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## **BODY WEIGHT AND RISK OF BREAST CANCER IN BRCA1/2 MUTATION CARRIERS**

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## ABSTRACT

**Obesity is an established risk factor for postmenopausal breast cancer in the general population. However, it is still unclear whether this association also exists in BRCA1/2 mutation carriers. We investigated the association between self-reported anthropometric measures and breast cancer risk in a nationwide retrospective cohort study, including 719 BRCA1/2 mutation carriers, of whom 218 had been diagnosed with breast cancer within 10 years prior to questionnaire completion. All time-varying Cox proportional hazards analyses were stratified by menopausal status. For premenopausal breast cancer, no statistically significant associations were observed for any of the anthropometric measures. The association between Body Mass Index (BMI) at age 18 and premenopausal breast cancer risk suggested a trend of decreasing risk with increasing BMI (HR<sub>22.50-24.99 vs 18.50-22.49 kg/m<sup>2</sup></sub> = 0.83, 95%CI=0.47-1.44 and HR<sub>≥25.00 vs 18.50-22.49 kg/m<sup>2</sup></sub> = 0.41, 95%CI=0.13-1.27). For postmenopausal breast cancer, being taller than 1.66 m increased the risk 1.7-fold (HR=1.67, 95%CI=1.01-2.74) when compared to a height ≤1.67 m. Compared with a current body weight <72 kg, a current body weight of ≥72 kg increased the risk of postmenopausal breast cancer 2.1-fold (95%CI=1.23-3.59). A current BMI of ≥25.0 kg/m<sup>2</sup>, an adult weight gain of 5 kg or more, and a relative adult weight gain of 20% or more were all statistically non-significantly associated with a 50-60% increased risk of postmenopausal breast cancer (HR=1.46 (0.86-2.51), HR=1.56 (95%CI=0.85-2.87), and HR=1.60 (95%CI=0.97-2.63), respectively), as compared to having a healthy or stable weight. No associations for body weight or BMI at age 18 with postmenopausal breast cancer risk were observed. In conclusion, menopausal status seemed to modify the association between body weight and breast cancer risk among BRCA1/2 mutation carriers. We observed no clear association between body weight and premenopausal breast cancer, while overweight and weight gain increased postmenopausal breast cancer risk. Carriers may reduce their risk of postmenopausal breast cancer by maintaining a healthy body weight throughout life.**

## INTRODUCTION

The estimated lifetime risk of developing breast cancer for BRCA1/2 mutation carriers varies between 40 and 80%<sup>1-7</sup>. Reasons for variation may include different mutations in the same gene (allelic variation)<sup>8-10</sup>, the effect of modifying genes<sup>11-13</sup>, and non-genetic modifiers<sup>14</sup>. Several studies have indicated that the penetrance of BRCA1 and BRCA2 mutations has increased in recent generations<sup>4,6,15</sup>, which supports the concept that non-genetic risk factors, of which the prevalence has increased, also affect the risk. Overweight might be such a risk factor, because its prevalence has gradually increased over the last decades<sup>16</sup>. Moreover, in the general population overweight and obesity or, more specifically, adult weight gain are established risk factors for postmenopausal breast cancer<sup>17,18</sup>. By contrast, overweight and obesity may reduce the risk of premenopausal breast cancer<sup>17,18</sup>. Few studies examined the effects of body weight and/or weight change on breast cancer risk in BRCA1/2 mutation carriers. Three relatively small studies showed inconsistent results<sup>6,19,20</sup>. A large study by Kotsopoulos et al. observed no association between body weight and breast cancer risk among BRCA1/2 mutation carriers and was the only study examining menopausal status as a potential effect modifier<sup>21</sup>. The aim of the present study was to assess whether different anthropometric measures, i.e. height, body weight, Body Mass Index (BMI) and body weight change throughout life, affected the risk of pre- and postmenopausal breast cancer in a large population of BRCA1/2 mutation carriers, while adjusting for physical activity.

## METHODS

### STUDY POPULATION

The present study was conducted within the framework of the HEBON study, of which the design was described earlier<sup>22</sup>. In brief, the HEBON study is an ongoing nationwide retrospective cohort study with prospective follow-up among members of BRCA1/2 families in the Netherlands. The study population for the present study consisted of all female BRCA1/2 mutation carriers who had participated in the HEBON study in the period January 1999 through August 2007 (N=1,120; response 80%). We excluded seven carriers of whom the age at end of follow-up was missing. BRCA1/2 mutation carriers of whom at least 50% of the information on anthropometric measures was missing were excluded (N=133). The study population for the analyses consisted of 980 carriers, among whom 38 (4%) obligate carriers (not tested themselves but considered as carrier because there was at least one proven carrier among their children, while inheritance was not paternal). Five percent (N=49; response 76%) of the questionnaires was completed by a proxy, because the woman herself had died. This was equivalent to 76% of questionnaires completed by a proxy among obligate carriers (N=29/38).

### ANALYTIC COHORT

Previous studies showed that excess body weight (defined as overweight (BMI between 25.0 to 29.9 kg/m<sup>2</sup>) or obesity (BMI≥30.0 kg/m<sup>2</sup>)) decreases overall survival<sup>17,23-25</sup> and

breast cancer-specific survival<sup>16,26-28</sup>. The association between obesity and poor prognosis of breast cancer was present in both pre- and postmenopausal carriers. To reduce (potential) survival bias, we restricted the analysis to person-years within 10 years prior to questionnaire completion<sup>22,29-31</sup>. We excluded cases who died a long time ago whose prognosis might have been influenced by BMI. In total, 218 cases were diagnosed with breast cancer within the 10-year period in 4,992 person-years.

### ASSESSMENT OF ANTHROPOMETRIC MEASURES

Carriers were asked to report their height (m), exact body weight at age 18 (kg) and exact body weight (kg) at the time of questionnaire completion. Cases were also asked to report their exact body weight in the year prior to their breast cancer diagnosis. Additionally, for different age periods, carriers were asked to complete a grid with body weight in 5 kg categories (i.e. <55, 55-59, 60-64, 65-69, 70-74, 75-79, 80-84, 85-89, 90-94, >94 kg) from age 18 years onwards in 10-year age-periods (i.e. 20-29, 30-39, 40-49, 50-59, 60-69, ≥70 years), excluding periods of pregnancy. Body weight at age 18 and height were used to calculate BMI at age 18. Height, body weight at age 18 and BMI at age 18 were analyzed as fixed variables. Current body weight was calculated for each year (i.e. from age 18 until time of diagnosis/censoring) with the use of the completed grid and where an exact value of body weight was available this value was used (i.e. body weight at diagnosis (N=218 cases) and body weight at questionnaire completion for unaffected carriers who were censored at time of questionnaire completion (N=29)). Current body weight and BMI, adult weight change (calculated as the difference between the age-specific body weight and at age 18), and relative adult weight change (calculated as the adult weight change divided by body weight at age 18), changed over time and were therefore determined for each age (year) of observation and included as time-varying variables.

### STATISTICAL ANALYSIS

The adjusted hazard ratios (HRs) as estimates of relative risk and 95% confidence intervals (95%CI) were obtained using a time-varying multivariate Cox proportional hazards model with age (in years) as time scale. Follow-up started at 10 years prior to questionnaire completion and ended at date of first breast cancer diagnosis, date of risk reducing mastectomy, date of linkage with Netherlands Pathology Database (PALGA) and the Netherlands Cancer Registry (NCR), date of completing the questionnaire if no informed consent for linkage was given, or date of death, whichever occurred first. All analyses were adjusted for age at the start of follow-up, intrinsically stratified for birth cohort (≤1945, 1946-1955, 1956-1964, ≥1965) and gene (BRCA1 and BRCA2) and clustered on family to correct for potential within family correlations in risk factors. Because menopausal status has been shown to modify the association between overweight and breast cancer risk in the general population<sup>16</sup>, all analyses were stratified according to menopausal status. Carriers were considered postmenopausal 12 months after their last menstrual period (age of menopause). Forty-two percent of carriers in the analytic cohort (299/719) were postmenopausal at the end of follow-up.

For these 299 women, age at menopause was the censoring event within the premenopausal cohort, while it was the starting age within the postmenopausal cohort. Additionally, for the premenopausal cohort all analyses were weighted according to the weighted cohort approach to correct for potential testing bias<sup>32</sup>. Unfortunately, for the postmenopausal cohort it was not possible to conduct weighted analyses as the power was too low to create stable cohort-specific weights.

For the premenopausal cohort (N=609 among which 155 cases in 3,013 person years), height, body weight at age 18, current body weight, adult weight change, and relative adult weight change were categorized based on the distribution of the total cohort at the end of follow-up. For the postmenopausal cohort (N=299 among which 63 cases in 1,979 person years), dichotomized variables based on the median values were created because of the smaller numbers. For both cohorts, BMI at age 18 and current BMI were categorized according to the World Health Organization<sup>33</sup>.

For the multivariate models, stepwise forward confounder selection was performed, evaluating the effect of adding one confounder at a time, based on a more than 10% change in (at least one of) the HRs of the main exposure variables under investigation, i.e. current BMI and adult weight change. BRCA1/2 mutation carriers with a healthy and/or stable weight were considered as the reference group. Confounders (categorized based on the distribution of the entire cohort (N=980) at the end of follow-up) were: lifetime sports activity (never sports activity, <11.0, 11.0-22.7, >22.7 mean MET-hours/week; time-varying) for the premenopausal cohort, and parity (nulliparous, 1-2 children, >2 children), type of menopause and use of hormonal replacement therapy (HRT) (natural menopause and never HRT use; natural menopause and ever HRT use; bilateral prophylactic (salpingo) oophorectomy (BPSO) and never HRT use; BPSO and ever HRT use; surgical (ovarian cancer) and never HRT use), and lifetime sports activity (never sports activity, <11.0, 11.0-22.7, >22.7 mean MET-hours/week; time-varying) for the postmenopausal cohort. Age at menarche, oral contraceptive use, age at first full term pregnancy, breast-feeding, smoking, alcohol consumption, and family history did not change the HRs by more than 10% and were omitted from our final models. No violation of the proportional hazards assumption by any of the confounding variables or by current BMI and adult weight change was observed.

Two-sided p-values ≤0.05 were considered statistically significant. All analyses were performed using STATA/SE 10.0 (StataCorp LP).

## RESULTS

The age (mean ± standard deviation) at end of follow-up in the premenopausal and postmenopausal cohort was 38.9 ± 7.9 years and 54.9 ± 11.4 years, respectively. In both cohorts, cases were older than noncases at end of follow-up (premenopausal: 44.0 ± 8.1 and 41.5 ± 9.9 years, respectively, P=0.002; postmenopausal: 61.1 ± 8.7 and 53.4 ± 11.8 years, respectively, P<0.001). In general, the characteristics of the premenopausal cases were reasonably similar to the noncases (Table 1). However, in the postmenopausal cohort there were some differences, for example, cases were born earlier, more often

**TABLE 1. Characteristics of the study population (N=719) by menopausal status**

Characteristic	Premenopausal cohort (N=609)				Postmenopausal cohort (N=299)			
	Cohort		Cases		Cohort		Cases	
	N <sup>a</sup>	%	N <sup>a</sup>	%	N <sup>a</sup>	%	N <sup>a</sup>	%
<b>Gene</b>								
BRCA1	468	77%	120	77%	223	76%	50	79%
BRCA2	141	23%	35	23%	76	24%	13	21%
<b>Proxy data</b>								
No	601	99%	151	97%	289	97%	61	97%
Yes	8	1%	4	3%	10	3%	2	3%
<b>Birth cohort</b>								
≤1945	26	4%	3	2%	109	36%	42	67%
1946-1955	137	23%	45	29%	96	32%	17	27%
1956-1964	207	34%	65	42%	68	23%	3	5%
≥1965	239	39%	42	27%	26	9%	1	1%
<b>Age at end of follow-up</b>								
≤34 years	185	30%	49	32%	5	2%	1	2%
35-40 years	167	27%	41	26%	16	5%	1	2%
41-49 years	186	31%	52	34%	82	27%	7	11%
≥50 years	71	12%	13	8%	196	66%	54	86%
<b>Lifetime sports activity</b>								
Never	218	36%	53	34%	138	46%	35	55%
<11.0 MET-hours/week	195	32%	43	28%	72	24%	8	13%
11.0-22.7 MET-hours/week	175	29%	52	34%	66	22%	12	19%
≥22.7 MET-hours/week	21	3%	7	4%	23	8%	8	13%
<b>Family history</b>								
No	269	45%	63	41%	120	41%	15	25%
Yes	324	55%	91	59%	171	59%	46	75%
<b>Age at menarche</b>								
≤12 years	207	34%	56	36%	83	28%	21	33%
13 years	152	25%	36	24%	75	25%	13	21%
≥14 years	244	41%	62	40%	139	47%	29	46%
<b>Parity</b>								
Nulliparous	185	30%	43	28%	49	16%	7	11%
Parous	424	70%	112	72%	250	84%	56	89%
<b>Number of children</b>								
1-2 children	313	74%	88	79%	153	61%	41	73%
>2 children	111	26%	24	21%	97	39%	15	27%
<b>Age at first full term pregnancy</b>								
≤22 years	74	17%	22	20%	66	26%	13	23%
23-25 years	87	21%	24	21%	67	27%	15	27%
26-27 years	67	16%	15	13%	48	19%	15	27%
≥28 years	196	46%	51	46%	69	28%	13	23%
<b>Breastfeeding</b>								
Never	106	25%	29	26%	62	25%	14	25%
Ever	317	75%	82	74%	188	75%	42	75%
<b>Oral contraceptive use</b>								
Never	39	6%	10	7%	58	19%	13	21%
Ever	569	94%	144	93%	241	81%	50	79%

Type of menopause and HRT use	NA		NA		NA		NA	
	N	%	N	%	N	%	N	%
Natural – never HRT use	106	35%	42	67%				
Natural – ever HRT use	23	8%	5	8%				
Surgical, prophylactic – never HRT use	83	28%	5	8%				
Surgical, prophylactic – ever HRT use	60	20%	9	14%				
Surgical, ovarian cancer – never HRT use	27	9%	2	3%				
<b>Alcohol consumption</b>								
Never	235	39%	69	45%	116	39%	21	33%
Ever	374	61%	86	55%	183	61%	42	67%
<b>Smoking</b>								
Never	270	44%	74	48%	132	44%	28	45%
Ever	339	56%	81	52%	166	56%	34	55%

NA, not applicable

a Numbers do not always add up to 100% due to missing values; number of children, age at first full term pregnancy and breastfeeding apply to parous carriers only (100%); type of menopause and HRT use applies to postmenopausal carriers only (100%)

had a positive family history, and had less children than noncases. Forty-three percent of postmenopausal carriers had experienced a natural menopause, while 57% had a menopause that was surgically induced (48% and 9% by BPSO and ovarian cancer diagnosis, respectively). Furthermore, cases had more often experienced a natural menopause (75%) than noncases (35%). Twenty percent of postmenopausal carriers had ever used HRT.

The median body weight at age 18 was 58 kg for both cohorts. Because the median height was somewhat larger for premenopausal carriers than for postmenopausal carriers (median: 1.69 m and 1.65 m, respectively; data not shown), the median BMI at age 18 was slightly lower among premenopausal carriers than in postmenopausal carriers (20.4 kg/m<sup>2</sup> versus 21.2 kg/m<sup>2</sup>; data not shown). The median current body weight and adult weight change were also lower in the premenopausal cohort than in the postmenopausal cohort (65 kg and 5 kg weight gain versus 70 kg and 10 kg weight gain, respectively; data not shown). Approximately one-third of the premenopausal carriers and half of the postmenopausal carriers were overweight (≥25.00 kg/m<sup>2</sup>) at the end of follow-up.

For premenopausal breast cancer, no statistically significant associations were observed for any of the anthropometric measures (Table 2). The association between BMI at age 18 and premenopausal breast cancer risk suggested a trend of decreasing risk with increasing BMI (HR=0.83, 95%CI=0.47-1.44 and HR=0.41, 95%CI=0.13-1.27 for 22.50-24.99 kg/m<sup>2</sup> and ≥25.00 kg/m<sup>2</sup> when compared to 18.50-22.49 kg/m<sup>2</sup>, respectively). When compared to a current body weight of 58-62 kg, both a low current weight (HR<sub>≤57 kg</sub>=1.52, 95%CI=0.84-2.78) and a high current weight (HR<sub>≥68 kg</sub>=1.35, 95%CI=0.74-2.47) were non-significantly associated with an increased premenopausal breast cancer risk. A trend of decreasing premenopausal breast cancer risk with increasing current BMI was suggested (HR=0.87, 95%CI=0.53-1.42 and HR=0.75, 95%CI=0.43-1.31 for 22.50-

**TABLE 2. Anthropometric measures and the risk of premenopausal breast cancer**

	Person-years	Cases	Multivariate	
			HR (95% CI) <sup>a</sup> unweighted	HR (95% CI) <sup>a</sup> weighted
<b>Height (m)</b>				
≤1.64	537	32	0.84 (0.53-1.36)	0.73 (0.39-1.38)
1.65-1.68 <sup>c</sup>	796	43	1.00	1.00
1.69-1.72	690	36	1.20 (0.76-1.89)	1.36 (0.75-2.47)
≥1.73	990	44	0.99 (0.76-1.89)	0.89 (0.46-1.71)
<b>Body weight at age 18 (kg)</b>				
≤53	500	26	1.16 (0.70-1.93)	1.02 (0.53-1.97)
54-57 <sup>c</sup>	688	39	1.00	1.00
57-60	791	46	1.26 (0.79-2.02)	1.01 (0.52-1.95)
≥61	1,034	44	1.04 (0.64-1.70)	0.94 (0.50-1.78)
<b>BMI at age 18 (kg/m<sup>2</sup>)</b>				
≤18.49	544	28	0.84 (0.55-1.28)	0.72 (0.41-1.27)
18.50-22.49 <sup>c</sup>	1,785	101	1.00	1.00
22.50-24.99	421	19	0.89 (0.57-1.38)	0.83 (0.47-1.44)
≥25.00	263	7	0.73 (0.31-1.75)	0.41 (0.13-1.27)
<b>Current body weight (kg)</b>				
≤57	554	42	1.42 (0.88-2.30)	1.52 (0.84-2.78)
58-62 <sup>c</sup>	592	24	1.00	1.00
63-67	521	25	0.88 (0.52-1.51)	0.92 (0.48-1.76)
≥68	1,346	64	1.40 (0.87-2.25)	1.35 (0.74-2.47)
<b>Current BMI (kg/m<sup>2</sup>)</b>				
≤18.49	83	3	0.47 (0.14-1.60)	0.41 (0.09-1.85)
18.50-22.49 <sup>c</sup>	1,334	63	1.00	1.00
22.50-24.99	786	48	0.98 (0.68-1.44)	0.87 (0.53-1.42)
≥25.00	810	41	1.05 (0.68-1.61)	0.75 (0.43-1.31)
<b>Adult weight change (kg)</b>				
≥5 kg loss	170	9	1.00 (0.51-1.95)	1.03 (0.45-2.37)
<5 kg loss and <5 kg gain <sup>c</sup>	1,091	48	1.00	1.00
≥5 kg and <10 kg gain	637	28	0.62 (0.38-1.01)	0.67 (0.37-1.21)
≥10 kg and <15 kg gain	512	33	1.29 (0.78-2.12)	1.02 (0.56-1.86)
≥15 kg gain	603	37	0.96 (0.59-1.56)	0.77 (0.41-1.45)
<b>Relative weight change</b>				
≤3.9%	869	38	0.99 (0.61-1.60)	0.85 (0.47-1.53)
4% and <13% <sup>c</sup>	799	39	1.00	1.00
13% and <25%	725	39	1.01 (0.65-1.57)	0.75 (0.42-1.34)
≥25%	620	39	1.10 (0.71-1.72)	0.85 (0.48-1.51)

HR, hazard ratio; CI, confidence interval

a A time-varying Cox proportional hazards model, stratified for genes (BRCA1 and BRCA2) and birth cohort (≤1945, 1946-1955, 1956-1964, ≥1965), clustered on family (326 clusters), and adjusted for lifetime sports activity (mean MET-hours/week in active period; time-varying)

b Weighted cohort approach

c Reference category

24.99 kg/m<sup>2</sup> and ≥25.00 kg/m<sup>2</sup> when compared to 18.50-22.49 kg/m<sup>2</sup>, respectively). We found no association for adult weight change and relative weight change with risk of premenopausal breast cancer. Analyses restricted to BRCA1 carriers showed very similar results compared to all carriers combined (data not shown).

For the postmenopausal cohort it was not possible to conduct weighted analyses as the power was too low to create stable cohort-specific weights. Therefore, the presented unweighted estimates (Table 3) might be slightly biased towards unity. Being taller than 1.66 m increased the risk of postmenopausal breast cancer 1.7-fold (HR=1.67, 95%CI=1.01-2.74) when compared to a height ≤1.67 m. We observed no associations for body weight or BMI at age 18 with postmenopausal breast cancer risk. When compared to a current body weight below 72 kg, weighing 72 kg or more increased the risk of postmenopausal breast cancer 2.1-fold (HR=2.10, 95%CI=1.23-3.59). A current BMI of ≥25.00 kg/m<sup>2</sup>, an adult weight gain of 5 kg or more, and a relative adult weight gain of 20% or more were all statistically non-significantly associated with a 1.5- to 1.6-fold increased risk of postmenopausal breast cancer (HR=1.46 (0.86-2.51), HR=1.56 (95%CI=0.85-2.87), and HR=1.60 (95%CI=0.97-2.63), respectively) as compared to having a healthy or stable body weight.

**TABLE 3. Anthropometric measures and the risk of postmenopausal breast cancer**

	Person-years	Cases	Multivariate	
			HR (95% CI) <sup>a</sup> unweighted	
<b>Height (m)</b>				
<1.67 <sup>b</sup>	1,589	35	1.00	
≥1.67	1,333	28	1.67 (1.01-2.74)	
<b>Body weight at age 18 (kg)</b>				
<58 <sup>b</sup>	1,146	20	1.00	
≥58	1,776	43	1.18 (0.62-2.23)	
<b>BMI at age 18 (kg/m<sup>2</sup>)</b>				
<22.50 <sup>b</sup>	2,157	42	1.00	
≥22.50	765	21	0.94 (0.37-2.39)	
<b>Current body weight (kg)</b>				
<72 <sup>b</sup>	1,764	29	1.00	
≥72	1,158	34	2.10 (1.23-3.59)	
<b>Current BMI (kg/m<sup>2</sup>)</b>				
<25.00 <sup>b</sup>	1,608	27	1.00	
≥25.00	1,314	36	1.46 (0.86-2.51)	
<b>Adult weight change (kg)</b>				
<5 kg gain <sup>b</sup>	695	14	1.00	
≥5 kg gain	2,227	49	1.56 (0.85-2.87)	
<b>Relative adult weight change</b>				
<20% <sup>b</sup>	1,520	31	1.00	
≥20%	1,402	32	1.60 (0.97-2.63)	

HR, hazard ratio; CI, confidence interval

a A time-varying Cox proportional hazards model, stratified for genes (BRCA1 and BRCA2) and birth cohort (≤1945, 1946-1955, 1956-1964, ≥1965), clustered on family (185 clusters), and adjusted for parity (nulliparous, 1-2 children, >2 children), type of menopause and HRT use (natural menopause – never HRT use, natural menopause – ever HRT use, BPSO – never HRT use, BPSO – ever HRT use, surgical (ovarian cancer) – never HRT use) and lifetime sports activity (mean MET-hours/week in active period; time-varying)

b Reference category

## DISCUSSION

The results of our study on the association of body weight and weight change on breast cancer risk among BRCA1/2 mutation carriers are generally in line with the literature based on the general population. For premenopausal breast cancer no statistically significant associations were observed with any of the anthropometric measures. If any, we observed a decreasing risk of premenopausal breast cancer with increasing BMI at age 18. Among postmenopausal women, we observed that height, current overweight, and increased relative weight change were all associated with an increased postmenopausal breast cancer risk. In the present study, all observed associations were independent of the effect of physical activity.

So far, few studies examined the effects of anthropometric measures on breast cancer risk in BRCA1/2 mutation carriers, with inconsistent results. The largest study on the association between body weight and breast cancer risk in carriers was a study among 1,073 case-control pairs by Kotsopoulos et al.<sup>21</sup>. They focused on changes in body weight and observed that a loss of at least 10 pounds between ages 18 to 30 years was associated with a decreased risk of breast cancer at ages 30–40. We did not observe an association between adult weight loss and risk of premenopausal breast cancer, but numbers in our weight loss category were small. The study of Kotsopoulos et al. was intrinsically stratified on menopausal status of the cases, but it is not quite clear how the menopausal status of the controls was taken into account. The authors reported that no effect modification of menopausal status was present. The other studies in BRCA1/2 mutation carriers did not adjust for or stratify on menopausal status, which hampers the comparison with our results. In a case-only study among 104 carriers, a healthy body weight at menarche and a lighter body weight at age 21 were associated with a significant delay in the age at onset of breast cancer; however, it is not clear whether these findings were adjusted for birth cohort<sup>6</sup>. Similar to our observation of increased postmenopausal breast cancer risk after adult weight gain, Nkondjock et al. observed a trend of increased breast cancer risk with increasing weight gain since age 18 and age 30<sup>20</sup>. This effect was independent of physical activity and energy intake. Chang-Claude et al. observed no association between BMI and breast cancer risk in carriers<sup>19</sup>. The two last studies were relatively small and body weight might not always apply to the prediagnostic period for cases and similar age ranges for the unaffecteds. For women diagnosed with breast cancer it has been shown that they frequently gain body weight after diagnosis<sup>34</sup>.

The present study has some strong and weak points that should be considered in the interpretation of the results. The primary strengths of our study include the large sample size, the detailed lifetime information on various anthropometric measures, stratification by menopausal status, adjustment for lifetime physical activity (time-varying) and other confounders, and the possibility to cluster on family. In addition, we used the weighted cohort approach for the premenopausal cohort<sup>32</sup>. However, the retrospective character of the present study, the type of study population, consisting of carriers tested in the clinical setting, and the lack of weighting in the postmenopausal

cohort may have caused some biases in our results, which are discussed below. The association between obesity and poor overall survival<sup>17,23–25</sup> or prognosis of breast cancer<sup>16,26–28</sup>, might, if also true for BRCA1/2 mutation carriers, have influenced our study results. The inclusion of prevalent cases, may have led towards bias to the null, overweight/obese prevalent cases may have been underrepresented because they had died prior to study entry<sup>23,26,27</sup>. We reduced this potential survival bias by restricting the analyses to person-years within 10 years prior to questionnaire completion. In general, the difference in HRs between the entire cohort, starting follow-up at birth, and the analytic cohort indeed suggested that survival bias might be present in our entire cohort (e.g. the HR for adult weight change and the risk of postmenopausal breast cancer in the entire cohort was 1.33 (95%CI=0.80–2.22; data not shown) and 1.56 (95%CI=0.85–2.87) in the analytic cohort). Although the effect was small, we cannot exclude the presence of some survival bias in our analytic cohort, because overweight/obese BRCA1/2 mutation carriers with early-onset breast cancer and a poor prognosis may not have survived 10 years to participate in our study. Furthermore, we had a priori tried to reduce potential survival bias by including proxy data for obligate carriers who had died before study entry. However, a large proportion (67%) of these questionnaires had to be excluded because at least 50% of information on anthropometric measures was missing. A second limitation is the use of self-reported anthropometric data. Although self-reported and objective weights are correlated<sup>18</sup>, studies on the validity of self-reported anthropometric measures show consistent underreporting of self-reported body weight and overreporting of height, especially among overweight and obese individuals<sup>18,35</sup>. We are not aware of studies that examined potential differential misclassification according to breast cancer case-control status. However, if we assume that misclassification is non-differential, the systematic underreporting of obesity might have resulted in an overestimated risk for a specific category.

In the general population, the increased postmenopausal breast cancer risk observed in obese women is generally explained by the higher rates of conversion of androgenic precursors to estradiol through increased aromatase enzyme activity in adipose tissue<sup>17</sup>. The reduced risk of breast cancer after a BPSO in BRCA1/2 mutation carriers<sup>36</sup> suggests that hormonal influences are important in carriers, despite the fact that the majority of BRCA1 associated breast cancers have negative estrogen receptor status<sup>37</sup>. In vitro studies indicate that estrogens may play a role in BRCA1-related carcinogenesis<sup>38</sup> and suggest that BRCA1 may function as part of a feedback mechanism to regulate estrogen signaling<sup>39</sup>. Subgroup analysis of the association between anthropometric measures and breast cancer risk among premenopausal BRCA1 carriers showed similar results as in the total analytic premenopausal cohort. BRCA2-associated breast tumours tend to be similar to sporadic cases. A subgroup analysis among BRCA2 carriers was not possible due to lack of power.

The results of our study on the association of height on breast cancer risk among BRCA1/2 mutation carriers are generally in line with the literature based on the general population, where height is an independent factor that has been shown to have a modest contribution to the development of postmenopausal breast cancer<sup>40,41</sup>, whereas

in premenopausal women the association is less clear and not significant<sup>41</sup>. Potential biological mechanisms include amongst others childhood energy intake plus related growth hormone release and increased levels of insulin-like growth factor<sup>42-44</sup>, childhood physical activity<sup>45</sup>, and number of ductal stem cells that develop in the breast in utero<sup>46</sup>. Previous studies in the general population showed that HRT use interacts with obesity in the development of postmenopausal breast cancer<sup>41,47,48</sup>, probably by sharing hormonal carcinogenic pathways. However, in the Netherlands HRT has not been widely used by postmenopausal carriers (see Table 1 and <sup>49</sup>). The results of multivariate analyses restricted to never users were not markedly different, e.g. the HRs for current body weight and the risk of postmenopausal breast cancer in the total analytic postmenopausal cohort (HR=2.10, 95%CI=1.23–3.59) and the cohort of never users (HR=2.05, 95%CI=1.10–3.82; data not shown) were similar. In the present study, we adjusted our analysis for HRT use as it proved to be a confounder, but the power was too low to test whether interaction was present.

In conclusion, the results of the present study indicate that there was no clear association between any of the anthropometric measures and premenopausal breast cancer risk in BRCA1/2 mutation carriers. For postmenopausal breast cancer risk, we observed associations similar to what is known for the general population, i.e. overweight and adult weight gain increased the risk of postmenopausal breast cancer in BRCA1/2 mutation carriers. Body weight is one of the few non-genetic modifiers for breast cancer. Carriers may reduce postmenopausal breast cancer risk by maintaining a healthy body weight throughout life. Our findings require confirmation by future studies focusing on prospective follow-up in larger sample sizes. Future research should also focus on the potential interaction between body weight, physical activity and lifestyle/behavioural determinants of adult weight change for BRCA1/2 mutation carriers.

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## APPENDIX

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