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Prenatal DES exposure in relation to breast size

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Abstract

Purpose—Prenatal DES exposure has been associated with increased risk of breast cancer, but the mechanisms are unknown. Larger bra cup size has also been associated with increased breast cancer risk, although not consistently. We investigated the relation of prenatal DES exposure to mammary gland mass, as estimated by bra cup size.

Methods—In 2006, 3,222 DES-exposed and 1,463 unexposed women reported their bra cup size, band size (chest circumference), and weight at age 20. Prevalence ratios (PR) were calculated for DES exposure in relation to large bra cup size, with control for year of birth and study cohort. Primary analyses were carried out among women who reported a chest circumference of no more than 32 inches because their cup size would be less influenced by fat mass.

Results—Within this group, DES-exposed women had an estimated 45% increased prevalence (95% CI 0.97-2.18) of large cup size (C or greater) relative to unexposed women. The PR was further increased among women in this group who had a body mass index of <21 at age 20: PR = 1.83 (95% CI 1.11-3.00). The PR for high-dose DES exposure relative to no exposure was 1.67, 95% CI 1.02-2.73, whereas there was no association of bra cup size with low-dose exposure.

Conclusions—These results provide support for the hypothesis that *in utero* DES exposure may result in greater mammary gland mass. Taken together with previous research on bra size and breast cancer risk, these findings suggest a mechanism for a possible association of *in utero* DES exposure with increased risk of breast cancer.

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INTRODUCTION

Diethylstilbestrol (DES) is a synthetic estrogen widely prescribed to pregnant women from the 1940s to 1960s. Recent findings suggest that women exposed *in utero* to DES may have an increased risk of developing breast cancer as adults (1). If there is indeed a causal relation between prenatal DES exposure and breast cancer risk, one mechanism may involve DES effects on the total number of mammary gland cells formed during gestation (2). A number of studies have found a lower breast cancer risk in women who self-report small breast size (3-6), small bra cup size (7, 8) or have relatively small breasts as evidenced by self-report of not wearing a bra (9), or having undergone breast augmentation surgery (10, 11); other studies have found no evidence of an association between breast size and breast cancer risk (12-15). We examined the relation between prenatal DES exposure and bra cup size, a surrogate for mammary gland mass, in a cohort of women being followed for health outcomes as part of the National Cancer Institute Collaborative Follow-up of DES-exposed cohorts.

METHODS

Study participants

Since 1994, women in previously existing cohorts of DES-exposed and comparison unexposed persons have been followed by questionnaire in a uniform manner in the NCI Collaborative Follow-up of DES-Exposed Cohorts. As described previously (16), the existing cohorts of daughters were from (a) the National Cooperative Diethylstilbestrol Adenosis Project (DESAD) (17); (b) a randomized clinical trial of DES carried out at the University of Chicago in 1951-1952 (Dieckmann) (18), and (c) a large private infertility practice in Massachusetts (Horne). In addition, in 1994, a new cohort of DES-exposed and unexposed daughters was identified through Maine, New Hampshire, and Massachusetts mothers who previously participated in an early 1980's study of DES-related outcomes in women who were given DES during pregnancy (Women's Health Study) (19). Each of these four sub-cohorts included both DES-exposed daughters and unexposed daughters. Review of the mother's obstetrical record provided documentation of DES exposure (or lack thereof) for all exposed and unexposed daughters. There were 4,817 exposed and 2,073 unexposed women eligible for inclusion in the study in 1994.

Data collection

Questionnaires were mailed to all eligible participants in 1994 and were completed by 3,932 exposed (82%) and 1,735 unexposed (84%) women. The questionnaires ascertained information on adult height, current weight, weight at age 20, reproductive factors, use of female hormones, cigarette smoking, alcohol use, and health outcomes. Follow-up questionnaires have been mailed every 3-5 years since then (in 1994, 1997, 2001, 2006 and 2011). The 2006 follow-up questionnaire, completed by 3,222 DES-exposed and 1,463 unexposed women, asked about bra size at age 20, including bra band size (chest circumference, e.g., 32, 34, 36, 38, 40 etc.) and bra cup size (AA, A, B, C, D, DD, etc., with AA representing the smallest and DD the largest size).

Data on total dose of DES taken by the participant's mother was available for 38% of participants. Previous analyses from the DES Collaborative Follow-up Study have used original cohort as a crude marker for low and high dose of exposure because there were regional prescribing patterns, with physicians in Massachusetts and California generally prescribing DES to be taken throughout the pregnancy with a resulting high total dose and exposure late in pregnancy as well as early in pregnancy, and physicians at the Mayo Clinic prescribing the drug for a markedly shorter interval. Women born to mothers who participated in the University of Chicago randomized trial all received a very high total dose. We used this previously established variable to classify DES-exposed participants as having received a "low-dose", "high-dose", or unknown level of dose (women born in Maine or New Hampshire).

Statistical Analysis

All women who completed the 2006 questionnaire and answered the questions on bra size were included in the analysis. The log binomial model for prevalence ratios (20) was used to estimate the prevalence ratio of large bra cup size (C or greater) in DES-exposed women relative to unexposed women. Women who are overweight will have a relatively large cup size simply because of excess fat in the breast. Overall adipose tissue also contributes to a larger chest circumference. Bra cup size has been shown to be a more accurate measure of breast volume among women who have a small chest circumference than among all women considered together(21). For this reason, we stratified all analyses on bra band size (chest circumference). We also carried out a sub-analysis restricted to thin women (those with a body mass index of <21 kg/m2 at age 20). All analyses were controlled for year of birth and cohort. The primary exposure variable was DES exposure (ever vs. never); additional analyses assessed high and low-dose DES exposure relative to no exposure.

RESULTS

Almost all participants reported bra band size in even integers, with 97% reporting a size of 32, 34, 36, or 38. We used these data to create three strata of chest circumference ≤32, 33 or 34, and 35 or higher. As shown in Table 1, there was no association of DES exposure with bra cup size among women in the two highest categories of band size. However, among women with a band size of ≤32, the prevalence ratio (PR) for DES exposure in relation to large cup size (C or larger) was 1.45 (95% confidence interval (CI) 0.97-2.18). We further restricted the analysis to women who were both thin at age 20 (body mass index <21 kg/m^2) and had a bra band size of 32 or less; the PR for large cup size in DES exposed compared with unexposed was 1.83 (95% CI 1.11-3.00).

To assess whether birth weight could be an intervening variable in a possible causal pathway between prenatal DES exposure and bra cup size, we carried out additional analyses that included indicator terms for birth weight. The PR for DES in relation to large cup size among women with band size ≤32 was 1.56, 95% CI 1.04-2.32, similar to that obtained in the model without terms for birth weight.

Table 2 presents results according to whether total DES exposure was likely to have been high or low. High-dose DES was associated with an increased prevalence of large bra cup size: PR = 1.67, 95% CI 1.02-2.73). There was no association with low-dose exposure.

DISCUSSION

The present analyses were designed to assess whether DES exposure influences breast volume. The study question was posed because preliminary data indicate a positive association between prenatal DES exposure and risk of breast cancer, the mechanisms for such an association are unknown, and several lines of evidence point to an association between breast size and risk of breast cancer. Our primary analyses were restricted to women who had a small chest circumference at age 20, as indicated by bra band size, under the assumption that these women would have had relatively little adipose tissue in their breasts and therefore bra cup size would closely approximate mammary gland mass. We found that these DES-exposed women had an estimated 45% greater prevalence of large bra cup size, and that the increase was even greater among women who were lean at age 20. These results provide evidence in support of the hypothesis that *in utero* DES exposure may cause an increase in the total number of breast epithelial cells formed prenatally and in early life. In addition, the prevalence ratio was highest among women who received the highest doses of DES. The observed increase in bra cup size associated with DES exposure did not appear to be mediated by birth weight.

The mechanisms by which prenatal exposures, including exposure to DES, influence breast development or breast cancer risk, have not been established (22). Trichopoulos and others have postulated that altered prenatal hormone exposure may lead to fetal development of a greater number of mammary gland cells, and therefore, a greater number of ductal stem cells at risk of carcinogenic stimulation after birth (23, 24). Animal studies have demonstrated long-lasting effects of neonatal DES exposure on proliferation and differentiation of mammary glands in mice (25). Mammary gland development involves an interplay of endogenous androgens, estrogens, prolactin, and insulin (26, 27), occurring in mid and late gestation.

Epidemiologic studies relating bra size or other measures of breast volume to risk of breast cancer have had conflicting findings. Several studies that restricted the analysis to women with a smaller bra band size or to non-overweight women found an increased risk of breast cancer in those with the larger cup sizes (7, 8). Others with similar methods found no association (12-15). A recent analysis of GWAS data from the customer base of 23andMe, Inc., a consumer genetics company, identified seven genetic variants that were associated at genome-wide significance with self-reported bra cup size, after control for bra band size (28). Two of the variants associated with larger bra cup size were variants in high linkage disequilibrium with variants previously associated with increased breast cancer risk in large GWAS studies (29, 30). These findings indicate that genetic variants in the same regions are involved in both breast size and breast cancer risk.

A limitation of our study is the reliance on bra cup size as a marker for mammary gland mass. Misclassification of the outcome is likely because we asked middle-aged women to

recall their bra size at age 20. Misclassification would have been unrelated to DES exposure status and would have reduced power to detect a true association. Despite nondifferential misclassification, we observed a 50% increase in the prevalence of large bra cup size among DES exposed women who had a narrow chest circumference. Another limitation is the lack of comprehensive data on total dose of DES exposure. Nevertheless, when study subjects were grouped according to whether mothers in their region of birth typically received a high or low total dose of DES, large cup size was associated with high-dose exposure relative to no exposure, but was not associated with low-dose exposure.

It is not clear which factors, if any, would be potential confounders of a relation of *in utero* DES exposure with breast volume in young adulthood. We controlled for year of birth, which may be important because other unmeasured endocrine disruptors have changed over time in the United States.

In conclusion, the present findings provide support for the hypothesis that *in utero* DES exposure may result in greater mammary gland mass. Taken together with previous research on bra size and breast cancer risk, these findings suggest a mechanism for a possible association of *in utero* DES exposure with increased risk of breast cancer.

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Table 1

Prenatal DES exposure in relation to large bra cup size, according to bra band size and body mass index (BMI) at age 20

	Bra cup size		
DES exposure	**Small (AA,A,B)**	**Large (C,D,DD)**	**Prevalence ratio (95% CI)**
Bra band size >34			
Unexposed	228	220	Reference
Exposed	468	462	0.97 (0.86 - 1.10)
Bra band size 33, 34			
Unexposed	531	138	Reference
Exposed	1,180	325	0.94 (0.78 - 1.13)
Bra band size ≤32			
Unexposed	314	32	Reference
Exposed	685	102	1.45 (0.97 - 2.18)
Bra band size ≤ 32 and BMI <21			
Unexposed	261	22	Reference
Exposed	520	76	1.83 (1.11-3.00)

* Prevalence ratios adjusted for year of birth and study cohort

Table 2

Total DES dose in relation to large bra cup size among women with bra band size ≤32

	Bra cup size		
DES exposure	**Small (AA,A,B)**	**Large (C,D,DD)**	**Prevalence ratio* (95% CI)**
Unexposed	314	32	1.00 Reference
“Low-dose”	275	33	0.94 (0.45 - 1.94)
“High-dose”	383	64	1.67 (1.02 – 2.73)

* Prevalence ratios adjusted for year of birth and study cohort “Low-dose” cohorts: Texas, Minnesota, Wisconsin "High-dose" cohorts: Massachusetts, Illinois, California Excluded: New Hampshire, Maine