BRIEF REPORT

The HOXB13 p.Gly84Glu mutation is not associated with the risk of breast cancer

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Abstract Recently, the HOXB13 gene has been shown to be a susceptibility gene for prostate cancer. HOXB13 is overexpressed in breast cancer tissues and HOXB13 expression in combination with low expression of IL17BR is predictive for a tamoxifen response in ER-positive breast cancers. Based on observations, we hypothesized that the HOXB13 p.Gly84Glu mutation might be associated with breast cancer risk. We genotyped this mutation in the germline DNA of 4,037 women with breast cancer (including 1,082 familial cases) and in 2,762 controls from Canada and Poland. Seven heterozygous carriers of the HOXB13 p.Gly84Glu mutation were found in the cases (0.17 %) compared to four carriers among the controls (0.14 %; OR = 1.2, 95 % CI = 0.34–4.1, p = 1.0). Only one of the seven carriers had a family history of breast cancer. This study does not support the hypothesis that women who carry the HOXB13 Gly84Glu mutation are at increased risk of breast cancer.

Keywords Breast cancer · HOXB13 · p.Gly84Glu

Abbreviations BRCA1 Breast cancer susceptibility gene 1
BRCA2 Breast cancer susceptibility gene 2
BRIP1 BRCA1 interacting protein 1
CHEK2 Checkpoint kinase 2
CI Confidence interval
DNA Deoxyribonucleic acid
ER Estrogen receptor
Glu Glutamine
Gly Glycine
HOXB13 Homeobox B13
OR Odds ratio
PALB2 Partner and localizer of BRCA2

Introduction

The risk of breast cancer varies between women, and genetic susceptibility plays an important role in the etiology of the disease. Two major breast cancer susceptibility genes, BRCA1 [1] and BRCA2 [2], were identified in the 1990s. Hereditary breast cancer cases are estimated to account for 10 % of all breast cancers and BRCA1 and BRCA2 genes are responsible for only 15–20 % of the inherited breast cancers [3]. More recently, other breast cancer genes such as BRIP1 [4], CHEK2 [5], and PALB2 [6] have been discovered, but mutations in those genes are rare and account for only a small percentage of families.

Recently, the HOXB13 p.Gly84Glu mutation was reported to be associated with the increased risk of prostate cancer [7]. This mutation increases the risk of prostate cancer by 5- to 10-fold [8, 9]. HOXB13 belongs to the HOX family of transcription factor genes, each containing a homeodomain.
These genes are located in four clusters throughout the human genome (HOXA, B, C, and D) and are involved in embryonic development [10], but they are also expressed in different organs in the adult, including breast tissue [11]. HOXB13 is overexpressed in breast cancers compared to the normal breast [12, 13], and high expression of HOXB13 in combination with low expression of IL17BR has been reported to be predictive of a poor response to tamoxifen therapy in ER-positive breast cancer patients [14, 15]. Based on these observations, we hypothesized that the HOXB13 p.Gly84Glu mutation might be associated with breast cancer risk.

Methods

We genotyped the HOXB13 p.Gly84Glu mutation in germline DNA of 4,037 breast cancer cases and in 2,762 controls. Subjects included 1,804 breast cancer cases and 925 controls from Canada and 2,233 breast cancer cases and 1,837 controls from Poland. The Canadian cases were all white women and were selected from patients who received genetic counseling at the cancer genetics clinics in Toronto between 1998 and 2011. The mean age at diagnosis of these patients was 52.2 years (range 18–89). The Polish cases include prospectively ascertained series of invasive breast cancer diagnosed throughout Poland with early onset breast cancer (age range 20–50; mean 44.4). They were ascertained from 18 different hospitals between 1996 and 2003. Of the total 4,037 breast cancer cases, 1,085 patients had at least one affected individual among their first-degree relatives (familial cases). Canadian controls were obtained from the Healthwatch (HW) screening program at the Women’s College hospital. These are healthy women with no prior history of cancer who had attended a multimodal screening clinic for well women at the Women’s College Hospital in Toronto. The Polish controls consisted of 1,837 unselected cancer-free women (age range 24–84 years; mean age 54.0 years), selected at random from the computerized patient lists of five large family practices located in the region of Szczecin in 2003. Cases and controls were restricted to women of European origin as previous studies in prostate cancer [7, 8] found the HOXB13 p.Gly84Glu mutation only among men of European decent. The study was approved by the ethics review board of the participating institutions.

Genotyping of the HOXB13 p.Gly84Glu mutation among cases and controls was performed using the TaqMan assay on ABI 7500 fast and 7900 real-time systems (Applied Biosystems Co., Foster City, CA, USA). All mutation carriers identified by genotyping were confirmed by direct sequencing using the BigDye Terminator Cycle Sequencing kit on an ABI 3500XL DNA Analyzer (Applied Biosystems Co., Foster City, CA, USA). We compared the frequency of the HOXB13 p.Gly84Glu mutation between cases and controls using Fisher’s exact test and calculated odds ratios (OR) and their 95% confidence intervals (CI) based on 2x2 table analysis of the cases and controls. All statistical tests were two-sided and p values <0.05 were considered statistically significant.

Table 1 Clinical characteristics of 11 individuals carrying germline HOXB13 G84E mutation

| No. | Origin | Subjects | Age at diagnosis, y | Estrogen receptor | Progesterone receptor | HER2 | Grade | Family history b |
|-----|--------|----------|--------------------|------------------|---------------------|------|-------|------------------|
| 1   | Canada | Case     | 78                 | +                | +                   | ?    | Ca ductal G3 | None             |
| 2   | Canada | Case     | 74                 | +                | +                   |      | Ca ductal G2 | Father (PrCa), maternal aunt (BrCa) |
| 3   | Poland | Case     | 50 a               | +                | ++                  | –    | Ca ductal Gx | None             |
| 4   | Poland | Case     | 50                 | –                | –                   | –    | Ca ductal G3 | None             |
| 5   | Poland | Case     | 47                 | +                | –                   | ?    | Ca ductal G1, G2 | Mother (BrCa), paternal aunt (PrCa) |
| 6   | Poland | Case     | 47                 | –                | –                   | +    | Ca ductal G1, G2 | Father (PrCa), paternal grandfather (PrCa) |
| 7   | Poland | Case     | 41                 | –                | –                   | +    | Ca medullar | None             |
| 8   | Canada | Control  | 57                 | NA               | NA                  | NA   | NA                | None             |
| 9   | Poland | Control  | 50                 | NA               | NA                  | NA   | NA                | None             |
| 10  | Poland | Control  | 70                 | NA               | NA                  | NA   | NA                | None             |
| 11  | Poland | Control  | 40                 | NA               | NA                  | NA   | NA                | None             |

a Poland cases were selected for their age and they were all under 50 years of age

b Family history of breast cancer (BrCa) and prostate cancer (PrCa) among the first-and second-degree relatives of the probands (+) or (++) positive; (−) negative; ? unknown; NA not available; HER2 Human Epidermal Growth Factor Receptor 2
Results

The detailed clinical characteristics of the cases and controls carrying the HOXB13 p.Gly84Glu mutation are shown in Table 1. Seven heterozygous carriers of the HOXB13 p.Gly84Glu mutation were identified among the 4,037 cases (0.17 %) and four carriers were seen among the 2,762 controls (0.14 %) \((OR = 1.2, 95 \% CI = 0.3–4.1, p = 1.0)\). Only one of the seven carrier cases had a first-degree relative with breast cancer, and after limiting the comparison to familial cases, still no association was seen between HOXB13 p.Gly84Glu and breast cancer \((OR = 0.6, 95 \% CI = 0.1–5.7, p = 0.9)\).

Discussion

The association of HOXB13 p.Gly84Glu and increased risk of prostate cancer has been confirmed in several large scale case–control studies \([8–10]\). In a follow-up study, Alanee et al. reported that this mutation was also associated with an increased risk of familial breast cancer \((OR = 5.7, 95 \% CI: 1.0–40.7, p = 0.02)\). The authors found that, among study subjects with familial breast cancer who were negative for BRCA1/2 mutations, the carrier frequency of p.Gly84Glu (0.7 %) was seven times higher than in their control group (0.1 %). This study was based on 877 familial cases, out of which 6 had a mutation. In our much larger series of familial and non-familial cases, we observed similar frequencies between all cases and controls \((0.17 \text{ and } 0.14 \% \text{ respectively})\) and even lower frequency \((0.09 \%)\) was seen among familial cases and we did not confirm the association between HOXB13 p.Gly84Glu and increased risk of breast cancer. However, the HOXB13 p.Gly48Glu allele is very rare \((\text{approximately one in a thousand})\) and we are unable to rule out a small effect.

The relatively high expression level of HOXB13 in breast tumor cells compared to normal breast cells \([12–15]\) suggests a possible oncocenic role for HOXB13, although activating mutations have not been reported. However, the prostate cancer studies are unable to distinguish between an oncogenic or tumor suppressor effect for HOXB13 \([17]\), given that it is not known if the p.Gly84Glu mutation leads to loss or gain of function and truncating variants which are suggestive of a tumor suppressor effect have not been seen \([7]\).

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Conflict of interest None

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