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Original Paper

BRCA1-Positive Patients are Small for Gestational Age Compared with their Unaffected Relatives

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The *BRCA1* gene is thought to exert its main function early in life. We, therefore, studied the effects of *BRCA1* mutations on birth weight and birth length. This was carried out by comparing 33 women with and without mutations. Birth weight and length were obtained from a self-administered questionnaire. *BRCA1* mutation carriers had a significantly lower birth weight ($P = 0.0041$) compared with non-carriers, after adjustment for gestational age. They were also significantly shorter at birth compared with their unaffected relatives ($P = 0.0060$), after adjustment for gestational age. The *BRCA1* gene thus seems to influence the carriers *in utero*. The findings could imply that humans heterozygotic for the *BRCA1* mutations may be influenced by the mutations during development *in utero*.

Key words: *BRCA1*, birth weight, birth length
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INTRODUCTION

The *BRCA1* gene was first localised in 1990 by Hall and associates [1] via linkage analysis in families with early onset breast cancer. This finding was later confirmed by Narod and colleagues in 1991 [2]. Cloning of the gene was successfully carried out in 1994 [3]. Numerous mutations have been found within the gene [4]. The *BRCA1* gene is located on 17q21 and acts as a tumour suppressor gene. The lifetime risk of developing cancer in an affected woman is approximately 80% for breast cancer [4, 5]. Another breast cancer susceptibility gene *BRCA2* has been identified on 13q12–13 [6–8]. *BRCA2* predisposes for both male and female breast cancer, but to a lesser extent ovarian cancer. Disease-causing mutations are found in approximately 25% of families with true dominant pedigrees, *BRCA1* more frequently than *BRCA2* [9, 10]. Few studies have yet addressed the function of *BRCA1* or *BRCA2*.

In mice, a homologue gene to the *BRCA1* gene in humans has been identified [11]. There is a 72% identity between the human and the murine *BRCA1* gene at the nucleotide level [11]. Mice with deletions of both alleles die *in utero*, whereas heterozygotic mice do not differ phenotypically from mice with both alleles intact [12–14]. However, size differences in *BRCA1* heterozygotic mice compared with wild-type littermates have not been carefully examined. Therefore, previous studies in mice should be cautiously interpreted. Whether heterozygotic *BRCA1* mutations in humans have any embryonal effects or not has not yet been studied. We, therefore, wanted to see if a *BRCA1* mutation influenced birth weight and birth length, as well as final height and weight, in affected family members compared with their unaffected relatives.

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MATERIALS AND METHODS

Patients belonging to families in which a *BRCA1* mutation has been found were included in this study. Patient data were obtained both from women who had sought advice at the oncogenetic reception at Lund University Hospital, Sweden, as well as from research families. At the time of genetic testing, individuals were requested to fill out a detailed questionnaire, from which we obtained our information. Questions covered birth weight, birth length and gestational age, as well as present weight and height. Birth weight and length were measured immediately after birth and registered on the birth certificate. Length was measured from the head to the feet with the baby lying on its back.

Genetic testing for the *BRCA1* and *BRCA2* genes was offered through an oncogenetic clinic in Lund if an individual had at least three first degree relatives with breast cancer and with one diagnosed before the age of 50 years; or two first degree relatives with breast cancer and with one diagnosed

before the age of 40 years; or one first degree relative with breast cancer diagnosed before the age of 30 years. 33 women belonging to families with known *BRCA1* mutation agreed to undergo genetic testing themselves and completed the aforementioned questions in their questionnaires prior to testing. All were born between 1936 and 1971. 13 women were *BRCA1* mutation carriers, 11 of whom had inherited the gene from their mother. 20 women were non-carriers and 7 had a *BRCA1*-positive mother. Birth weight was missing for one *BRCA1*-negative woman. Birth length was missing for one *BRCA1*-positive woman and two *BRCA1*-negative women (Table 1). Because of the rarity of testing men, only data from women were used in this study.

BRCA1 *analyses*

The entire coding region of the *BRCA1* gene was screened for mutations using genomic DNA and the protein truncation test (PTT) or single-stranded conformation polymorphism (SSCP) analysis, followed by direct sequence analysis in samples scored as positive in the PTT and SSCP analysis [10]. Women belonging to families Lund 1, 3, 8, 9, 30, 44, 49, 79, 93 and 133 were included in this study. Lund 1 was analysed by linkage analysis. Details concerning mutation sites in the other families have been described in two previous articles [10, 15].

Statistics

Differences between affected and unaffected patients and constitutional parameters were analysed by a linear regression models using the statistical programme SPSS. ANOVA was used to study the separate effects on birth weight and birth length depending on whether the mother and/or daughter were mutation carriers. Mann–Whitney's *U*-test and Student's *t*-test were used for univariate analysis.

RESULTS

Birth weight in relation to *BRCA1* carrier status was studied in a univariate model. *BRCA1* gene carriers had a significantly lower birth weight compared with non-carriers ($P = 0.0044$; Figure 1). In a multivariate model with birth weight as the dependent variable, including *BRCA1* carrier status and gestational age, a lower birth weight was significantly associated with both *BRCA1* mutations ($P = 0.0041$) and lower gestational age ($P = 0.0271$; Table 2).

Birth length as a function of *BRCA1* carrier status was studied in a univariate model. *BRCA1* gene carriers were significantly shorter at birth compared with non-carriers ($P = 0.0047$; Figure 2). In a multivariate model with birth length as the dependent variable, including *BRCA1* carrier status and gestational age, a shorter length at birth was significantly associated with *BRCA1* mutations ($P = 0.0060$) after adjustment for gestational age (Table 3). *BRCA1* mutation carriers were, thus, significantly smaller for gestational age than their unaffected relatives. There was no interaction between gestational age and *BRCA1* mutation status. *BRCA1* mutation carriers did not have a significantly shorter gestational age than non-carriers ($P = 0.9671$).

As birth weight and length may have varied over time, birth weight was also studied in a multivariate model including birth date in addition to the *BRCA1* mutation and gestational

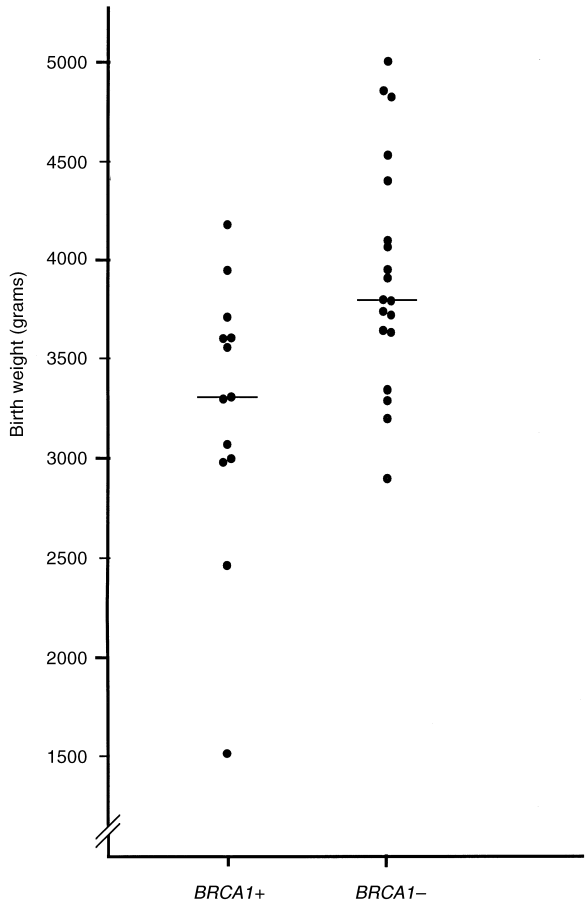

Figure 1. Birth weight as a function of *BRCA1* carrier status was studied in a univariate model. *BRCA1* gene carriers had a significantly lower birth weight compared with non-carriers ($P = 0.0044$).

Table 1. *Characteristics of the* 33 *women included in the study. Birth weight was missing for one* BRCA1*-negative woman. Birth length was missing for one* BRCA1*-positive woman, and two* BRCA1*-negative women*

	BRCA1-positive women ($n = 13$) Median (range)	*BRCA1*-negative women ($n = 20$) Median (range)
Birth weight (g)	3310 (1510–4180) ($n = 13$)	3800 (2900–5000) ($n = 19$)
Birth length (cm)	50.5 (44–58) ($n = 12$)	53 (50–59) ($n = 18$)

Table 2. *Birth weight as a function of* BRCA1 *mutation and gestational age in the* 32 *women included. Birth weight was significantly independently associated both with* BRCA1 *mutation and with gestational age.* BRCA1 *mutation was significantly negatively associated with birth weight, and gestational age significantly positively associated with birth weight. The model explains* 35.9% *of the birth weight variation*

Variable	B	SEM B	*T* value	*P* value
BRCA1	−653.928	209.632	−3.119	0.0041
Gestational age	250.135	107.473	2.327	0.0271
Constant	3823.887	141.680	26.990	0.0000

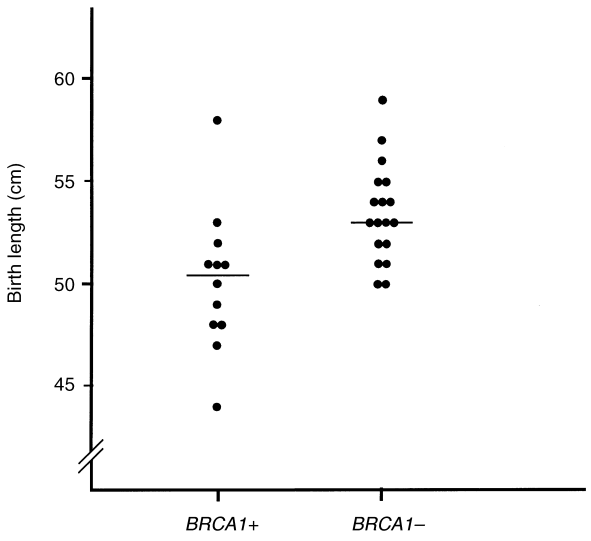

Figure 2. Birth length as a function of *BRCA1* carrier status was studied in a univariate model. *BRCA1* gene carriers were significantly shorter at birth compared with non-carriers ($P = 0.0047$).

age, but birth date did not significantly influence birth weight. The same was true for another model studying birth length in relation to *BRCA1* mutation, gestational age and birth date, with no significant effect seen for birth date on birth length.

No significant differences were seen on either present weight ($P = 0.4504$) or height ($P = 0.6337$) between the group with a mutant *BRCA1* gene and their unaffected relatives.

In studying the effect on birth weight of a woman having inherited the gene on the maternal side or not, it was found that among women whose mother was a known *BRCA1* gene carrier, women with a *BRCA1* mutation had a significantly lower birth weight than their unaffected relatives ($P = 0.0230$), after adjustment for gestational age. A non-significantly shorter length at birth was also seen in this group ($P = 0.1405$), after adjustment for gestational age. There were too few women to allow a comparison between women whose mother was not a *BRCA1* mutation carrier (Table 4).

The highest birth weight was thus found among women whose mother was *BRCA1* positive but were themselves *BRCA1* negative. The lowest birth weight was found among those whose mother was *BRCA1* negative and who were themselves *BRCA1* positive. The shortest children were born to *BRCA1*-negative mothers who themselves were *BRCA1* positive (Table 5). In non-carriers, the carrier status of the mother did not significantly influence the birth weight or the birth length of the child. Too few women inherited the mutation from the paternal side to allow statistical analysis on maternal versus paternal effects in *BRCA1* carriers.

Table 3. *Birth length as a function of* BRCA1 *mutation and gestational age in the* 30 *women included. Birth length was significantly independently associated both with* BRCA1 *mutation and with gestational age.* BRCA1 *mutation was significantly negatively associated with birth length, and gestational age significantly positively associated with birth length. The model explains* 26.7% *of the birth length variation*

Variable	B	SEM B	*T* value	*P* value
BRCA1	−3.222	1.080	−2.985	0.0060
Gestational age	0.400	0.538	0.744	0.4634
Constant	53.256	0.727	73.262	0.0000

Table 4. *Mean birth weight (g) as a function of* BRCA1 *carrier status in mother and child. The number of women in each group is indicated*

	BRCA1-positive child	*BRCA1*-negative child	*P* value
BRCA1-positive mother	3291 ($n = 11$)	4029 ($n = 7$)	0.029
BRCA1-negative mother	3005 ($n = 2$)	3882 ($n = 12$)	0.097
P value	0.612	0.604	

DISCUSSION

The main finding in this study was that female *BRCA1* mutational carriers were significantly smaller for gestational age than their unaffected relatives. This applied both to birth weight and to birth length. The birth weights and birth lengths used in this study were self-reported. As this is registered on the birth certificates in Sweden, we felt the information obtained from the questionnaires was quite reliable. One study has evaluated mother's self-report on their children's birth weight. The correlation was high between self-reported birth weight and medical records ($r = 0.98$) [16]. As mutation analyses have been performed on only very few males, we did not include them in this study.

Studies on mice indicate that the *BRCA1* gene is thought to exert its effect early in life, i.e. during fetal life and early adolescence, as well as in early pregnancy [11–14]. Mice with homozygotic mutations in the *BRCA1* analogue region do not survive fetal life, whereas heterozygotic mice do not differ phenotypically from mice with both alleles intact [12–14]. This is in contrast with our finding with a lower birth weight and a shorter birth length among heterozygotic mutation carriers. However, size differences in *BRCA1* heterozygotic mice compared with wild-type litter mates have not been carefully examined. Therefore, previous studies in mice should be cautiously interpreted. *BRCA1* expression is higher in female than in male mice entering puberty [17, 18]. The highest expression occurs in the terminal end buds in the breast epithelium [17, 18], i.e. in the structure with the highest proliferation [19]. Interesting to note is that the terminal end buds lack hormone receptors [19]. Following puberty, *BRCA1* expression decreases, and a renewed increase in *BRCA1* expression is seen during pregnancy [18]. Marquis and associates [17] found that if 17β-oestradiol and

Table 5. *Mean birth length (cm) as a function of* BRCA1 *carrier status in mother and child. The number of women is indicated*

	BRCA1-positive child	*BRCA1*-negative child	*P* value
BRCA1-positive mother	50.7 ($n = 10$)	53.3 ($n = 6$)	0.131
BRCA1-negative mother	47.5 ($n = 2$)	53.5 ($n = 12$)	0.007
P value	0.254	0.894	

progesterone were administered simultaneously to mice, this triggered a 5-fold increase in *BRCA1* mRNA, whereas if administered separately, 17β-oestradiol hardly had any effect and progesterone only had a weak effect on *BRCA1* mRNA levels. The fact that this increase in *BRCA1* is a direct effect of hormone stimulation is further supported by the finding of hormone binding regions within the *BRCA1* region [20]. New findings indicate that *BRCA1* expression may not be a direct result of the mitogenic activity of oestrogen [21]. It is, therefore, of great importance to study effects of *BRCA1* mutations not only in breast cancer patients, but also in healthy individuals during fetal life, childhood, adolescence and pregnancy.

The Swedish medical birth registry has registered birth weight in infants born since 1973. Between 1973 and 1995 the mean birth weight for singleton girls born after 37 or more weeks of gestation has varied between 3433 g (1974) and 3533 g (1995). The women included in this study were born between 1936 and 1971. *BRCA1* carriers had a mean birth weight of 3247 g and non-carriers a mean birth weight of 3936 g. As the general nutrition status has improved over recent years, the mean birth weight for girls was probably somewhat lower during the years of 1936–1971. No significant effect from birth date on birth weight was seen when this factor was entered into the multivariate model in our study sample, while the effect from *BRCA1* mutation remained significantly negatively associated with birth weight. The fact that the women in this study were born over a number of years may indicate that different birth cohorts did not affect the results. The majority of newborns have a birth weight between 3000 and 3999 g (1988). *BRCA1* carriers tend to have a birth weight towards the lower end of the normal curve, whereas non-carriers born in these families tend to have a birth weight towards the higher end of the normal curve. What is the reason for this difference? The fact that the *BRCA1* gene plays an important role during growth *in utero* could possibly explain why *BRCA1* mutation carriers were small for gestational age, but does not explain why the non-carriers were big. There were too few women who had inherited the gene from the paternal side to be able to study any additional effects on birth weight and birth length due to conditions *in utero* in women whose mother also carried the defect gene. Currently, there are no data available on the effect of maternal versus paternal carriership on the fetus.

As there was no significant difference in present weight and height between *BRCA1* mutation carriers and non-carriers, it can be concluded that there is a 'catch-up' growth among *BRCA1* mutation carriers between infant life and adult life. Whether this 'catch-up' growth is mediated via hormones and growth factors, such as insulin-like growth factor-1, is not yet known. We are presently collecting plasma from young women belonging to *BRCA1* mutation families to see if there are any differences in hormone or growth factor levels between carriers and non-carriers that can explain the 'catch-up' effect.

In conclusion, we found that *BRCA1* carriers had a significantly lower birth weight, were significantly shorter at birth, and thus smaller for gestational age than their unaffected relatives.

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