



Published in final edited form as:

*Cancer Epidemiol Biomarkers Prev.* 2013 April ; 22(4): 670–674. doi:10.1158/1055-9965.EPI-12-1133.

## Dairy Intakes in Older Girls and Risk of Benign Breast Disease in Young Women

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### Abstract

Previous investigations found high dairy intakes in girls associated with rapid height growth and excess weight gain, which had opposite relationships with benign breast disease (BBD) in young women. We use data from the longitudinal Growing Up Today Study (GUTS) to investigate whether dairy intakes, in older children/adolescents, are associated with BBD risk in young women. GUTS includes 9039 females, aged 9–15yrs in 1996, who completed questionnaires annually through 2001, then in 2003, 2005, 2007, and 2010. Dietary food frequencies (1996–2001) obtained milk, yogurt, and cheese intakes. On 2005–2010 surveys, 7011 females (18–29yrs) reported whether a health care provider ever diagnosed them with BBD (n=250), and if confirmed by breast biopsy (n=105). Logistic regression models estimated associations between prevalent biopsy-confirmed BBD and dairy intakes, adjusted for age and energy. Multivariable-adjusted models additionally included menarche age, childhood adiposity, adolescent alcohol consumption, and pregnancy. Further analyses stratified by family history. Age-energy-adjusted models of dairy (milk, yogurt, cheese, total dairy servings, dairy protein, dairy fat) intakes at 14yr found no significant associations with BBD risk (milk OR=0.90/(serving/day), 95%CI:0.76–1.05; dairy protein OR=0.98/(10gm/day), 95%CI:0.82–1.17). Separate analyses of dairy intakes at 10yr, intakes before the growth spurt, during the growth spurt, before menses-onset, and after menses-onset provided no significant associations with BBD. Multivariable-adjustment, and family history stratification, did not alter the above findings. We conclude that dairy intakes by older girls have no strong relation with BBD risk in young women. Due to small number of cases, it is important to continue follow-up and re-examine later.

### Keywords

adolescent; pre-teen; milk; cheese; yogurt; dairy protein; dairy fat; height growth spurt; BBD; breast cancer; prospective cohort; longitudinal

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**Conflict of Interest:** The authors have no conflicts, including financial, to disclose.

## Introduction

Substantial evidence points to the period in life before a woman's initial pregnancy, when mammary gland cells are undergoing rapid proliferation, as a critical time for carcinogenic exposures that may increase her lifetime risk for breast cancer (1). Human studies indicate childhood and adolescent exposures can be more important than adult exposures in breast cancer development (2–4). Because benign breast disease (BBD) is a well-established risk factor for breast cancer (5), the investigation of exposures in older girls and their subsequent development of BBD may provide insight into the etiology of breast cancer and present possible new strategies for prevention.

Previously we reported that high intakes of certain dairy products (milk, yogurt, dairy protein) were associated with more rapid height growth spurts (peak height velocity, PHV) (6), and a subsequent study on the same cohort found that higher PHV's were associated with increased risk for BBD in young women (7). In other analyses, high dairy intakes promoted excess weight gain (8), but heavier children had lower BBD risk (7). Taken together, these investigations infer associations, between dairy intakes and BBD, in opposite directions.

We investigate whether childhood dairy intakes are independently associated with BBD in young women, using data from a prospective cohort study, initiated in 1996, of 9–15yr old females followed to 29yr.

## Methods

### Study Population

The Growing Up Today Study (GUTS; founding PI, Dr. Colditz) includes 9039 girls from all 50 states who are daughters of Nurses' Health Study II participants (9). The study, approved by Institutional Review Boards at Harvard School of Public Health and Brigham and Women's Hospital, is described elsewhere (10). Mothers provided informed consent, and their 9–15yo daughters assented by completing baseline questionnaires. The cohort returned questionnaires annually (by mail or Internet) from 1996 through 2001, then in 2003, 2005, 2007, and 2010. The girls' response rate to one or more follow-ups after baseline was 97%. Over 78% (n=7053) returned at least one of the 2005 through 2010 (up to September, 2011) surveys inquiring about BBD.

### Benign Breast Disease

The 2005, 2007 and 2010 surveys asked "Has a health care provider ever diagnosed you as having Benign Breast Disease?" and, if yes, whether it had been "Confirmed by breast biopsy". A total of 7011 females (aged 18–29yr) reported whether a health care provider ever diagnosed them with BBD (n=250), and if any diagnosis had been confirmed by breast biopsy (n=105). After excluding six girls whose mothers reported childhood cancer in their daughters, those 6755 females who responded they had never been diagnosed with BBD provide the non-cases for analyses.

Most BBD cases were likely diagnosed because participants (or their physicians) found a clinically palpable mass which was then biopsied, as participants were too young to be undergoing routine screening mammography. The most common type of BBD occurring in adolescents and young women is fibroadenoma, which accounts for nearly 70% of benign breast lesions (11). The remaining types are primarily cysts and fibrocystic changes (11). A validation study conducted in a large cohort of women confirmed the accuracy of women's self-reports of biopsy-confirmed BBD (12).

## Dairy Intakes

Our self-administered semi-quantitative food frequency questionnaire (FFQ), designed specifically for older children and adolescents, has good validity and reproducibility for children ages 9–18yr (13). The mean correlation for nutrients from the FFQ compared to three 24-hour recalls was  $r=0.54$ , comparable to the performance of a similar FFQ for adults. Milk and dairy products, including yogurt and cheese, had particularly high validity among adult women (14). Another youth FFQ, similar to ours, provided estimates of milk and dairy food consumption by adolescent girls that correlated well with 7-day dietary records (15).

Surveys from 1996 through 2001 annually inquired about usual frequency of past-year intake of white and chocolate milk, cheese, and yogurt; the white milk question indicated the serving size was a “glass or with cereal”, for chocolate milk the serving was a “glass”, the cheese serving was “1 slice”, and yogurt serving was “1 cup – not frozen”. We combined white and chocolate milk intakes to get servings/day of dairy milk (soy-milk excluded). Children also reported the fat content of milk they usually drink (whole, 2%, 1%, skim). Total dairy fat and dairy protein intakes were calculated from milk, butter, yogurt and cheese as whole foods and as ingredients in other foods. Total daily caloric intakes below 500 kcal/day or greater than 5,000 kcal/day were considered implausible and set to missing. Further details on our childhood FFQ and derivation of nutrient values were provided earlier (6).

## Other Variables

At baseline, participants reported their race/ethnic group by marking all (of six) options that applied; most are white/non-Hispanic (95%), as are BBD cases. We computed ages (to the month) from dates of questionnaire return and birth. Early surveys annually asked “Have you started having menstrual periods?” and “If yes, age when periods began”. The derivation of peak height growth velocity and childhood adiposity were described earlier (7). Multiple surveys asked participants whether they had been pregnant. Cumulative alcohol intake was obtained from alcohol reported on the 2000–2003 surveys (16). Participants’ family history of breast disease was derived earlier (17).

## Statistical Analyses

We compared, using baseline (1996) data, females in these analyses (returned 2005, 2007 and/or 2010 surveys with BBD questions) with females not included.

Because we did not have information regarding when cases were diagnosed, prevalent biopsy-confirmed BBD ( $n=105$ ) became the outcome for all analyses. Logistic regression models, estimated using SAS (18), provided odds ratios (OR) and 95% confidence intervals (CI). Because age was related to each female’s chance of being diagnosed during follow-up, all models adjusted for baseline age (to month). Nutrients (dairy protein, dairy fat) were adjusted for energy by the residual method, and foods were energy-adjusted by including total energy intake in relevant models. Multivariable-adjusted models additionally included age at menarche, childhood BMI, adolescent alcohol consumption, and ever-pregnant.

We investigated dairy intakes at 14yr, which falls within the critical period between menses-onset and first pregnancy. We also examined intakes at 10yr, but there were fewer participants as many were older than 10 at baseline. We further derived dairy intakes specific to developmental periods: diet prior to the growth spurt was the cumulative average of intakes reported from baseline up to the survey before the growth spurt, while diet during the growth spurt was computed as the mean of two intakes nearest peak velocity. Diet before menses-onset was the mean of intakes from baseline up to the survey before menses-onset,

while diet after onset was the mean of intakes reported post-menses-onset. Because risk factors may differ by family history, we fit stratified models.

## Results

Surveys returned up to September 2011 provided responses to our BBD question for 7011 females, who reported 105 biopsy-confirmed BBD cases and 6755 non-cases. Baseline data of females who returned at least one of the 2005 to 2010 surveys (78% of original cohort) are compared with females returning none. At baseline, the included girls were slightly younger (by 5.5 weeks) than those not included, and were slightly taller (+0.17 inch), consumed more milk (+0.18 glass/day), more yogurt (+0.02 cups/day), and more dairy protein (age-adjusted, dietary variables energy-adjusted, all  $p < .05$ ). However, baseline body fatness, energy intake, cheese, dairy fat, and menarche status were similar between the included and missing females. Thus, some older participants more likely to have BBD may be missing, but also some shorter participants less likely to have BBD, and some who consumed less milk and yogurt. This slightly limited range of exposures on these variables may reduce our ability to detect associations.

In age-energy-adjusted models of dairy intakes (total dairy, milk, yogurt, cheese, dairy protein, dairy fat) at age 14yr, we found no significant associations with BBD risk in these young women (Table 1). Multivariable-adjustment did not alter these findings (Table 1). Re-analyzing milk as a categorical variable, 14yo girls who drank the most (2+ servings/day) were not at increased risk relative to those who drank the least (never to 1 serving/week) (age-energy-adjusted OR=1.05, 95% CI: 0.52–2.15,  $p = .89$ ; multivariable-adjusted OR=1.06, 95% CI: 0.52–2.18,  $p = .86$ ). Analyses of intakes at age 10yr provided no significant associations between dairy and BBD (not shown). Models investigating dairy intakes during developmental stages (before height growth spurt, during growth spurt, before menses-onset, and after menses-onset), again found no significant associations (Table 2).

Because previous work suggested that some risk factors for BBD may vary by family history (17), we further investigated dairy variables separately by family history of breast disease, and observed no associations between dairy and BBD (not shown). Admittedly, the numbers of BBD cases in family history subgroups were small (ranging from 17 to 57 cases), so power is limited.

## Discussion

This is the first investigation of dairy intakes reported during adolescence, rather than recalled later in life, and risk of BBD in young women. Previous research found that dairy products promoted height growth (6,19), and more rapid height growth was associated with increased risk for BBD (7) and breast cancer (20–21). Whether rapid growth itself or related factors, such as growth-promoting dietary intakes or hormones, are cancer initiators/promoters is unknown. However, we found no link between adolescent dairy intakes and BBD in young women. Other factors in milk, such as vitamin D added by fortification, could modify the risk of BBD.

Our findings are reasonably consistent with the literature. Analyses of nurses, who at 33–53yrs of age recalled their high school diets, found no associations between proliferative BBD risk (based on centralized pathology specimen review) and milk or dairy intakes (22). Studies of childhood diet and adult breast cancer provided similarly null conclusions; the few studies did not consistently support any association between early dairy consumption and breast cancer (23–25), though in most childhood diets were recalled in adulthood.

The longitudinal design of this investigation is a strength; dietary data were collected, in real time, years prior to the reporting of BBD in this large cohort of girls from all over the US. Though we controlled for age and energy and included other potential confounders in multivariable-adjusted models, and further analyses stratified by family history, some residual and unmeasured confounding may remain. We cannot exclude the possibility of incomplete adjustment, or confounding through variables not considered, but little is known about childhood risk factors for BBD. Although our cohort is not representative of US females, the comparison of risks within our cohort should still be valid and generalizable. Because participants are daughters of nurses, this reduces confounding by socioeconomic and other unmeasured factors, while enhancing the accuracy of the data. The racial/ethnic makeup of our cohort (95% white/non-Hispanic) hinders generalization to other races/ethnicities.

The most serious limitation is our small number of cases, but we previously reported significant associations between BBD and adolescent alcohol consumption (16) and growth variables (7). Even if insufficient power were responsible for our null findings, most odds ratios were in the opposite direction from supporting our expectation that milk would increase BBD risk. Because some of the not-biopsy-confirmed (excluded) cases may be valid, we further re-estimated all the multivariable-adjusted odds ratios (Tables 1, 2) with them included among the BBD cases. In spite of more than doubling the number of cases, only one odds ratio (of 30) became significant, likely due to chance. Another limitation was the necessity to collect data by self-report on (paper and web-based) questionnaires, but with our large, geographically dispersed cohort, alternatives were not feasible. Dietary-intake reporting errors are likely non-differential with respect to subsequent BBD. Many of the dietary variables in our analyses were cumulative averages of intakes reported over multiple years, which should minimize measurement error bias in risk estimates.

We assessed BBD risk in young women in relation to their dairy intakes during adolescence, a critical period for the development of breast cancer and other adult diseases, and found no significant associations. Because our number of cases was small, and because certain dietary factors may require longer time between exposure and development of the disease, continued follow-up of this cohort will be critical to re-assess these null results as new cases of BBD are diagnosed.

## Acknowledgments

The authors appreciate the ongoing, since 1996, dedication of our female GUTS participants.

**Funding:** Supported by a grant from The Breast Cancer Research Foundation (NYC, NY) and by DK46834 from the National Institutes of Health (Bethesda, MD). Dr. Frazier was supported by an award from the American Institute for Cancer Research. Dr. Colditz was supported, in part, by an American Cancer Society Clinical Research Professorship.

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**Table 1**

Associations between adolescent (age 14yr) dairy intakes and risk of BBD in young women: dietary intake means (and standard deviations, SD) by BBD status, and odds ratios (OR) \*

Total N=	Never reported BBD	Biopsy-confirmed BBD	Age-Energy-Adjusted		Multivariable-Adjusted	
	6755	105	OR	(95%CI)	OR	(95%CI)
<u>Intakes at 14 years</u>	<u>Mean (SD)</u>	<u>Mean (SD)</u>				
Total dairy (serv/day)	2.32 (1.53)	2.20 (1.32)	0.94	(0.81–1.08)	0.94	(0.82–1.09)
Milk (serv/day)	1.72 (1.31)	1.56 (1.13)	0.90	(0.76–1.05)	0.89	(0.76–1.05)
Yogurt (cup/day)	0.15 (0.26)	0.18 (0.27)	1.69	(0.88–3.25)	1.79	(0.93–3.47)
Cheese (slice/day)	0.46 (0.60)	0.45 (0.48)	0.99	(0.71–1.39)	0.99	(0.71–1.40)
Dairy protein (10gm/day)	2.68 (1.08)	2.64 (1.06)	0.98	(0.82–1.17)	0.99	(0.82–1.18)
Dairy fat (10gm/day)	1.75 (0.74)	1.73 (0.72)	1.04	(0.80–1.36)	0.99	(0.76–1.31)

\* A total of 6860 GUTS females, born between 1981 and 1987 and followed from childhood (1996) to young adulthood, returned surveys between years 2005 and 2011 with BBD information. When dairy variables were not available at age 14yrs, values between ages 13.5 and 15.5 were used, and if still missing, values obtained from 13.0 to 15.99yr. Multivariable-adjustment further included age at menarche, childhood adiposity (BMI at 10yr), adolescent alcohol intake, and pregnancy ever. Yogurt's larger OR's do not imply a threshold effect at high levels; girls consuming 1+ cups/day (OR=1.39 versus none, p=.54) had risk below that of some smaller intake categories.



**Table 2**

Associations between dairy intakes during developmental stages and risk of BBD in young women: dietary intake means (and standard deviations) by BBD status, and odds ratios (OR)<sup>\*</sup>

	Never reported BBD	Biopsy-confirmed BBD	Age-Energy- Adjusted		Multivariable- Adjusted	
	Mean (SD)	Mean (SD)	OR	(95%CI)	OR	(95%CI)
<b>Intakes prior to growth spurt</b>	(N=3932)	(N=53)				
Total dairy (serv/day)	2.50 (1.42)	2.44 (1.29)	0.94	(0.76–1.16)	0.94	(0.76–1.17)
Milk (serv/day)	1.96 (1.23)	1.83 (1.15)	0.89	(0.71–1.13)	0.89	(0.70–1.13)
Yogurt (cups/day)	0.13 (0.21)	0.13 (0.23)	0.88	(0.23–3.37)	0.88	(0.23–3.38)
Cheese (slices/day)	0.42 (0.54)	0.49 (0.58)	1.16	(0.74–1.83)	1.18	(0.75–1.86)
Dairy protein (10gm/day)	2.83 (1.07)	2.70 (0.98)	0.90	(0.70–1.17)	0.90	(0.70–1.17)
Dairy fat (10gm/day)	1.76 (0.72)	1.66 (0.66)	0.82	(0.55–1.22)	0.80	(0.53–1.20)
<b>Intakes during growth spurt</b>	(N=3924)	(N=53)				
Total dairy (serv/day)	2.44 (1.32)	2.51 (1.28)	0.97	(0.78–1.22)	0.97	(0.78–1.22)
Milk (serv/day)	1.88 (1.14)	1.88 (1.09)	0.95	(0.74–1.22)	0.95	(0.74–1.22)
Yogurt (cup/day)	0.13 (0.19)	0.14 (0.21)	1.10	(0.28–4.29)	1.11	(0.28–4.32)
Cheese (slices/day)	0.44 (0.51)	0.49 (0.44)	1.05	(0.63–1.75)	1.06	(0.64–1.76)
Dairy protein (10gm/day)	2.77 (1.02)	2.67 (0.95)	0.92	(0.69–1.21)	0.92	(0.70–1.22)
Dairy fat (10gm/day)	1.75 (0.68)	1.66 (0.61)	0.83	(0.54–1.28)	0.82	(0.53–1.27)
<b>Intakes prior to menses onset</b>	(N=4337)	(N=60)				
Total dairy (serv/day)	2.46 (1.34)	2.43 (1.20)	0.94	(0.77–1.16)	0.94	(0.76–1.16)
Milk (serv/day)	1.91 (1.16)	1.81 (1.06)	0.89	(0.71–1.12)	0.89	(0.70–1.12)
Yogurt (cup/day)	0.13 (0.20)	0.13 (0.23)	0.94	(0.26–3.38)	0.96	(0.27–3.48)
Cheese (slices/day)	0.43 (0.51)	0.49 (0.49)	1.16	(0.74–1.82)	1.18	(0.75–1.86)
Dairy protein (10gm/day)	2.79 (1.02)	2.71 (0.89)	0.93	(0.72–1.19)	0.93	(0.72–1.20)
Dairy fat (10gm/day)	1.75 (0.69)	1.71 (0.63)	0.93	(0.64–1.35)	0.91	(0.62–1.33)
<b>Intakes after menses onset</b>	(N=6503)	(N=103)				
Total dairy (serv/day)	2.22 (1.26)	2.15 (1.25)	0.93	(0.79–1.11)	0.94	(0.79–1.12)
Milk (serv/day)	1.59 (1.09)	1.49 (1.01)	0.91	(0.75–1.10)	0.91	(0.75–1.11)
Yogurt (cup/day)	0.15 (0.20)	0.16 (0.19)	1.14	(0.43–3.03)	1.16	(0.44–3.05)
Cheese (slices/day)	0.48 (0.48)	0.50 (0.42)	1.02	(0.67–1.55)	1.03	(0.68–1.55)
Dairy protein (10gm/day)	2.65 (1.01)	2.56 (0.99)	0.93	(0.75–1.15)	0.94	(0.76–1.17)
Dairy fat (10gm/day)	1.82 (0.71)	1.77 (0.71)	1.02	(0.75–1.38)	0.97	(0.72–1.32)

\* GUTS females, born between 1981 and 1987 and followed from childhood (in 1996) to young adulthood, returned surveys between years 2005 and 2011 with BBD information. Numbers of BBD cases and non-cases who provided data during each developmental period are shown. Multivariable-adjustment further included age at menarche, childhood adiposity (BMI at 10yr), adolescent alcohol intake, and pregnancy ever.