. A. Daly, W. Ormiston, S. L. Neuhausen, and D. Ford, personal communication). Initial mutation studies showed a wide variety of *BRCA2* alterations located throughout the coding sequence (Wooster et al. 1995; Couch et al. 1996; Phelan et al. 1996; Tavtigian et al. 1996; Gayther et al. 1997; Serova et al. 1997). However, several recurrent mutations have been identified, most notably in specific populations such as the Ashkenazi Jews (Berman et al. 1996; Neuhausen et al. 1996; Oddoux et al. 1996; Roa et al. 1996; Tonin et al. 1996) or the Icelanders (Johannesdottir et al. 1996; Thorlacius et al. 1996).

To determine the spectrum of *BRCA2* mutations in French kindreds, we have screened the entire coding region of *BRCA2* in a sample of 77 breast cancer families accrued through genetic-counseling clinics in nine centers located in different parts of France: Institut Curie, Paris; Institut Gustave Roussy, Villejuif; Centre Léon Bérard, Lyon; Centre Jean Perrin, Clermont-Ferrand; Centre Hospitalier Universitaire, Grenoble; Institut Bergonié, Bordeaux; Centre René Gauducheau, Nantes; Centre René Huguenin, St. Cloud; and Centre Hospitalier Universitaire, Rouen. The families selected met one of the following criteria in first- and second-degree relatives: at least either (1) two cases of invasive breast cancer diagnosed at <50 years of age; (2) three cases of breast cancer, with at least one diagnosed at <40 years of age; (3) one case of breast cancer diagnosed at <50 years of age and at least one case of ovarian cancer diagnosed at any age; or (4) one case of male breast cancer and at least one case of female breast cancer diagnosed at any age. Sixty-six families were shown to be negative in a *BRCA1* mutation search done by either denaturing gradient gel electrophoresis, SSCP, heteroduplex analysis (HDA) combined with the protein-truncation test (PTT), or direct DNA sequencing, depending on the technique(s) used in each center. Eleven families were not tested for *BRCA1* status, since, on the basis of the presence of at least one case of male breast cancer, they were presumed to be *BRCA2* families. Forty-three of the 66 were female breast cancer families (mean number of cases 3.5), and 23 were breast/ovarian cancer families (mean number of ovarian cancer cases 1.3; mean number of breast cancer cases 2.5). The number of affected people in each of the 77 families was 2–10.

The analysis was performed by use of genomic DNA of the youngest affected individual available in each family. The average age of those actually tested was 40 years for female breast cancer patients, 54 years for ovarian cancer patients, and 47 years for male breast cancer patients. Exons 10 and 11 were examined with the PTT, and the rest of the coding region was examined by HDA, as described elsewhere (Serova et al. 1997). cDNA was available for 22 families and was analyzed by HDA using primer pairs published by Serova et al. (1997).

We identified 14 frameshift, nonsense, and splice-site truncating mutations and 2 missense variants, distributed widely across the gene, each of them being detected only once in our series of families (table 1). It remains to be explored whether these missense alterations represent

Table 1

Germ-Line *BRCA2* Mutations in French Families

Family	Female Breast Cancer	Male Breast Cancer	Ovarian Cancer	Type	Exon(s)	Codon	Nucleotide	Mutation
CU404	5	0	0	Frameshift	3	23	297	delT, resulting in ter24
CRG32	8	0	0	Nonsense	10	414	1469	Leu414ter
CU538	2	0	0	Frameshift	11	936	3034	del4, resulting in ter958
IGR1241	2	0	1	Frameshift	11	1782	5573	delAA, resulting in ter1785
IGR665	3	0	0	Frameshift	11	1906	5946	delCT, resulting in ter1909
IGR1675	4	0	0	Nonsense	11	1953	6085	GluI953ter
IGR682	2	0	1	Frameshift	11	2068	6431	delT, resulting in ter2069
IGR684	3	0	0	Frameshift	11	2215	6872	del4, resulting in ter2227
CU579	2	0	1	?	12 and 13			Exons 12 and 13 missing in transcript, resulting in tcr2311
CU487	4	0	0	Splice mutation	13	2336	7235	G→C, resulting in exons 12 and 13 missing in transcript, resulting in ter2311
CLB95052	5	0	0	Frameshift	16	2561	7909	insT, resulting in ter2565
CFD80	3	2	0	Nonsense	18	2714	8368	Gln2714ter
CLB94090	3	1	0	Missense	22	2951	9079	Ala2951Thr
CU127	4	0	2	Frameshift	23	3009	9254	del5, resulting in ter3015
CLB95122	2	0	1	Frameshift	23	3030	9317	insA, resulting in ter3043
CLB94002	2	0	1	Missense	25	3098	9520	Tyr3098His

deleterous mutations or rare polymorphisms; neither of them, however, was detected in 94 control chromosomes from the French population.

Eight of 43 female breast cancer families were found to carry mutations in *BRCA2*, a proportion similar to those found in other studies on *BRCA2* mutations in *BRCA1*-negative breast cancer families satisfying the same selection criteria as were met by families in our set (Gayther et al. 1997; Serova et al. 1997). The proportion of *BRCA2*-positive families was much higher in the group of families containing four or more affecteds (6 of 18) than in small families of two or three breast cancer cases (4 of 25).

We detected only five frameshifts and one missense alteration in the 23 breast-ovarian cancer families tested, 18 kindreds remaining attributable to neither *BRCA1* nor *BRCA2*. Surprisingly low mutation yield (one nonsense variant and one missense variant) also was found in male breast cancer families. The low proportion of *BRCA2* mutations in this sample of breast-ovarian and male breast cancer families might be explained by chance aggregation of cancer cases in some of them, since eight of the mutation-negative breast-ovarian cancer families each have only one case of breast cancer diagnosed at <50 years of age and one case of ovarian cancer, and four male breast cancer families each contain only one female breast cancer case. This is consistent both with the results reported by Couch et al. (1996), who found only seven *BRCA2* mutations in 50 males with breast cancer, 80% having an unspecified family history of breast cancer, and with the results reported by Friedman et al. (1997), who have detected two *BRCA2* mutations in 54 male breast cancer patients, 30% of whom had a positive family history of breast and/or ovarian cancer. Because of the possibility that the only person screened was a phenocopy, we could have failed to identify some mutations. The techniques used here for mutation screening have been shown to be efficient in other studies (Keen et al. 1991; Tassabehji et al. 1992; White et al. 1992; van der Luijt et al. 1994; Gayther et al. 1995, 1997; Hogervorst et al. 1995). The *BRCA2* coding region was analyzed by use of cDNA as well as genomic DNA, in 22 families. One of the mutations (in family CU579) appeared to be detectable on cDNA only, thus suggesting that some mutations might have been missed in the group of families in which only genomic DNA was examined.

Gayther et al. (1997) have reported that *BRCA2* truncating mutations in families with a high proportion of ovarian cancer appear to be clustered in a region of 3.3 kb in exon 11, between nucleotides 3035 and 6629. Data from our families provide some support for this clustering. The four families with mutations in this 3.3-kb region contain 2 ovarian and 11 breast cancer cases, compared with 4 ovarian and 38 breast cancer cases in 10 families with mutations elsewhere (odds ratio 1.7; $P = .05$).

Eight of 14 truncating mutations have not been published before. The frameshift mutation 9254del5 (in family CU127) has been identified previously in another family (CU159) also collected by Institut Curie (Tavtigian et al. 1996). Both families appeared to originate from Catalonia. The missense variant Ala2951Thr found in an affected niece and aunt in family CLB94090 also have been detected in a French breast cancer patient with no family history of cancer (O. M. Serova-Sinilnikova, unpublished data). These findings indicate the possibility of a founder effect, within the French population, for these two *BRCA2* alterations. Five other mutations already have been reported to the Breast Cancer Information Core (1997) web site (http:/www.nchgr.nih.gov/Intramural_research/Lab_transfer/Bic/index.html) and/or in the literature, all identified in families with geographic residence other than France, one in Belgium and the others in the United Kingdom and the United States. 3034del4 was found five times, 9325insA and 5946delCT three times, and Glu1953ter and 5573delAA twice (Breast Cancer Information Core 1996; Takahashi et al. 1996; Gayther et al. 1997; Serova et al. 1997). It will be of interest to investigate whether some of these mutations have a common origin or constitute mutation hot spots in the coding region of the *BRCA2* gene.

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OLGA M. SEROVA-SINILNIKOVA,[1,2,3] LAETITIA BOUTRAND,[1] DOMINIQUE STOPPA-LYONNET,[5] BRIGITTE BRESSAC-DE-PAILLERETS,[6] VALÉRIE DUBOIS,[3] CHRISTINE LASSET,[4] NICOLAS JANIN,[6] YVES-JEAN BIGNON,[7] MICHEL LONGY,[8] CHRISTINE MAUGARD,[9] ROSETTE LIDEREAU,[10] DOMINIQUE LEROUX,[11] THIERRY FREBOURG,[12] SYLVIE MAZOYER,[1,2] AND GILBERT M. LENOIR[1,2,3]

[1]*Centre International de Recherche sur le Cancer,* [2]*Laboratoire de Génétique, UMR5641 CNRS,* [3]*Hôpital Edouard Herriot, and* [4]*Centre Léon Bérard, Lyon;* [5]*Institut Curie, Paris;* [6]*Institut Gustave Roussy, Villejuif, France;* [7]*Centre Jean Perrin, Clermont-Ferrand, France;* [8]*Institut Bergonié, Bordeaux;* [9]*Centre René Gauducheau, Nantes;* [10]*Centre René Huguenin, Saint Cloud, France;* [11]*Centre Hospitalier Universitaire de Grenoble, Grenoble; and* [12]*Centre Hospitalier Universitaire, Hôpital Ch. Nicolle, Rouen*

References

Berman DB, Costalas J, Schultz DC, Grana G, Daly M, Godwin AK (1996) A common mutation in *BRCA2* that predisposes to a variety of cancers is found in both Jewish Ashkenazi and non-Jewish individuals. Cancer Res 56:3409–3414

Breast Cancer Information Core (1997) http:/www.nchgr.nih.gov/Intramural_research/Lab_transfer/Bic

Couch FJ, Farid LM, DeShano ML, Tavtigian SV, Calzone K, Campeau L, Peng Y, et al (1996) *BRCA2* germline mutations in male breast cancer cases and breast cancer families. Nat Genet 13:123–125

Friedman LS, Gayther SA, Kurosaki T, Gordon D, Noble B, Casey G, Ponder BAJ, et al (1997) Mutation analysis of BRCA1 and BRCA2 in male breast cancer population. Am J Hum Genet 60:313–319

Gayther SA, Mangion J, Russel P, Seal S, Barfoot R, Ponder BAJ, Stratton MR, et al (1997) Variation of risks of breast and ovarian cancer associated with different germline mutations of the *BRCA2* gene. Nat Genet 15:103–105

Gayther SA, Warren W, Mazoyer S, Russel PA, Harrington PA, Chiano M, Seal S, et al (1995) Germline mutations of the *BRCA1* gene in breast/ovarian cancer families: evidence for a genotype/phenotype correlation. Nat Genet 11:428–433

Hogervorst FBL, Cornelis RS, Bout M, van Vliet M, Oosterwijk JC, Olmer R, Bakker B, et al (1995) Rapid detection of *BRCA1* mutations by the protein truncation test. Nat Genet 10:208–212

Johannesdottir G, Gudmundsson J, Berghthorsson JT, Arason A, Agnarsson BA, Eiriksdottir G, Johannsson OT, et al (1996) High prevalence of the 999del5 mutation in Icelandic breast and ovarian cancer patients. Cancer Res 56:3663–3665

Keen J, Lester D, Inglehern C, Curtis A, Bhattacharya S (1991) Rapid detection of single base mismatches as heteroduplexes on Hydrolink gels. Trends Genet 7:5

Neuhausen SL, Gilewski T, Norton L, Tran T, McGuire P, Swensen J, Hampel H, et al (1996) Recurrent *BRCA2* 6174delT mutations in Ashkenazi Jewish women affected by breast cancer. Nat Genet 13:126–128

Oddoux C, Streuwing JP, Clayton CM, Neuhausen S, Brody LC, Kaback M, Haas B, et al (1996) The carrier frequency of the *BRCA2* 6174delT mutation among Ashkenazi Jewish individuals is approximately 1%. Nat Genet 13:188–190

Phelan CM, Lancaster JM, Tonin P, Gumbs C, Cochran C, Carter R, Ghadirian P, et al (1996) Mutation analysis of the *BRCA2* gene in 49 site-specific breast cancer families. Nat Genet 13:120–122

Roa BB, Boyd AA, Volcik K, Richards CS (1996) Ashkenazi Jewish population frequencies for common mutations in *BRCA1* and *BRCA2*. Nat Genet 14:185–187

Serova OM, Mazoyer S, Puget N, Dubois V, Tonin P, Shugart YY, Goldgar D, et al (1997) Mutations in *BRCA1* and *BRCA2* in breast cancer families: are there more breast cancer–susceptibility genes? Am J Hum Genet 60:486–495

Takahashi H, Chiu H-C, Bandera CA, Behbakht CA, Liu PC, Couch FJ, Weber BL, et al (1996) Mutations of the *BRCA2* gene in ovarian carcinomas. Cancer Res 56:2738–2741

Tassabehji M, Read AP, Newton VE, Harris R, Balling R, Gruss P, Strachan T (1992) Waardenburg's syndrome patients have mutations in the human homologue of the *Pax-3* paired box gene. Nature 355:635–636

Tavtigian SV, Simard J, Rommens J, Couch F, Shattuck-Eidens D, Neuhausen S, Merajver S, et al (1996) The complete *BRCA2* gene and mutations in chromosome 13q-linked kindreds. Nat Genet 12:333–337

Thorlacius S, Olafsdottir G, Tryggvadottir L, Neuhausen S, Jonasson JG, Tavtigian SV, Tulinius H, et al (1996) A single *BRCA2* mutation in male and female breast cancer families from Iceland with varied cancer phenotypes. Nat Genet 13:117–119

Tonin P, Weber B, Offit K, Couch F, Rebbeck T, Neuhausen S, Godwin A, et al (1996) Frequency of recurrent *BRCA1* and *BRCA2* mutations in Ashkenazi Jewish breast cancer families. Nat Med 2:1174–1183

van der Luijt R, Khan PM, Vasen H, van Leeuwen C, Tops C, Roest P, den Dunnen J, et al (1994) Rapid detection of translation-terminating mutations at the adenomatous polyposis coli (APC) gene by direct protein truncation test. Genomics 20:1–4

White MB, Carvalho M, Derse D, O'Brien SJ, Dean M (1992) Detecting single base substitutions as heteroduplex polymorphisms. Genomics 12:301–306

Wooster R, Bignell G, Lancaster J, Swift S, Seal S, Mangion J, Collins N, et al (1995) Identification of the breast cancer gene *BRCA2*. Nature 378:789–791

Wooster R, Neuhausen SL, Mangion J, Quirk Y, Ford D, Collins N, Nguyen K, et al (1994) Localization of a breast cancer susceptibility gene, *BRCA2*, to chromosome 13q12-13. Science 265:2088–2090

Address for correspondence and reprints: Dr. Gilbert M. Lenoir, Centre International de Recherche sur le Cancer, 150 cours Albert Thomas, 69372 Lyon Cedex 08, France. E-mail: lenoir@iarc.fr

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Frequently Occurring Germ-Line Mutations of the *BRCA1* Gene in Ovarian Cancer Families from Russia

To the Editor:

Germ-line mutations of the *BRCA1* gene are thought to be responsible for the cancer in 80%–90% of families containing multiple cases of both breast and ovarian cancer (Easton et al. 1993). In addition, *BRCA1* is predicted to be responsible for the cancer in the majority of site-specific ovarian cancer families containing three or more cases of epithelial ovarian cancer and no cases of early-onset breast cancer (Steichen-Gersdorf et al. 1994). Since the isolation of *BRCA1* (Miki et al. 1994), >130 distinct germ-line mutations have been reported, primarily in multiple-case breast/ovarian cancer families (Couch et al. 1996). The majority of these mutations