Case–control study of breast cancer and exposure to synthetic environmental chemicals among Alaska Native women

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Background. Exposure to environmental chemicals may impair endocrine system function. Alaska Native (AN) women may be at higher risk of exposure to these endocrine disrupting chemicals, which may contribute to breast cancer in this population.

Objective. To measure the association between exposure to select environmental chemicals and breast cancer among AN women.

Design. A case–control study of 170 women (75 cases, 95 controls) recruited from the AN Medical Center from 1999 to 2002. Participants provided urine and serum samples. Serum was analyzed for 9 persistent pesticides, 34 polychlorinated biphenyl (PCB) congeners, and 8 polybrominated diethyl ether (PBDE) congeners. Urine was analyzed for 10 phthalate metabolites. We calculated geometric means (GM) and compared cases and controls using logistic regression.

Results. Serum concentrations of most pesticides and 3 indicator PCB congeners (PCB-138/158; PCB-153, PCB-180) were lower in case women than controls. BDE-47 was significantly higher in case women (GM = 38.8 ng/g lipid) than controls (GM = 25.1 ng/g lipid) (p = 0.04). Persistent pesticides, PCBs, and most phthalate metabolites were not associated with case status in univariate logistic regression. The odds of being a case were higher for those with urinary mono-(2-ethylhexyl) phthalate (MEHP) concentrations that were above the median; this relationship was seen in both univariate (OR 2.16, 95% CI 1.16–4.05, p = 0.02) and multivariable (OR 2.43, 95% CI 1.13–5.25, p = 0.02) logistic regression. Women with oestrogen receptor (ER)/progesterone receptor (PR)-tumour types tended to have higher concentrations of persistent pesticides than did ER+/PR+ women, although these differences were not statistically significant.

Conclusions. Exposure to the parent compound of the phthalate metabolite MEHP may be associated with breast cancer. However, our study is limited by small sample size and an inability to control for the confounding effects of body mass index. The association between BDE-47 and breast cancer warrants further investigation.

Keywords: Alaska Native; breast cancer; environmental chemical exposure; persistent pesticides; polychlorinated biphenyl ethers; phthalates

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Cancer is the leading cause of death among Alaska Native (AN) men and women (1). Breast cancer is among the most frequently diagnosed cancers in this population; from 2004 to 2008, breast cancer accounted for 14% of all cancer deaths among AN women (2). In the 40 years between 1969 and 2008, the incidence of breast cancer among AN women tripled (2). Although breast cancer rates of AN women have been lower than that of US white women, the rates are now similar (2).

Exposure to organochlorine pesticides and polybrominated biphenyls (PCBs), polybrominated diethyl ethers (PBDEs), and phthalates (collectively referred to here as environmental chemicals) may impair the normal functioning of the endocrine system (3). Organochlorine pesticides and PCBs in particular have been studied for their association with breast cancer incidence, with conflicting results (4–16). In 2006, Rubin et al. conducted a pilot study evaluating the relationship between breast cancer and concentrations of p,p′-dichlorodiphenylchloroethylene (DDE), a metabolite of dichlorodiphenylchloroethylene (DDT), and PCBs among AN women. The results showed ubiquitous exposure to a number of persistent chemicals at levels similar to those reported in the contiguous US states. The study reported slightly higher levels of DDE, but slightly lower levels of PCBs among AN women diagnosed with breast cancer compared to women who were not diagnosed with breast cancer. However, this relationship was not statistically significant when controlling for the risk factors such as diet, ethnicity, and geographic region of residence. Some studies reported an association between breast cancer and DDE exposure (7–9), while others reported no association (10–14). Similarly, studies of breast cancer and exposure to PCBs reported conflicting results (7–9,12,15,16).

PBDEs have demonstrated endocrine disrupting capabilities in animal and in vitro studies (17–20). The PBDE congener BDE-47 is consistently detectable and at higher concentrations than other BDE congeners in human, animal, and environmental samples. A recent case–control study was conducted to determine whether risk of breast cancer was associated with concentrations of PBDE congeners measured in adipose tissue. No association was found (21). Phthalates have also demonstrated possible hormone-disrupting properties, although the human health effects of chronic low-dose exposure to phthalates are currently unknown (22). Only one study was found that examined the association between exposure to phthalates and breast cancer, and reported a positive association between urinary concentrations of monoethyl phthalate (MEP) and breast cancer (23).

AN populations may be at higher risk for exposure to certain environmental chemicals for a number of reasons. Organochlorine pesticides and PCBs resist degradation and persist in the environment. Marine animals caught in the Arctic, a large part of a traditional AN diet, are reported to have higher concentrations of synthetic persistent organic pollutants (4,24,25) than their southern counterparts as a result of atmospheric and oceanic transport and deposition of environmental chemicals in the Arctic (26).

Technical pentaBDE containing PBDE congeners with 3–6 bromine atoms were added to consumer products such as furniture cushions and wall-to-wall carpet pads to decrease flammability. Technical octa- and decaBDE with higher degrees of bromination were used in hard plastics, such as casings for electrical appliances. The predominant routes of human exposure to PBDE mixtures are ingestion and/or inhalation of indoor residential dust and ingestion of food containing these chemicals (27). For AN people who consume foods comprising a traditional diet, ingesting food containing these chemicals is quantitatively a more important route of exposure than for the general US population.

Added to common household, personal care, medical, and commercial products, human exposure to phthalates is ubiquitous in the United States (22). Unlike the other environmental chemicals discussed here, phthalates do not persist in the environment and do not bioaccumulate (22,28). Although phthalates are easily metabolized and excreted in the urine, their pervasiveness and potential hormone-disrupting effects present a concern for chronic low-dose exposure (28).

The objective of this study was to investigate possible associations between breast cancer among AN women and concentrations of select environmental chemicals measured at the time of enrolment. A strength of this study is the use of pre-treatment samples to quantify chemical exposures. To determine whether different types of tumours have different risk factors, we examined differences in concentrations of environmental chemicals in women with oestrogen receptor (ER) and progesterone receptor (PR) positive and ER/PR-negative tumours. We discuss environmental chemicals measured in serum (organochlorine pesticides, PCBs, and PBDEs) and urine (phthalate metabolites) collected at enrolment in relation to demographic characteristics, health history information, and breast tissue pathology, including ER/PR status.

Methods

Screening and enrolment
The protocol for this study was reviewed and approved by the Institutional Review Boards (IRB) of the Alaska Area and the US Centers for Disease Control and Prevention (CDC). We invited women who presented to the Alaska Native Medical Center (ANMC) in Anchorage, Alaska for excisional biopsy, lumpectomy, mastectomy, or breast reconstruction between March 1999 and March 2002 to enrol in this study. Consenting participants could take part in one or more of the study's
3 components: interview; biologic specimen collection and analysis; and analysis of banked serum collected from participants during previous studies. Participants could withdraw from the study at any time, and were not provided personal results; only aggregated data were provided to participants. ANMC physicians did not know if their patients were enrolled in this study to prevent any real or perceived conflict of interest in healthcare delivery. We did not collect data about reasons for refusal to participate.

A case was defined as an enrolled participant whose surgical procedure resulted in a diagnosis of invasive or in situ breast cancer based on ANMC pathology results. Following department guidelines, biopsies with invasive cancer, or initial biopsies where invasion could not be ruled out, were sent to a contract laboratory to be analyzed for ER and PR status. A control was defined as an enrolled participant whose tissue analysis was returned from pathology without a cancer diagnosis. All control women had diagnoses of benign breast conditions, such as fibrocystic changes and hyperplasia; none had lesions suspicious for cancer. Women receiving only core needle biopsy were excluded from the study because the procedure did not yield sufficient tissue for analysis.

**Biologic specimen collection**

Blood and urine samples were obtained by the study’s research nurse in the surgery outpatient clinic during a pre-operative surgery clinic visit. Blood samples were obtained by venipuncture and collected in a 10 ml plain red-top Vacutainer tube. The ANMC laboratory separated serum from blood cells and placed the serum in glass vials, which were stored frozen at CDC Arctic Investigations Program (AIP). Urine samples were collected in glass and frozen within 2 hours of collection.

Biologic specimens were matched to the study participant by their unique identification number. To limit potential incidental contamination with chemicals of interest to the study, at no time were enrolment serum or urine samples collected or stored in plastic vials. All glass receptacles were provided by the National Center of Environmental Health’s (NCEH) Division of Laboratory Sciences (DLS) at CDC in Atlanta, Georgia. All samples collected at the time of enrolment were frozen, batched, packaged and shipped by CDC AIP to CDC DLS at intervals throughout the enrolment period.

**Participant interview**

Participants were interviewed for demographic and risk factor information, and dietary, pregnancy, and residence histories. Patient information was recorded on paper forms identified only by the participant’s unique study ID number. Completed questionnaires containing no personal identifying information were mailed to the primary investigator at CDC NCEH where data were entered and stored in a secure electronic database.

**Laboratory analysis**

Serum samples collected at enrolment were analyzed for a number of environmental chemicals, including 9 persistent pesticides (including 2 metabolites of DDT), 34 PCB congeners, and 8 PBDE congeners (29). Urine samples were analyzed for 10 phthalate metabolites (30). Concentrations of chemicals measured in serum are lipid-adjusted; concentrations of phthalate metabolites measured in urine are creatinine corrected. All laboratory analyses were completed by fall 2004.

**Data analysis**

All statistical analyses were conducted using SAS 9.1. PCB and pesticide concentrations were normalized by natural log transformation. Concentrations below the limit of detection (LOD) were assigned a value equal to the LOD divided by the square root of 2 (31). Geometric means (GM) were calculated for persistent pesticides, indicator PCBs, and BDE-47 measured in serum, and for 7 phthalate metabolites measured in urine. We report only the indicator PCBs and BDE-47 because they are the congeners most frequently found in serum and usually at the highest concentrations. These congeners are reported widely in the literature as indicators of exposure to PBDEs and PCBs. The indicator PCB congeners selected are those from dietary sources. Three of the 10 phthalate metabolites were detectable in fewer than 60% of the samples analyzed and were excluded from the analysis. GM concentrations were compared between cases and controls and between oestrogen-positive/progesterone-positive (ER+/PR+) and oestrogen-negative/progesterone-negative (ER−/PR−) cases by t-tests. Chi-square tests were used to compare categorical risk factors between cases and controls and between ER+/PR+ and ER−/PR− tumour types in cases.

The demographic variables, “place of birth” and “current residence,” were categorized into 5 recognized regions of the state: northwest, southwest, south central, interior, and southeast Alaska. Ethnicity was based on self-report at interview – participants were categorized as Eskimo, Indian, or Aleut. Study participants who were American Indian of non-Alaskan origin were categorized as “other.”

Logistic regression was used to explore the association between case status and exposure to environmental chemicals. Each chemical was assessed in 2 different univariate models: once as a continuous variable, and once as a dichotomized variable. For the latter, we dichotomized at the median. We also assessed these relationships in multivariable models that were adjusted for age at enrolment, ethnicity, smoking and alcohol use history, family history of breast cancer, number of live births, number of months of breastfeeding children, menopausal status, and hormone use history. P-value <0.05 was considered statistically significant.
Results
During enrolment, of the approximately 470 women who visited the surgery clinic for breast-related health issues, 203 were enrolled into the study. This paper presents results of serum and urine samples collected at the time of enrolment, limited to the 170 women (75 cases, 95 controls) for whom we had sufficient quantities for laboratory analyses. The 33 women who were excluded from analyses did not differ with respect to demographic variables from the 170 women who were included (data not shown).

Descriptive characteristics are shown in Table I. Age at enrolment ranged from 30 to 88 years, with a mean age of 51 years. Women of Eskimo (54.7%), Indian (21.8%), and Aleut (15.9%) ethnicities were represented. All 5 regions of the state were represented within the study population. The majority (58.2%) of participants resided in Alaska’s south central region at the time of enrolment. The fewest lived in the southeast (5.9%) and the interior (1.2%) regions. Almost one-fifth (17.6%) of women reported a family history of breast cancer. Almost all (92.4%) of the women reported having children. A history of smoking was reported by 77.6% of the women; however, at the time of enrolment, only 59.3% of respondents were current smokers. Similarly, 90% of the women reported ever drinking alcohol, but only 51.3% of respondents reported any alcohol use at the time of enrolment.

Case women reported a significantly lower median cumulative number of months of breast feeding (2 months) than control women (8 months). The variable “expecting more periods” was used as a proxy measure for menopausal status – women who responded “no” were assumed to be menopausal. More case women (63.9%) responded “no, they were not expecting more menstrual periods” than control women (45.6%). That is, more case women were menopausal than control women, a statistically significant difference (p = 0.03). No other descriptive variables differed significantly between cases and controls.

Sixty-two of 75 (83%) case women had invasive cancer tumours. We compared the descriptive characteristics and known risk factors for breast cancer shown in Table I between case women with invasive vs. non-invasive cancers and found no significant differences between them for these variables (data not shown).

GM concentrations and confidence intervals for persistent pesticides, indicator PCBs, and BDE-47 measured in serum, and phthalate metabolites measured in urine are shown in Table II. GM serum concentrations of most pesticides were lower in case women than in controls. Only gamma (γ)-hexachlorocyclohexane was higher in cases (8.3 ng/g lipid) than controls (7.7 ng/g lipid); however, this difference was not statistically significant.

The GM serum concentrations of all 3 indicator PCBs were also lower among cases than controls, although again, these differences were not statistically significant. The GM serum concentration of BDE-47 was statistically significantly higher in case women (38.84 ng/g lipid) than in controls (25.11 ng/g lipid) (p = 0.04). Although GM urinary concentrations for most of the 7 phthalate metabolites were higher among cases than controls, these differences were not statistically significant.

Results of the univariate logistic regression analyses showed no association between the median concentrations of persistent pesticides, PCBs, or most of the phthalate metabolites and breast cancer status (Table III). However, urinary concentrations above the median of mono-(2-ethylhexyl) phthalate (MEHP) were associated with breast cancer, and the association was statistically significant in both univariate and multivariable analyses (OR 2.16, 95% CI 1.16–4.05, p = 0.02 and ORadj 2.43, 95% CI 1.13–5.25, p = 0.02, respectively). The result of univariate logistic regression analysis of BDE-47 approached statistical significance (OR 1.79, 95% CI 0.97–3.32; p = 0.06). However, this association was not statistically significant in the multivariable analysis. Conversely, the result of the same analysis for p,p’-DDT suggested a slight protective effect (OR 0.55, 95% CI 0.30–0.91, p = 0.05). No significant associations between continuous concentrations of environmental chemicals and breast cancer status were found.

Of the 62 women who had invasive tumours, 40 (65%) had tumours that were positive for both oestrogen- and progesterone-binding capacity (ER+/PR+), while 13 (17.3%) were negative for both hormone markers (ER−/PR−). Women with ER−/PR− tumours (n = 13) had GM serum concentrations of persistent pesticides, PCBs, and BDE-47 that were consistently (7.7–48%) higher, although not significantly different, than women with ER+/PR+ tumours (n = 40). Urine concentrations were higher in 3 of 7 phthalate metabolites among women with ER−/PR− tumours. The differences were not statistically significant (Table IV). We estimated the odds ratios for the ER+ and ER− groups, modelling the probability of ER−. While results for MEHP were not significant, the estimates indicated a positive association.

Discussion
The scientific literature contains conflicting reports about the association between exposure to persistent organic pollutants and breast cancer (4–16). Results of some studies demonstrated an association between the DDT metabolite DDE and breast cancer (7–9), while others did not (10–14). Studies of exposure to PCBs and breast cancer incidence also reported conflicting results (7–9,12,15,16). Few studies have focused on the association between exposure to environmental contaminants and breast cancer specifically among AN women, and only one epidemiologic study was found that examined the relation between PBDE and breast cancer in any population (21). Rubin et al. (4) confirmed DDT and PCB exposure in AN women, but did not find that exposures to
Table I. Statistics for characteristics and known risk factors for breast cancer of enrolled Alaska Native women (n = 170), 1999–2002

|                                      | Total<sup>a</sup> | Case | Control | p<sup>b</sup> |
|--------------------------------------|-------------------|------|---------|--------------|
| Number of women                      | 170               | 75   | 95      |              |
| Mean age at enrolment in years (range) | 51 (30–88)       | 53 (30–78) | 50 (30–88) | 0.08         |
| Mean age at menarche in years (range) | 13 (9–19)         | 13 (9–19) | 13 (10–17) | 0.68         |
| Ethnicity                            |                   |      |         |              |
| Eskimo                               | 93 (54.7)         | 36 (48.0) | 57 (60.0) | 0.06         |
| Indian                               | 37 (21.8)         | 22 (29.3) | 15 (15.8) |              |
| Aleut                                | 27 (15.9)         | 9 (12.0) | 18 (19.0) |              |
| Other                                | 13 (7.6)          | 8 (10.7) | 5 (5.3)  |              |
| Service units by region at birth      |                   |      |         |              |
| South central (Anchorage)            | 33 (19.6)         | 15 (20.3) | 18 (19.1) | 0.08         |
| Interior (Fairbanks)                 | 8 (4.8)           | 3 (4.1) | 5 (5.3)  |              |
| Northwest (Barrow, Kotzebue, Nome)   | 46 (27.4)         | 17 (23.0) | 29 (30.9) |              |
| Southeast (Bethel & Dillingham)      | 44 (26.2)         | 16 (21.6) | 28 (29.8) |              |
| Southeast (Sitka–Mt. Edgecumbe; Annette Isl) | 17 (10.1) | 13 (17.6) | 4 (4.3)  |              |
| Service units by region at time of enrolment |         |      |         |              |
| South central (Anchorage)            | 99 (58.2)         | 42 (56.0) | 57 (60.0) | NA           |
| Interior (Fairbanks)                 | 2 (1.2)           | 1 (1.3) | 1 (1.0)  |              |
| Northwest (Barrow, Kotzebue, Nome)   | 32 (18.8)         | 14 (18.7) | 18 (19.0) |              |
| Southeast (Bethel & Dillingham)      | 26 (15.3)         | 7 (9.3) | 19 (20.0) |              |
| Southeast (Sitka–Mt. Edgecumbe; Annette Isl) | 10 (5.9) | 10 (13.3) | 0        |              |
| Other                                | 1 (0.6)           | 1 (1.3) | 0        |              |
| Highest education level completed    |                   |      |         |              |
| ≤ 8th grade                          | 27 (15.9)         | 11 (14.7) | 16 (16.8) | 0.45         |
| 9th–11th                             | 18 (10.6)         | 5 (6.7) | 13 (13.7) |              |
| High school graduate                 | 109 (64.1)        | 52 (69.3) | 57 (60.0) |              |
| ≥ College graduate                   | 16 (9.4)          | 7 (9.3) | 9 (9.5)  |              |
| Smoking (yes)                        |                   |      |         |              |
| Ever                                 | 132 (77.7)        | 62 (82.7) | 70 (73.7) | 0.20         |
| Current (n = 135)                    | 80 (59.3)         | 33 (51.6) | 47 (66.2) | 0.11         |
| Alcohol (yes)                        |                   |      |         |              |
| Ever                                 | 153 (90.0)        | 71 (94.7) | 82 (86.3) | 0.08         |
| Current (n = 154)                    | 79 (51.3)         | 34 (47.9) | 45 (54.2) | 0.52         |
| Family history of breast cancer (yes) |                   |      |         |              |
| Mother                               | 7 (4.1)           | 3 (4.0) | 4 (4.2)  | 0.58         |
| Maternal grandmother                 | 7 (4.1)           | 4 (5.3) | 3 (3.2)  | 0.29         |
| Paternal grandmother                 | 4 (2.4)           | 2 (2.7) | 2 (2.1)  | 0.91         |
| Sister                               | 10 (5.9)          | 3 (5.0) | 7 (9.1)  | 0.51         |
| Daughters                            | 2 (1.2)           | 0      | 2 (2.9)  | 0.50         |
| Pregnancies (yes)                    |                   |      |         |              |
| Ever (n = 169)                       | 158 (93.5)        | 71 (96.0) | 87 (91.6) | 0.35         |
| Live births                          |                   |      |         |              |
| Median # births (range)              | 3 (0–11)          | 3 (0–10) | 3 (0–11) | 0.77         |
| Number of women (%)                  | 157 (92.4)        | 71 (96.0) | 86 (90.5) | 0.23         |
| Breast fed                           |                   |      |         |              |
| Median cumulative months (range)     | 4 (0–216)         | 2 (0–88) | 8 (0–216) | 0.05         |
| Number of women (%)                  | 106 (62.4)        | 45 (60.0) | 61 (64.2) | 0.63         |
| Hormone history (yes)                |                   |      |         |              |
| Ever birth control pills             | 111 (66.9)        | 49 (65.3) | 62 (68.1) | 0.74         |
these chemicals were associated with breast cancer. The study did not include PBDEs or phthalates. Day et al. (32) suggested that increases of breast cancer among AN women over the past 30 years may be attributable to increased body mass index (BMI) and a shift away from a traditional diet.

We believe this is the first study to measure and report PBDE serum concentrations among AN women and the first to suggest a possible association between breast cancer and PBDE exposure in this population. The serum concentrations of BDE-47 measured in our participants ranged from 0.71 to 1,370 ng/g lipid, and the GM concentration of BDE-47 was 30.4 ng/g lipid (95% CI 24.9–37.2). AN women have GM serum concentrations of BDE-47 similar to concentrations previously reported in studies from other areas of the United States. A representative sample of US women (22) had a lipid-adjusted GM serum BDE-47 concentration of 19.6 ng/g lipid (95% CI 16.4–23.5). AN women in our study had GM concentrations of BDE-47 that more closely

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**Table II.** Geometric mean serum concentrations (lipid adjusted), 95% confidence intervals, and p-values of selected persistent pesticides, polychlorinated biphenyls (PCB), and polybrominated diphenyl ethers (PBDE), and urinary concentrations (μg/g creatinine) of phthalate metabolites in a population of Alaska Native women: 1999–2002

| Analyte                                                                 | Total (N = 170) | Case (N = 75) | Control (N = 95) | p   |
|------------------------------------------------------------------------|----------------|--------------|------------------|-----|
| **Persistent pesticides (ng/g lipid)**                                 |                |              |                  |     |
| p,p'-Dichlorodiphenylethylene (DDE)                                     | 3,912 (3,372–4,539) | 3,866 (3,103–4,866) | 3,944 (3,229–4,817) | 0.91 |
| Hexachlorobenzene                                                        | 404 (340–480) | 340 (270–433) | 464 (365–590) | 0.08 |
| Mirex                                                                  | 39.9 (33.3–47.8) | 33.1 (25.8–42.5) | 45.6 (35.5–59.2) | 0.08 |
| Oxychlordane                                                           | 241 (200–290) | 215 (166–276) | 265 (200–347) | 0.28 |
| β-Hexachlorocyclohexane                                                | 151 (126–180) | 143 (110–183) | 158 (123–202) | 0.57 |
| γ-Hexachlorocyclohexane                                                | 8.02 (7.60–8.47) | 8.33 (7.77–8.94) | 7.77 (5.99–8.50) | 0.23 |
| o,p'-Dichlorodiphenytrichloroethane (DDT)                              | 15.6 (14.1–17.3) | 15.0 (13.1–17.3) | 16.1 (13.9–18.7) | 0.51 |
| p,p'-DDT                                                               | 62.8 (55.0–71.8) | 56.3 (46.5–68.0) | 68.7 (56.8–82.3) | 0.16 |
| trans-Nonaclor                                                          | 344 (282–420) | 302 (226–403) | 380 (290–503) | 0.27 |
| **Polychlorinated biphenyls (ng/g lipid)**                             |                |              |                  |     |
| 138/158                                                                | 413 (347–491) | 386 (304–490) | 436 (340–558) | 0.50 |
| 153                                                                   | 590 (494–705) | 541 (424–689) | 632 (491–815) | 0.39 |
| 180                                                                   | 345 (297–401) | 325 (266–398) | 362 (291–450) | 0.49 |
| **Polybrominated diphenyl ether (ng/g lipid)**                         |                |              |                  |     |
| 47                                                                    | 30.4 (24.9–37.2) | 38.8 (28.6–52.7) | 25.1 (19.3–32.7) | 0.04 |
| **Phthalate metabolites (μg/g creatinine)**                            |                |              |                  |     |
| Mono-butyl phthalate (MBP)                                              | 15.9 (13.3–19.1) | 14.6 (10.7–19.8) | 17.1 (13.8–21.2) | 0.41 |
| Mono-benzyl phthalate (MBzP)                                            | 7.1 (6.1–8.4) | 7.6 (5.8–10.0) | 6.7 (5.6–8.2) | 0.46 |
| Mono-(2-ethyl-5-hydroxyhexyl) phthalate (MEHHP)                        | 32.8 (27.7–38.9) | 36.4 (27.6–48.0) | 30.1 (24.4–37.2) | 0.28 |
| Mono-(2-ethylhexyl) phthalate (MEHP)                                   | 4.2 (3.3–5.4) | 5.3 (3.6–7.7) | 3.5 (2.6–4.7) | 0.10 |
| Mono-(2-ethyl-5-oxohexyl) phthalate (MOEHp)                            | 21.9 (18.7–25.7) | 24.3 (18.8–31.5) | 20.1 (16.4–24.5) | 0.24 |
| Mono-ethyl phthalate (MEP)                                              | 57.4 (45.8–71.9) | 50.5 (36.4–70.0) | 63.8 (46.8–87.0) | 0.31 |
| Mono-methyl phthalate (MMP)                                            | 1.7 (1.4–2.1) | 1.7 (1.3–2.3) | 1.7 (1.4–2.2) | 0.98 |
**Table III.** Univariate and multivariable odds ratios (OR), 95% confidence intervals, and p-values for the association between breast cancer and selected persistent pesticides, polychlorinated biphenyls (PCB), polybrominated diphenyl ethers (PBDE), and phthalate metabolites among Alaska Native women – 1999–2002

| Analyte                                      | Univariate OR (95% confidence interval) | p-value | Multivariable OR (95% confidence interval) | p-value |
|----------------------------------------------|----------------------------------------|---------|--------------------------------------------|---------|
| **Persistent pesticides**                    |                                        |         |                                            |         |
| p,p′-Dichlorodiphenyletherylene (DDE)        | 1.05 (0.57–1.92) 0.88                   |         | 0.60 (0.26–1.43) 0.25                      |         |
| Hexachlorobenzene                            | 0.78 (0.43–1.44) 0.44                   |         | 0.47 (0.19–1.16) 0.10                      |         |
| Mirex                                        | 0.82 (0.45–1.50) 0.55                   |         | 0.65 (0.28–1.55) 0.33                      |         |
| Oxychlorodiane                               | 1.05 (0.57–1.92) 0.88                   |         | 0.91 (0.35–2.35) 0.84                      |         |
| β-Hexachlorocyclohexane                      | 1.19 (0.65–2.18) 0.58                   |         | 1.21 (0.44–3.31) 0.71                      |         |
| γ-Hexachlorocyclohexane                      | 1.70 (0.92–3.12) 0.09                   |         | 1.88 (0.88–4.03) 0.10                      |         |
| o,p′-Dichlorodiphenyltrichloroethane (DDT)   | 0.99 (0.54–1.82) 0.99                   |         | 1.00 (0.48–2.11) 1.00                      |         |
| p,p′-DDT                                     | 0.55 (0.30–1.01) 0.05                   |         | 0.49 (0.22–1.10) 0.08                      |         |
| trans-Nonachlor                              | 0.98 (0.53–1.80) 0.95                   |         | 0.65 (0.26–1.66) 0.37                      |         |
| **Polychlorinated biphenyls**                |                                        |         |                                            |         |
| 138/158                                      | 0.95 (0.52–1.75) 0.88                   |         | 0.64 (0.27–1.52) 0.31                      |         |
| 153                                          | 0.95 (0.52–1.75) 0.88                   |         | 0.55 (0.22–1.41) 0.21                      |         |
| 180                                          | 0.87 (0.47–1.59) 0.64                   |         | 0.43 (0.17–1.09) 0.08                      |         |
| **Polybrominated diphenyl ether**            |                                        |         |                                            |         |
| 47                                           | 1.79 (0.97–3.32) 0.06                   |         | 1.58 (0.75–3.33) 0.23                      |         |
| **Phthalate metabolites**                    |                                        |         |                                            |         |
| Mono-butyl phthalate (MBP)                   | 0.78 (0.43–1.45) 0.44                   |         | 0.66 (0.32–1.39) 0.28                      |         |
| Mono-benzyl phthalate (MBzP)                 | 1.22 (0.66–2.25) 0.52                   |         | 1.53 (0.73–3.22) 0.26                      |         |
| Mono-(2-ethyl-5-hydroxyhexyl) phthalate (MEHP)| 1.28 (0.69–2.35) 0.44                   |         | 1.50 (0.71–3.17) 0.28                      |         |
| Mono-(2-ethoxyhexyl) phthalate (MEHP)        | 2.16 (1.16–4.05) 0.02                   |         | 2.43 (1.12–5.24) 0.02                      |         |
| Mono-(2-ethyl-5-oxoehexyl) phthalate (MEOHP) | 0.95 (0.52–1.76) 0.88                   |         | 1.15 (0.54–2.44) 0.71                      |         |
| Mono-ethyl phthalate (MEP)                   | 0.71 (0.39–1.31) 0.28                   |         | 0.55 (0.26–1.18) 0.13                      |         |
| Mono-methyl phthalate (MMP)                  | 0.95 (0.52–1.76) 0.88                   |         | 0.86 (0.40–1.85) 0.70                      |         |

Approximated concentrations at the 75th percentile (38.4 ng/g lipid; 95% CI 31.8–46.4) of the US sample (22). BDE-47 concentrations measured in 50 serum samples collected in the 1990s from Laotian women living near San Francisco ranged from less than the LOD (10 ng/g lipid) to 511 ng/g lipid (mean 30.6 ng/g lipid; median 16.5 ng/g lipid) (33). The concentration of BDE-47 measured in a 2005 study of pooled serum from 100 discarded anonymous US samples collected in 2003 was 32.5 ng/g lipid (34).

A recent case–control study that measured concentrations of the 5 major PBDE congeners, including BDE-47, in adipose tissue collected from 134 women in the San Francisco Bay Area found no association between exposure to PBDEs and breast cancer (21). In that study population, the median concentration of BDE-47 measured in adipose was 22.0 ng/g lipid. For case women, the concentration was 18.83 ng/g lipid, and for control women, it was 26.85 ng/g lipid. The authors state that these adipose concentrations of PBDE were among the highest ever reported. The GM concentrations of BDE-47 measured in serum in our study population (30.4 ng/g lipid) and among our case women (38.8 ng/g lipid, respectively) were higher than concentrations reported in adipose.

The results of our study suggest a possible association between breast cancer and BDE-47 (OR 1.79, p = 0.06). However, the association is not supported in a multivariable analysis of BDE-47 with other known risk factors including age, menopause status, and having a relative with breast cancer. Our data seem to show a protective effect from exposure to p,p′-DDT. This association has not been reported previously. It is possible that the result is spurious, and should be interpreted with care.

No reports describing phthalate exposure among AN people were found in the literature. Our results suggested an association between urinary concentrations dichotomized at the median of the phthalate metabolite MEHP and breast cancer, an association that held in both univariate and multivariable analyses. However, MEHP is a minor metabolite of DEHP, and is the metabolite from which all others are formed. Therefore, we would expect to see associations with the other metabolites as well. The fact that associations were not found among the other DEHP metabolites may suggest the association between MEHP and breast cancer is a chance finding.
Table IV. Geometric mean serum concentrations (ng/g lipid) of selected persistent pesticides, polychlorinated biphenyls (PCB), and polybrominated diphenyl ethers (PBDE), and urinary concentrations (µg/g creatinine) of phthalate metabolites in a population of Alaska Native women with specific tumour hormone receptor status – 1999–2002

| Persistent pesticides | Geometric mean (95% confidence interval) | ER+/PR+ cases (n = 40) | ER–/PR– cases (n = 13) | p |
|-----------------------|------------------------------------------|------------------------|------------------------|---|
| p,p’-Dichlorodiphenylethylene (DDE) | 3,103 (2,231–4,316) | 4,273 (2,566–7,115) | 0.31 |
| Hexachlorobenzene | 290 (200–420) | 441 (240–804) | 0.25 |
| Mirex | 26.8 (18.7–38.5) | 40.9 (18.2–91.8) | 0.27 |
| Oxychlordane | 166 (113–242) | 273 (134–556) | 0.19 |
| β-Hexachlorocyclohexane | 110 (74–162) | 185 (103–334) | 0.16 |
| γ-Hexachlorocyclohexane | 7.92 (7.24–8.76) | 8.58 (6.69–11.02) | 0.49 |
| o,p’-Dichlorodiphenyltrichloroethane (DDT) | 13.9 (11.3–17.1) | 17.0 (11.7–24.5) | 0.35 |
| p,p’-DDT | 49.4 (37.0–66.7) | 66.0 (40.9–108) | 0.32 |
| trans-Nonachlor | 217 (141–337) | 416 (192–907) | 0.13 |
| Polychlorinated biphenyls | | | |
| 138/158 | 327 (237–455) | 503 (270–925) | 0.19 |
| 153 | 424 (296–614) | 672 (340–1326) | 0.22 |
| 180 | 273 (202–372) | 354 (198–626) | 0.41 |
| Polychlorinated diphenyl ethers | | | |
| 47 | 34.1 (21.3–54.1) | 51.9 (23.1–116.8) | 0.36 |
| Phthalate metabolites | | | |
| Mono-butyl phthalate (MBP) | 15.3 (9.0–26.1) | 12.8 (6.7–24.8) | 0.71 |
| Mono-benzyl phthalate (MBzP) | 8.3 (5.4–12.8) | 6.2 (3.1–12.7) | 0.51 |
| Mono-(2-ethyl-5-hydroxyhexyl) phthalate (MEHP) | 35.5 (24.5–51.4) | 49.4 (20.7–119.1) | 0.40 |
| Mono-(2-ethylhexyl) phthalate (MEHP) | 4.6 (2.8–7.6) | 11.1 (3.7–33.8) | 0.10 |
| Mono-(2-ethyl-5-oxohexyl) phthalate (MEOHP) | 23.3 (16.8–32.5) | 36.6 (16.1–83.9) | 0.21 |
| Mono-ethyl phthalate (MEP) | 49.6 (32.3–76.1) | 47.7 (23.0–98.9) | 0.93 |
| Mono-methyl phthalate (MMP) | 1.54 (1.1–2.2) | 1.48 (0.5–4.4) | 0.95 |

Furthermore, the concentrations of phthalate metabolites were measured in a single spot urine sample. Urinary concentrations of some phthalate metabolites, including MEHP, can be highly variable over time (35). Although this variability could result in exposure misclassification, it would likely be non-differential with respect to the outcome examined, significantly reducing our ability to observe an effect of DEHP on cancer risk. For these reasons, the findings regarding MEHP should be interpreted with caution.

We found only one study that reported an association between exposure to phthalates and breast cancer (23). A case–control study of more than 450 Mexican women found that case women had higher concentrations (169.58 µg/g creatinine) of the phthalate metabolite MEP than did control women (106.78 µg/g creatinine) – concentrations that were almost double those of MEP among our study population – and that MEP concentrations were associated with breast cancer.

A recent in vitro study implicated phthalates as possible potentiators in breast carcinogenesis (36). One study examined the effect of 2 phthalates – neither of them MEHP – on ER-negative breast cancer cells. The authors reported that the exposure induced cell proliferation, migration, invasion, and tumour formation (37). Animal studies have demonstrated metabolic differences between species, necessitating human studies to determine health effects (22,29). Temporal differences in human exposure, inconsistent study design, and differing methods, however, appear to account for wide variability in findings reported from studies investigating human health effects of phthalate exposure (22,35,38).

It is widely accepted that oestrogen is an important contributor to breast cancer development and progression (39), an effect demonstrated best in studies of breast cancer among males; men who develop breast cancer have higher than normal levels of oestrogen (40). Reproduction-related characteristics, such as early age at menarche, later age at bearing first child, and postmenopausal obesity are associated with ER+ tumours, which respond to treatment with hormone therapy (41,42). ER– tumours are not influenced by oestrogen and do not respond to treatment by hormone therapy, are associated with higher mortality (43), and are believed by some to have different promoters
and aetiologies than ER+ tumours (41,44,45). An ecologic study based on county level breast cancer and exposure data concluded that ER+ tumours could be influenced by environmental factors with endocrine disrupting capabilities (44). Hoyer et al., in a nested case–control study of Danish women, concluded the opposite: that exposure to potential estrogenic organochlorines does not lead to developing ER+ breast cancer type. This study further reported an association between dieldrin and incidence of ER– tumours (46). Although the differences were not statistically significant, in our study, women with ER–PR– tumours type had consistently higher concentrations of the persistent pesticides we measured, than ER+/PR+ women.

Limitations of this study included a small sample size, which may have decreased our power to detect statistical differences, and the inability to include all potential confounders (e.g. BMI) in the analyses. Another limitation is the potential exposure misclassification due to the short half-life of phthalates, which can result in high variability of phthalate metabolites, particularly MEHP (35). However, any residual misclassification would be non-differential and bias results towards the null. Furthermore, our controls were women with benign breast disease. It is possible that one or more of the environmental chemicals we measured could be associated with benign breast disease, along with breast cancer. This could have resulted in some of our null findings. Future research should attempt to measure environmental chemical exposures in the general AN population.

Conclusion
AN women may be exposed to types and concentrations of environmental chemicals that are different from other populations of women in the U.S. Studies of DDT or PCBs have not demonstrated with any certainty that exposure to these chemicals results in increased risk of developing breast cancer. The health effects of exposure to relatively new environmental chemicals, such as PBDE and phthalates, are not well understood. The biologic plausibility exists for these chemicals to affect reproductive and other hormonally-regulated systems, as they are known endocrine disruptors. Larger population-based studies of AN people would provide more statistical power to better determine possible association between exposure to contaminants, in particular those with endocrine disrupting capabilities, and the risk of breast cancer.

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