ELECTRONIC LETTER

Novel *BRCA2* mutation in a Polish family with hamartoma and two male breast cancers

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Breast cancer is a rare disease in men and represents only approximately 1% of all breast cancer cases.[1] Several factors have been reported to increase the risk for male breast cancer, including alteration in hormonal status, Klinefelter's syndrome, family history of breast cancer, and occupational exposures to high temperature, electromagnetic fields, and radiation.[2 3] Germline mutations in the androgen receptor gene and, particularly, in the *BRCA2* gene are thought to increase breast cancer risk in male carriers.[4–9] Mutations in *BRCA2* may account for 75% of families in which there is at least one case of male breast cancer and at least three more cases of female breast cancer.[10] Most families have a single case of male breast cancer and familial clusters of male breast cancer cases are unusual. Recently, *BRCA2* mutation analyses in male breast cancer were performed in the Polish population and five novel mutations were detected; however, aggregation of male breast cancer cases was observed in only one family from north eastern Poland.[11 12] Here, we report a novel *BRCA2* mutation in a Polish family with two male breast cancer cases in a father and son and a first case of *BRCA2* mutation with loss of heterozygosity in a patient with lung hamartoma.

METHODS

Genomic DNA was isolated from peripheral blood lymphocytes of the proband (male breast cancer patient) and seven additional family members using the Wizard Genomic DNA Purification Kit (Promega, Madison, WI, USA). Paraffin embedded tumour samples were obtained for the proband and his brother diagnosed with hamartoma chondromyxoides. Mutation analysis of the *BRCA2* gene was performed by the PCR-SSCA-HA method. The polymerase chain reaction was used to detect loss of heterozygosity (LOH) at polymorphic microsatellite markers spanning the *BRCA2* region, by comparing the allelic pattern in tumour and blood DNA. Three microsatellite markers, D13S260, D13S171, and D13S267, were analysed. LOH was considered to be present if the intensity of alleles varied by more than 50%.

RESULTS AND DISCUSSION

The family pedigree is shown in fig 1. It includes two cases of male breast cancer, one case of female genital tract cancer, and one case of hamartoma chondromyxoides. The proband, III.1, developed breast cancer at the age of 61. Histological examination of resected breast tissue showed infiltrating ductal carcinoma (grade III). The proband's father (patient II.3) died at the age of 74 from breast cancer, two years after diagnosis. He underwent mastectomy, chemotherapy, and radiotherapy. The proband's brother (patient III.3) was diagnosed at the age of 56 with hamartoma chondromyxoides in the right lung and underwent surgery. Mammography performed at the age of 60 showed minor fibrosis with no influence on the gland structure. The father's sister (II.6) died from genital tract cancer. No other cancers have been observed in the family. DNA samples were available from patients III.1 and III.3, as well as from the unaffected members III.2, III.4, III.5, IV.1, IV.2, and IV.5.

Mutation analysis of the *BRCA2* gene showed a novel frameshift mutation 6621del4 in exon 11, predicted to generate premature termination of translation at codon 2136. The mutation was present in the proband (III.1) and the hamartoma patient (III.3). The mutation was also found in two (IV.1, aged 36 and IV.5, aged 19) of six unaffected family members and the remaining four persons (III.2, III.4, III.4, and IV.2) did not have the mutation. To evaluate the involvement of the *BRCA2* 6621del4 mutation in the male breast cancer and hamartoma progression, we mapped loss of heterozygosity at the *BRCA2* locus in tumour samples. Both cases were

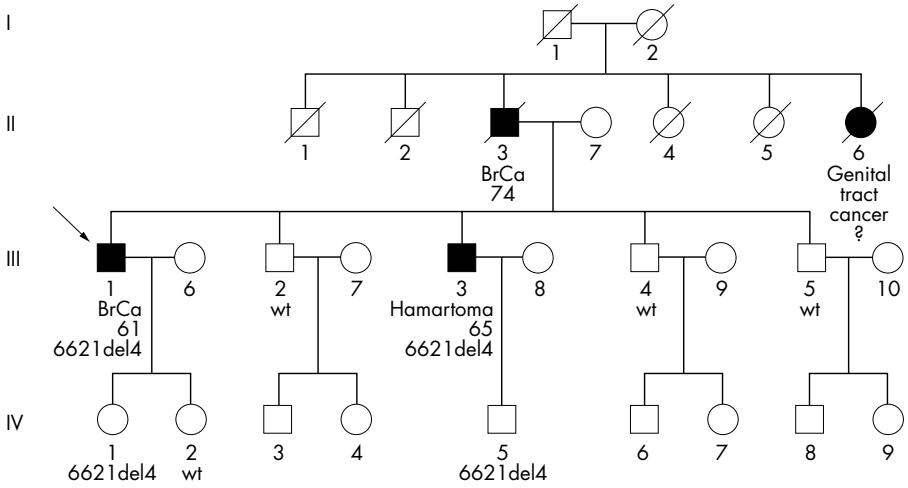

Figure 1 Male breast cancer and hamartoma family. Solid symbols = affected subjects, wt = wild type.

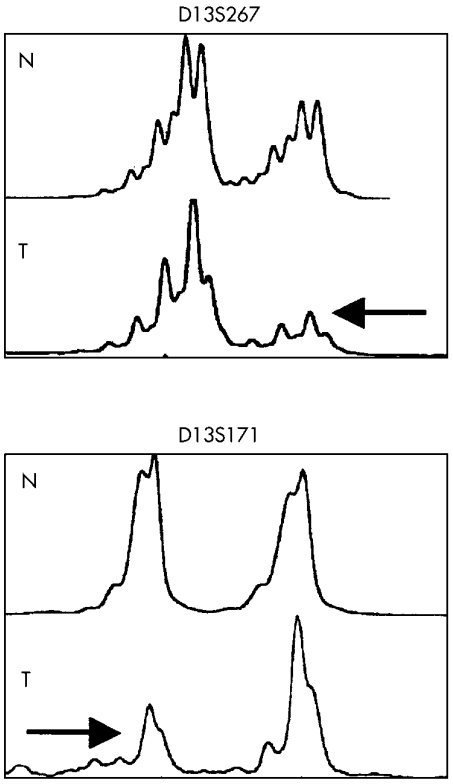

Figure 2 Loss of heterozygosity at the *BRCA2* locus (microsatellite markers D13S267 and D13S171) in hamartoma chondromyxoides. N = normal tissue, T = tumour tissue, arrow indicates deleted allele.

informative (heterozygous) for LOH at two *BRCA2* markers analysed, D13S171 and D13S267. LOH was seen at these markers in both breast tumour and hamartoma (fig 2).

Carriers of germline mutations in the *BRCA2* gene are known to be at high risk of breast and ovarian cancer.[10] There are several reports suggesting that *BRCA2* mutations confer a predisposition to other cancers including pancreatic cancer, prostate cancer, gallbladder and bile duct cancer, stomach cancer, and malignant melanoma.[13 14] Analysis of tumours occurring in cancer patients verified as *BRCA2* carriers showed a high frequency of LOH on chromosome 13q12-13 in female breast cancer, but also in male breast cancer as well as in tumours of the prostate, ovary, cervix, colon, and uterus.[15] The high frequency of loss of the wild type chromosome observed in tumours of *BRCA2* carriers suggest a strong selection of tumour cells with both alleles of the *BRCA2* gene being mutated.[15] This is the first report of mutation in the *BRCA2* gene in a benign neoplasm with loss of heterozygosity. Hamartomas are the most common tumorous lesions of the lung, occurring in approximately 0.3% of the general population.[16] For many years, hamartomas were classified as focal overgrowths of disorganised but otherwise normal lung tissue, such as cartilage, smooth muscle, other connective tissue elements, and respiratory tract epithelium.[17] Neoplastic transformation is a result of somatic cell mutations (point mutations, chromosomal aberration) occurring in a set of genes essential for proliferation and differentiation. Several studies have suggested that most hamartomas of the lung are characterised by clonal chromosomal abnormalities.[18–20] Different rearrangements involving chromosomes 6p21 and 14q24 were common, as were changes of 12q13-15 and 17p. Detection of acquired, specific, clonal chromosome aberrations in pulmonary hamartomas suggest a neoplastic genesis of such tumours.[20] LOH at the *BRCA2* locus in pulmonary hamartoma occurring in a *BRCA2* mutation carrier suggests a possible involvement of germline mutations of the *BRCA2* gene in the development of benign tumours. The number of different cancers seen in *BRCA2* carriers together with loss of the wild type allele in tumours suggest that mutations in the *BRCA2* gene may lead to the development of cancer in various organs.

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