

Biomarkers of Insecticide Exposure and Asthma in Children: A National Health and Nutrition Examination Survey (NHANES) 1999–2008 Analysis

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Pesticide exposure is a potential risk factor for increased asthma prevalence among children. The authors used National Health and Nutrition Examination Survey (1999–2008) biomarker data to evaluate dialkylphosphate (DAP) urinary concentrations, serum dichlorodiphenyldichloroethylene (DDE), and asthma among school-aged children (Mexican American, Non-Hispanic Black, Non-Hispanic White). Poisson logistic regression included age, sex, nativity, poverty index ratio, tobacco smoke exposure, and body mass index covariates. No association was found between DAP ($N = 2,777$) and asthma outcomes; adverse effect of DDE ($N = 940$) was suggested for Current Wheeze. Subgroup analyses identified positive associations with some asthma outcomes among Non-Hispanic Blacks, whereas inverse associations were identified among Mexican Americans. Results support previous associations observed among children's DDE exposure and wheeze. Characterization of risk factors for pesticide exposure and disease recognition among Mexican Americans is needed.

Keywords: asthma, children, DDE, epidemiology, pesticides

Introduction

There has been an increase in the asthma prevalence rate among US children during the past 30 years.^{1–3} Children have a higher asthma prevalence rate than adults, and low-income racial and ethnic groups continue to be disproportionately affected.^{4–6} The causes of asthma and the reasons for its increased occurrence have remained little understood⁷; however, a number of well-established environmental risk factors have been identified for its exacerbation in individuals.

The role of nonpersistent and bioaccumulative pesticide exposures in the development of asthma and the exacerbation of this disease has received attention due to the widespread and continued use of pesticides in both urban^{8–12} and rural^{13,14} environments and biological plausibility.¹⁵ Few epidemiological studies have explored the relationship between specific pesticide chemical types and asthma or asthma-related symptoms

in children. Prospective studies have found associations between prenatal and breast milk exposures to organochlorine pesticides and asthma, wheeze, and lower respiratory tract infections in newborn and young children in Spain.^{16–19} In the Ontario Farm Family Health Study, researchers did not observe clear associations between self-reported maternal pesticide exposures during pregnancy and asthma outcomes of their offspring.²⁰ Studies of adults in agricultural settings, in contrast, have identified links between self-reported lifetime or current exposure to pesticides and respiratory health.^{21–23} We explored the hypothesis that pesticides are associated with asthma in school-aged children 7–16 years of age using population-based data of biomarker concentrations. Given the disparities in asthma outcomes and exposure to pesticides among subpopulations, effects by country of birth and race/ethnicity were also examined.

Methods

Data Source

We obtained demographic, asthma, early childhood health, and chemical biomarker data collected by the National

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Health and Nutritional Examination Surveys (NHANES). NHANES is conducted following a complex, multistage area sampling protocol to generate cross-sectional representative data of the civilian noninstitutionalized population living in the United States. To increase sample sizes, we merged several NHANES survey cycle data sets from 1999 to 2008. During the greater part of this time frame, Mexican Americans, non-Hispanic African Americans, low-income Non-Hispanic White Americans, and adolescents were oversampled for the purpose of generating sufficient sample sizes to derive reliable population-based estimates.²⁴

Asthma and Asthma Symptoms

Recognizing that the likelihood of a clinical diagnosis or self-recognition of asthma may vary among subpopulations,^{25–27} we examined 3 dichotomous asthma outcome variables derived from NHANES medical examination and respiratory health survey questionnaires: (1) “Has a doctor or other health professional ever told you that you have asthma?” (Ever Asthma); (2) “Do you still have asthma?” (Current Asthma); and (3) “In the past 12 months have you had wheezing or whistling in your chest?” (Current Wheeze).

Biomarkers of Exposure

The primary independent variables of interest were (1) common metabolites of organophosphate insecticides (dialkyl phosphates), a nonpersistent insecticide, and (2) dichlorodiphenyldichloroethylene (*p,p'*-DDE), the major metabolite of the organochlorine insecticide dichlorodiphenyltrichloroethane (DDT). NHANES participants 6 years of age and older submitted a whole-blood specimen and spot urine specimen. From the blood specimen, cotinine serum levels, a metabolite of tobacco smoke exposure and covariate of interest, were measured for all participants.

A randomly selected subset of participants 6 years and older had urine specimens evaluated for concentrations of the 6 possible dialkylphosphate (DAP) metabolites of organophosphate pesticides: dimethylphosphate (DMP), dimethylthiophosphate (DMTP), dimethyldithiophosphate (DMDTP), diethylphosphate (DEP), diethylthiophosphate (DETP), and diethyldithiophosphate (DEDTP). For the analysis of DAP, we summed the 6 organophosphate metabolite concentrations after converting metabolites to nanomolar units by dividing the analyte concentrations by their molecular weight and multiplying by 1000. We adjusted the estimates of urinary biomarkers by correcting each organophosphate metabolite for urinary creatinine before conversion and DAP aggregation.^{28,29} Because specific organophosphate urinary metabolites had different percentages of observations above the limit of detection, ranging from 29% to 70%, we summed dimethyl alkylphosphate pesticides and diethyl alkylphosphate separately to yield DMAP and DEAP concentrations, for sensitivity analyses.

For a separate, randomly selected subset of participants 12 years of age and older, DDE was measured in serum.

Nearly 100% of the DDE observations were above the limit of detection.

Covariates

Our a priori selection of available NHANES covariates for descriptive statistics focused on risk factors associated with pesticide exposure and asthma or its diagnosis. These covariates included demographics and socioeconomic status (age, sex, race/ethnicity, country of birth, poverty index ratio [PIR], health insurance coverage), body mass index (BMI) by age, and exposure to tobacco smoke (prenatal smoking, cotinine). We examined data of participants 6–11 and 12–15 years of age available in the DAP data set, and data of adolescents 12–15 years of age available in the DDE data set. To evaluate disparities by race/ethnicity, we conducted stratified analyses of Non-Hispanic Whites, Mexican Americans, and Non-Hispanic Blacks. Although NHANES collects data from other racial/ethnic groups including Other Latinos (who do not have Mexican American heritage but who identify as Hispanic or Latino), these groups could not be considered for analysis due to sample size considerations. We dichotomized country of birth (US-born, foreign-born) and health insurance coverage (yes, no). The PIR was dichotomized to generally represent families meeting or not meeting poverty eligibility guidelines for federal programs: ie, PIR <1.85 and PIR ≥1.85, respectively.³⁰ Variables representing exposure to tobacco smoke included (1) secondhand smoke based on the question “Does anyone who lives here smoke cigarettes, cigars, or pipes anywhere inside the home?”; (2) prenatal smoking: “Did the biological mother smoke at any time while she was pregnant?”; and (3) serum cotinine, a metabolite of nicotine, commonly used to assess exposure to tobacco smoke. BMI by age categories were derived based on clinical guidelines: underweight, <5th percentile; average, 5th–85th percentile; overweight, 85th–95th percentile; and obese, >95th percentile.

Data Analysis

We compared sample sizes, frequencies, and missing observations of asthma outcomes and covariates of the larger NHANES data set from which we derived our 2 nonoverlapping NHANES pesticide subsets. For each pesticide data set, we calculated asthma outcomes as well as geometric means of biomarkers by race/ethnicity.

The relatively high Ever Asthma prevalence rate (>10%) of this cross-sectional study prompted use of Poisson logistic regression with robust variance estimation.³¹ To select exposure percentile cutpoints reflective of changes in risk of asthma with increasing pesticide exposure levels, we used linear splines to model the shape of the dose-response trend.³² Ever Asthma had the highest prevalence rate and overlaps with Current Asthma and Current Wheeze; therefore, cutpoints identified for Ever Asthma were used to evaluate all health outcomes. This process identified 25th percentile as cutpoint for creatinine-corrected DAP and the 40th and 80th percentiles as biomarker cutpoints for DDE. We used these cutpoints to define levels of exposure for each pesticide.

We estimated the unadjusted and adjusted asthma prevalence ratios for increasing exposure levels using the 25th percentile for DAP and the 40th percentile for DDE as the referent categories. Adjusted prevalence rate ratio (PRR) models for the DAP analysis included sex, race/ethnicity, BMI (continuous), PIR prenatal smoking, and log cotinine (continuous). As the DDE analysis had a smaller sample sizes and number of cases, for these models we reduced the number of covariates and adjusted for sex, BMI and the poverty index ratio. Due to the smaller race/ethnicity-stratified sample sizes, particularly for the DDE data set, we further reduced the number of covariates by using bivariate regression to select variables with the greatest influence in our sample. The stratified DAP models were adjusted for sex, cotinine, and BMI; the stratified analysis of DDE included BMI as a covariate. Statistical tests were 2-sided and our threshold for statistical significance was $p < .025$ and for marginal significance $.025 \leq p \leq .10$.

The geometric mean of DAP in the 1999–2001 cohort of participants was significantly higher than in the larger analytical sample, so we evaluated the influence of results specific to survey cycle. We assessed the impact of tobacco smoke exposure on our results by removing participants with high cotinine exposure levels indicative of an active smoker.³³ In addition, for DAP metabolites, we evaluated the potential impact of fasting time, which may influence the individual measurements of creatinine. Because we could not include country of birth in the stratified multivariate analysis, we conducted separate analyses restricted to US-born participants. This effort focused on Mexican Americans, since they had the largest percentage of foreign-born participants in the DAP (20%) and the DDE (25%) pesticide data sets, whereas Non-Hispanic Blacks and Non-Hispanic Whites had roughly 3% foreign-born in both pesticide data sets. For these analyses, only BMI was included as a covariate.

We used Stata 10³⁴ for our analysis. NHANES survey sample weights were used in estimation of unadjusted and adjusted geometric means, prevalence rate ratios, standard error estimates, and 95% confidence intervals, as well as exposure percentiles.

Results

NHANES participants from 1999 to 2008 included 10,077 6–15-year-old Non-Hispanic Whites, Mexican Americans, and Non-Hispanic Blacks (Table 1). Thirty-six percent (3,622) 6–15-year-old participants submitted urine samples for measurement of DAP concentrations. With the exception of oversampling differences of adolescents, individuals who contributed urine samples for DAP analysis were similar to the larger NHANES participant group.

During the 1999–2004 time frame, 962 participants aged 12–15 years were randomly selected to submit a blood sample for evaluation of DDE. For the analysis of the relationship between pesticides and asthma, the DAP data set was reduced by 845 (23%), primarily due to the incomplete analysis of all 6 dialkylphosphate metabolites needed to generate a DAP vari-

able. The DDE sample size was reduced by 22 (2%), primarily due to missing prenatal smoking and BMI observations.

In both pesticide data sets, Non-Hispanic Blacks had the highest prevalence rates of Ever Asthma and Current Asthma compared with Non-Hispanic Whites and Mexican Americans (Table 2). With the exception of Current Wheeze in the DDE data set, Non-Hispanic Blacks also had the highest estimates of Current Wheeze. There were some statistical differences of estimated uncorrected geometric mean DAP pesticide concentrations by race ethnicity; however, there were no differences in the creatine-corrected estimates (Table 2). The DDE geometric mean among Mexican Americans was roughly 3 times higher than those of Non-Hispanic Whites and Non-Hispanic Blacks. Mexican Americans also had the lowest geometric mean levels of cotinine in both data sets: Non-Hispanic Whites had cotinine levels roughly 3 times higher than Mexican Americans, and Non-Hispanic Blacks had estimates 4 times higher.

Unadjusted and adjusted asthma outcome prevalence rate ratios by increasing urinary DAP concentration percentiles were largely null, with point estimates generally greater than 1.0 for all participants (Tables 3a and 3b). Sensitivity analyses of DMAP and DEAP revealed similar null results (data in Appendices A and B). In stratified analysis, among Non-Hispanic Blacks, a positive trend was suggested for Current Wheeze among 6–11-year-old and Ever and Current Asthma among 12–15-year-olds. In contrast, an inverse trend was suggested for Ever and Current Asthma and Current Wheeze among Mexican American adolescents.

Increasing percentiles of serum DDE were associated with increased Current Wheeze but not Ever or Current Asthma (Table 4). Subgroup analyses were limited by small sample size but supported this association among Non-Hispanic Whites and Non-Hispanic Blacks. In contrast, an inverse effect among Mexican Americans was shown for both Current Asthma and Current Wheeze.

Our sensitivity analyses addressing survey cycle influence, number of hours fasted since specimen collection, and removing individuals with high body burden of cotinine did not alter the patterns reported in Tables 3a and 3b. Additional sensitivity analyses focused on US-born Mexican Americans. Increasing DAP and of DDE exposure levels did not differ from our core analyses (data available in Appendices A and B).

Comment

Overall, this study revealed no clear associations between asthma and biomarkers of DAP or DDT insecticides among school-aged children in the United States.

Although the analyses of asthma outcomes and urinary metabolites of DAP were largely null overall, subgroup analyses demonstrated marginally significant positive trends for Non-Hispanic Blacks for Current Wheeze among 6–11-year-olds, and Ever and Current Asthma among adolescents. Contradictory, marginally significant inverse trends were also

Table 1. Demographic Characteristics and Asthma Outcomes of All Participants 6–15 Years of Age, and by Pesticide Data Set; NHANES 1999–2008

Characteristic	1999–2008 6–15 years of age				1999–2004 12–15 years of age	
	All NHANES (<i>N</i> = 10,077)		OP data set (<i>N</i> = 3,622)		DDT data set [‡] (<i>N</i> = 962)	
	<i>n</i>	(%)	<i>n</i>	(%)	<i>n</i>	(%)
Age*						
6–11	5,164	(51.2)	2,027	(56.0)	No data	
12–15	4,913	(48.8)	1,595	(44.0)	962	(100)
Sex*						
Female	5,085	(50.5)	1,862	(51.4)	502	(52.2)
Male	4,992	(49.5)	1,760	(48.6)	460	(47.8)
Race*						
Non-Hispanic White	2,982	(29.6)	1,073	(29.6)	261	(27.1)
Mexican American	3,607	(35.8)	1,295	(35.8)	380	(39.5)
Non-Hispanic Black	3,488	(34.6)	1,254	(34.6)	321	(33.4)
Country of birth						
Mexico/Other	887	(8.8)	331	(9.1)	118	(12.3)
USA	9,188	(91.2)	3,291	(90.9)	844	(87.7)
Missing/refused	2	(0.0)				
BMI—for age**						
Underweight	230	(2.3)	76	(2.1)	9	(0.9)
Average	5,571	(55.3)	2,099	(58.0)	569	(59.1)
Overweight	1,673	(16.6)	625	(17.3)	160	(16.6)
Obese	2,178	(21.6)	781	(21.6)	213	(22.1)
Missing/refused	425	(4.2)	41	(1.1)	11	(1.1)
Poverty index ratio (PIR)*						
<1.85	5,194	(51.5)	1,882	(52.0)	496	(51.6)
1.85–5	4,883	(48.5)	1,740	(48.0)	466	(48.4)
Health insurance						
Noninsured	1,627	(16.1)	597	(16.5)	190	(19.8)
Insured	8,337	(82.7)	2,990	(82.6)	767	(79.7)
Missing/refused	113	(1.1)	35	(1.0)	5	(0.5)
Secondhand smoke						
No	7,935	(78.7)	2,870	(79.2)	727	(75.6)
Yes	2,024	(20.1)	717	(19.8)	231	(24.0)
Missing/refused	118	(1.2)	35	(1.0)	4	(0.4)
Prenatal smoking						
No	8,464	(84.0)	3,052	(84.3)	802	(83.4)
Yes	1,472	(14.6)	525	(14.5)	150	(15.6)
Missing/refused	141	(1.4)	45	(1.2)	10	(1.0)

*No missing/refused.

**Underweight (BMI <5th percentile), average (BMI 5th–85th percentile), overweight (BMI 85th–95th percentile), obese (BMI >95th percentile).

[‡]NHANES 1999–2004 available DDT biomarker data.

identified among Mexican American adolescents for Ever and Current Asthma.

Among our multiple analyses, we observed suggestive evidence for an association of increased DDE exposure with Current Wheeze; however, confidence intervals were wide. There was some suggested adverse impact of this pesticide on parent-reported Current Asthma in Non-Hispanic Blacks as well. These findings were in contrast to the significant inverse trends identified in the subgroup analyses of Current Asthma and Current Wheeze among Mexican Americans who had the highest body burdens measured for this DDT metabolite.

Exposure to DAP and DDT is widespread and variable in US children.³⁵ We observed marked higher metabolite levels among Mexican Americans relative to Non-Hispanic Whites and Non-Hispanic Blacks for DDE but not for the DAPs. In contrast to urinary DAP, which reflects exposure to these pesticides in the past 2–3 days primarily from dietary sources, serum DDE biomarker is a relatively good indicator of chronic, life-time, including prenatal, exposure due to its persistence in body tissues. Although the DDE body burden levels have significantly decreased since the 1970s, DDE serum concentrations did not decrease during the 1999–2004 time frame in

Table 2. Weighted Geometric Means (GM) of Crude and Creatinine-Corrected Dialkyl Phosphate Pesticide (DAP), Dichlorodiphenyldichloroethylene (*p,p'*-DDE), and Serum Cotinine, and Asthma Outcomes Stratified by Race/Ethnicity and Age Groups

DAP data set	Total		Non-Hispanic White (NHW)		Mexican American (MA)		Non-Hispanic Black (NHB)	
Ages 6–11	N = 1,484		N = 453		N = 517		N = 514	
Geometric means	GM	(95% CI)	GM	(95% CI)	GM	(95% CI)	GM	(95% CI)
Cotinine ^a (ng/mL)	0.12	(0.10–0.15)	0.13	(0.10–0.17)	0.04	(0.03–0.05)*	0.22	(0.18–0.27)*
DAP ^b (nmol/L)	72.3	(65.4–80.1)	71.1	(62.2–81.3)	65.5	(58.3–73.6)§	86.0	(72.0–103)§
DAP creatinine corrected (nmol/g)	87.7	(79.9–96.3)	87.7	(77.7–98.9)	88.2	(78.1–100)	87.3	(73.8–103)
Asthma outcomes ^c	PR ^d	(95% CI)	PR	(95% CI)	PR	(95% CI)	PR	(95% CI)
Ever Asthma ^a	14.5	(11.8–17.1)	13.3	(9.6–17.1)	10.6	(7.3–14.0)	22.9	(18.7–27.2)*
Current Asthma ^a	9.2	(7.5–10.8)	8.3	(6.0–10.6)	5.2	(3.0–7.4)§	16.5	(12.6–20.4)*
Current Wheeze ^a	11.9	(9.4–14.4)	12.4	(9.0–15.8)	6.6	(4.1–9.2)*	14.6	(11.1–18.2)
Ages 12–15	N = 1,239		N = 372		N = 480		N = 441	
Geometric means	GM	(95% CI)	GM	(95% CI)	GM	(95% CI)	GM	(95% CI)
Cotinine ^a (ng/mL)	0.13	(0.10–0.17)	0.14	(0.10–0.20)	0.05	(0.04–0.06)*	0.23	(0.18–0.29)*
DAP ^a (nmol/L)	73.9	(66.5–82.1)	74.8	(64.7–86.5)	61.3	(54.0–69.6)§	81.0	(71.1–92.4)§
DAP creatinine corrected (nmol/g)	57.7	(51.4–64.7)	60.0	(51.2–70.3)	53.0	(45.7–61.5)	52.0	(45.5–59.5)
Asthma outcomes	PR	(95% CI)	PR	(95% CI)	PR	(95% CI)	PR	(95% CI)
Ever Asthma	17.4	(14.3–20.6)	17.4	(13.5–21.4)	14.9	(10.2–19.6)	19.3	(14.6–23.9)
Current Asthma ^b	11.0	(8.4–13.6)	10.7	(7.5–14.0)	9.0	(5.5–12.5)§	13.6	(9.4–17.9)*
Current Wheeze	11.4	(10.0–14.1)	11.2	(7.7–14.7)	10.3	(6.6–14.1)*	12.8	(9.4–16.1)
DDE data set								
Ages 12–15	N = 940		N = 252		N = 371		N = 317	
Geometric means	GM	(95% CI)	GM	(95% CI)	GM	(95% CI)	GM	(95% CI)
Cotinine ^a (ng/mL)	0.18	(0.13–0.25)	0.19	(0.13–0.3)	0.07	(0.05–0.09)*	0.29	(0.22–0.37)
<i>p,p'</i> -DDE ^a (ng/g)	105	(93.0–118)	91.8	(79.9–105)	261	(224–306)	88.5	(80.8–97.0)
Asthma outcomes	PR	(95% CI)	PR	(95% CI)	PR	(95% CI)	PR	(95% CI)
Ever Asthma	12.5	(9.2–15.7)	10.5	(6.4–14.9)	14.4	(10.2–18.6)	18.6	(13.7–23.6)*
Current Asthma	8.4	(5.7–11.1)	7.8	(4.3–11.4)	9.2	(5.4–13.0)	9.9	(5.4–13.0)
Current Wheeze	10.4	(7.5–13.2)	11.0	(7.1–14.9)	9.3	(5.0–13.6)	8.6	(5.3–12.0)

^aStatistically significant difference between MA and NHB.^bMarginally statistically significant differences between MA and NHB.^cParent-reported asthma outcome.^dPrevalence rate.**p* ≤ .025. Statistically significant difference compared with NHW.§.025 < *p* ≤ .10. Marginally statistically significant difference compared with NHW.

which NHANES samples were collected and analyzed. In addition, the DDE geometric mean reported here is similar to estimates reported in a study evaluating the association between prenatal DDE exposure and respiratory infection and wheeze.¹⁹ The significantly higher geometric mean of DDE concentrations in serum of Mexican American children likely reflects the use of DDT in Mexico, which banned its use for agricultural purposes in 1991 but continued its use for malaria vector control until it was phased out in 1997.³⁶

There are few relevant prior epidemiological data on childhood asthma and DAP exposure. Several studies have evaluated possible mechanisms of exposure to DAP and the development of asthma or allergens.^{15,37} For example, the ongoing longitudinal birth cohort in the Salinas Valley, Califor-

nia, found that mothers who worked in an agricultural region where organophosphate pesticide use is relatively high were more likely to give birth to infants with elevated Th2 cytokines associated with the development of allergic diseases, including asthma, and the diagnosis of wheeze at 24 months of age.³⁸

In our cross-sectional analysis of DAP metabolites in school-aged children, we found no evidence of an increased risk of asthma and surprisingly observed an inverse trend related to Ever and Current Asthma among Mexican Americans in contrast to a positive trend among Non-Hispanic Blacks.

In the modest literature on DDT and childhood respiratory health, a cross-sectional study in Germany of 343 children 7–10 years of age found DDE biomarker exposure levels

Table 3a. Asthma Outcome Prevalence Rate Ratios by Creatinine-Corrected Dialkyl Phosphate Pesticide (DAP) Exposure Percentiles Among 6–11-Year-Olds, Stratified by Race/Ethnicity, NHANES 1999–2008

Percentiles	Ranges	All participants (N = 1,484)		Non-Hispanic White (N = 453)		Mexican American (N = 517)		Non-Hispanic Black (N = 514)	
		n	(%)	n	(%)	n	(%)	n	(%)
<25th percentile	3–30	357	(24)	113	(25)	120	(23)	124	(24)
25th–50th percentile	31–84	387	(26)	109	(24)	144	(28)	134	(26)
50th–75th percentile	85–216	365	(25)	113	(25)	127	(25)	125	(24)
>75th percentile	217–6,193	376	(25)	118	(26)	126	(24)	131	(25)
Respiratory morbidity	Cases ^a	UNADJ PRR ^b (95% CI)	ADJ ^c PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)
Ever Asthma									
<25th percentile	55	1.00 Referent	1.00 Referent	13	1.25 Referent	15	1.00 Referent	27	1.00 Referent
25th–50th percentile	57	1.08 (0.56–2.07)	1.19 (0.65–2.18)	14	2.15 (0.46–3.37)	13	1.07 (0.51–2.24)	30	1.22 (0.73–2.04)
50th–75th percentile	58	1.59 (0.87–2.90)	1.71 (0.97–2.99) [§]	20	1.04 (0.88–5.27) [§]	13	1.11 (0.43–2.84)	25	1.17 (0.75–1.83)
>75th percentile	62	1.05 (0.54–2.05)	1.16 (0.62–2.17)	13	0.73 (0.36–3.02)	15	1.53 (0.65–3.56)	34	1.30 (0.83–2.04)
Current Asthma									
<25th percentile	32	1.00 Referent	1.00 Referent	6	1.00 Referent	6	1.00 Referent	20	1.00 Referent
25th–50th percentile	38	1.34 (0.56–2.84)	1.49 (0.72–3.06)	9	1.81 (0.55–6.02)	9	1.24 (0.38–4.03)	20	1.18 (0.68–2.03)
50th–75th percentile	36	1.77 (0.77–3.23) [§]	1.89 (1.01–3.53) [§]	13	2.50 (0.89–7.03)	5	0.70 (0.16–3.12)	18	1.42 (0.87–2.32)
>75th percentile	40	1.29 (0.45–2.46)	1.42 (0.64–3.16)	9	1.41 (0.37–5.37)	9	1.65 (0.47–5.80)	22	1.36 (0.77–2.39)
Current Wheeze									
<25th percentile	36	1.00 Referent	1.00 Referent	11	1.00 Referent	7	1.00 Referent	18	1.00 Referent [†]
25th–50th percentile	40	1.08 (0.61–1.91)	1.07 (0.60–1.92)	13	1.09 (0.48–2.48)	10	0.92 (0.31–2.66)	17	1.16 (0.60–2.24)
50th–75th percentile	38	1.26 (0.71–2.24)	1.24 (0.70–2.23)	16	1.35 (0.59–3.09)	8	0.78 (0.25–2.45)	14	1.21 (0.64–2.30)
>75th percentile	47	1.45 (0.86–2.42)	1.43 (0.83–2.46)	16	1.45 (0.74–2.87)	9	1.45 (0.49–4.33)	22	1.54 (0.88–2.68)

^aParent-reported Asthma outcome.

^bPrevalence rate ratios.

^cPrevalence rate ratios adjusted for sex, race/ethnicity, body mass index, poverty income ratio, prenatal smoking, and cotinine.

^dStratified prevalence rate ratios adjusted for sex, body mass index, and cotinine.

[§].025 < p ≤ .10. Marginally statistically significant.

[†].025 < trend p ≤ .10. Marginally statistically significant.

Table 3b. Asthma Outcome Prevalence Rate Ratios by Creatinine-Corrected Dialkyl Phosphate Pesticide (DAP) Exposure Percentiles Among 12–13-Year-Olds, Stratified by Race/Ethnicity; NHANES 1999–2008

Percentiles	Ranges	All participants (<i>N</i> = 1,293)		Non-Hispanic White (<i>N</i> = 372)		Mexican American (<i>N</i> = 480)		Non-Hispanic Black (<i>N</i> = 441)	
		<i>n</i>	(%)	<i>n</i>	(%)	<i>n</i>	(%)	<i>n</i>	(%)
<25th percentile	2–21	329	(25)	95	(26)	118	(25)	116	(26)
25th–50th percentile	22–55	350	(27)	94	(25)	133	(28)	123	(28)
50th–75th percentile	56–146	331	(26)	94	(25)	118	(25)	119	(27)
>75th percentile	147–7,876	283	(22)	89	(24)	111	(23)	83	(19)
Respiratory morbidity	Cases ^a	UNADJ PRR ^b (95% CI)	ADJ ^c PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)
Ever Asthma									
<25th percentile	55	1.00 Referent	1.00 Referent	13	1.00 Referent	21	1.00 Referent†	21	1.00 Referent†
25th–50th percentile	51	1.19 (0.73–1.94)	1.14 (0.69–1.90)	16	1.32 (0.63–2.79)	15	0.91 (0.43–1.92)	20	0.98 (0.54–1.75)
50th–75th percentile	56	1.43 (1.01–2.03)§	1.44 (1.01–2.06)§	19	1.66 (0.95–2.89)§	17	0.88 (0.52–1.50)	20	1.37 (0.75–2.50)
>75th percentile	49	1.17 (0.66–2.08)	1.20 (0.66–2.20)	16	1.30 (0.55–3.06)	11	0.59 (0.30–1.16)	22	1.66 (0.97–2.84)§
Current Asthma									
<25th percentile	37	1.00 Referent	1.00 Referent	9	1.00 Referent	12	1.00 Referent†	16	1.00 Referent†
25th–50th percentile	31	1.09 (0.60–2.01)	1.04 (0.54–1.99)	10	1.30 (0.53–3.22)	10	1.00 (0.39–2.51)	11	0.69 (0.31–1.51)
50th–75th percentile	38	1.36 (0.72–2.57)	1.33 (0.74–2.40)	13	1.68 (0.65–4.34)	10	0.73 (0.34–1.57)	15	1.18 (0.58–2.43)
>75th percentile	33	1.11 (0.57–2.14)	1.12 (0.59–2.12)	10	1.24 (0.49–3.12)	7	0.45 (0.17–1.18)	16	1.59 (0.86–2.93)
Current Wheeze									
<25th percentile	34	1.00 Referent	1.00 Referent	10	1.00 Referent	11	1.00 Referent	13	1.00 Referent
25th–50th percentile	33	1.04 (0.53–2.02)	1.06 (0.54–2.11)	12	1.15 (0.44–3.02)	11	1.15 (0.48–2.77)	10	0.57 (0.25–1.32)
50th–75th percentile	36	1.06 (0.57–1.97)	1.11 (0.58–2.11)	12	1.18 (0.45–3.07)	11	0.74 (0.32–1.71)	13	1.10 (0.57–2.14)
>75th percentile	34	1.08 (0.54–2.15)	1.15 (0.56–2.34)	10	1.13 (0.42–3.06)	9	0.59 (0.22–1.62)	15	1.77 (0.73–4.30)

^aParent-reported Asthma outcome.

^bPrevalence rate ratios.

^cPrevalence rate ratios adjusted for sex, race/ethnicity, body mass index, poverty income ratio, prenatal smoking, and cotinine.

^dStratified prevalence rate ratios adjusted for sex, body mass index, and cotinine.

§.025 < *p* ≤ .10. Marginally statistically significant.

†.025 < trend *p* ≤ .10. Marginally statistically significant.

Table 4. Adjusted Asthma Outcome Prevalence Rate Ratios by Dichlorodiphenyldichloroethylene (*p,p'*-DDE) Exposure Percentiles Among 12–15-Year-Olds, Stratified by Race/Ethnicity, NHANES 1999–2004

Percentiles	Ranges	All participants (<i>N</i> = 940)		Non-Hispanic White (<i>N</i> = 252)		Mexican American (<i>N</i> = 371)		Non-Hispanic Black (<i>N</i> = 317)	
		<i>n</i>	(%)	<i>n</i>	(%)	<i>n</i>	(%)	<i>n</i>	(%)
<40th percentile	3–31	282	(30)	113	(45)	28	(8)	141	(44)
40th–80th percentile	97–245	366	(39)	101	(40)	122	(33)	143	(45)
>80th percentile	200–16,200	292	(31)	38	(15)	221	(60)	33	(10)
Respiratory morbidity		UNADJ PRR ^b (95% CI)	ADJ ^c PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)	Cases ^a	ADJ ^d PRR (95% CI)
Ever Asthma									
<40th percentile	52	1.00 Referent	1.00 Referent	14	1.00 Referent	7	1.00 Referent	31	1.00 Referent
40th–80th percentile	41	0.67 (0.39–1.15)	0.63 (0.35–1.14)	9	0.67 (0.27–1.69)	11	0.47 (0.15–1.46)	21	0.56 (0.34–0.90)*
>80th percentile	40	0.93 (0.49–1.77)	0.81 (0.33–2.00)	5	1.09 (0.34–3.56)	29	0.43 (0.16–1.15) [§]	6	0.77 (0.37–1.61)
Current Asthma									
<40th percentile	33	1.00 Referent	1.00 Referent	12	1.00 Referent	5	1.00 Referent [‡]	16	1.00 Referent
40th–80th percentile	25	0.51 (0.25–1.03) [§]	0.47 (0.21–1.05) [§]	9	0.39 (0.12–1.24)	8	0.43 (0.13–1.46)	12	0.66 (0.31–1.41)
>80th percentile	25	0.86 (0.38–1.96)	0.78 (0.24–2.55)	7	0.83 (0.21–3.33)	16	0.31 (0.13–0.79)*	5	1.41 (0.56–3.52) [§]
Current Wheeze									
<40th percentile	26	1.00 Referent	1.00 Referent [†]	14	1.00 Referent	3	1.00 Referent [†]	11	1.00 Referent
40th–80th percentile	25	1.01 (0.53–1.96)	1.24 (0.64–2.40)	9	1.10 (0.46–2.62)	10	0.83 (0.19–3.55)	16	1.08 (0.48–2.45)
>80th percentile	23	1.15 (0.59–2.26)	1.79 (0.79–4.09)	5	2.01 (0.83–4.86)	14	0.43 (0.11–1.68)	2	0.72 (0.18–2.83)

^aParent-reported Asthma outcome.

^bPrevalence rate ratios.

^cPrevalence rate ratios adjusted for sex, race/ethnicity, US- vs foreign-born, body mass index, poverty income ratio, and cotinine.

^dStratified prevalence rate ratios adjusted for body mass index.

**p* ≤ .025. Statistically significant.

[§].025 < *p* ≤ .10. Marginally statistically significant.

[‡]Trend *p* ≤ .025. Statistically significant.

[†].025 < trend *p* ≤ .10. Marginally statistically significant.

to be a predictor of asthma.³⁷ A prospective cohort study of 452 children observed that higher metabolite concentrations in infant cord blood or breast milk are associated with wheezing and asthma at 4 years of age and diagnosed asthma at 6 years of age.^{16,17} Another prospective cohort study of 520 mother-infant pairs found that increasing maternal serum concentration of DDE during the first trimester was associated with a higher frequency of lower respiratory tract infection of their infants, which may be a risk factor for subsequent development of asthma.¹⁸ Another study of 1,455 mother-child pairs saw an association between prenatal DDE exposure and risk of wheezing at 14 months. In contrast, results of a small study ($N = 175$) of infants suggested a negative association between pre- and postnatal DDE exposure and respiratory infection.³⁹

Our data show some concordance in that Current Wheeze prevalence was more commonly reported among the children with higher body burdens as represented by DDE. However, the inverse effect in the Mexican American subgroup for Current Asthma and Current Wheeze with much higher concentrations overall is puzzling. Inverse trend results for Mexican American adolescents in the DAP analysis raises similar questions. This likely reflects differential exposure patterns and risk factors for complex multifactorial diseases and their diagnoses, such as asthma, among subgroups.

We considered the observed strong differential exposure to tobacco smoke (Table 2), a key asthma risk factor, in our 3 ethnic/racial subgroups. Sensitivity analyses restricted to individuals with lower cotinine levels or self-reported no tobacco exposure, however, did not change the racial/ethnic differences in asthma outcomes with higher body burdens of DDE or DAP.

Lastly, we attempted to address disparities in recognition and diagnosis of asthma and asthma-related symptoms by using several outcome measures (doctor asthma diagnosis, Current Asthma, Current Wheeze) and by considering health insurance and income covariates. There may still be disparities in recognition of asthma and wheeze among ethnic/racial subgroups that could be due to health care access,^{25,40,41} nativity status, acculturation, or English language proficiency.^{42–49} Undiagnosed frequent wheezing is an issue affecting all racial/ethnic groups.^{26,50} Although we did not have extensive acculturation data, our sensitivity analyses restricted to US-born Mexican Americans did not alter the result of the “protective” effect of increasing DAP and DDE on asthma outcomes.

Other uncontrolled confounding is a possibility. Studies have suggested that early life exposure to components in rural environments (endotoxin, raw milk) and multiple siblings are associated with lower asthma and allergic disease prevalence, the so-called “hygiene hypothesis.”^{51–53} The NHANES data set and secondary analysis nature of this study precluded adequate assessment of hygiene hypothesis–associated early life factors.

Although the small sample size in subgroup analyses makes it difficult to draw conclusions, the results suggests the importance of considering differences in exposure patterns and risk factors among ethnic/racial subgroups in future assessments of the relationship between asthma and DDT exposure.

Although these are novel data in terms of looking at specific pesticide biomarkers and asthma among school-aged children, limitations include the cross-sectional nature of the data that preclude addressing specific windows of exposure, reliance on self-report surveys to categorize health outcomes, and potential for residual confounding due to the complex factors associated with asthma, and sample size limitations among ethnic/racial subgroups.

Conclusion

We found no clear support for our hypothesis that urinary biomarkers of DAP are associated with increased risk of asthma and wheeze in school-aged children. We found some support for a previously shown adverse role of DDE on wheeze in children. Subgroup analyses revealed several negative associations—contrary to our hypothesis—particularly among the most highly exposed subgroup, Mexican Americans.

Interpretation of the subgroup findings is uncertain given the small sample size but suggests the need to further define asthma risk factors among Mexican Americans.

Future studies that can address potential socioeconomic or acculturation factors that influence the diagnosis of asthma and/or reporting of asthma symptoms among racial and ethnic subgroups of US children could help determine whether their lower asthma prevalence rates are reliable, or whether other factors play a role. Better understanding of the role of pesticide exposure on respiratory health and asthma among children will require studies that can employ longitudinal measurements of exposure and other influential factors well into school age.

References

1. Akinbami LJ, Moorman JE, Bailey C, et al. *Trends in Asthma Prevalence, Health Care Use, and Mortality in the United States, 2001–2010*. NCHS Data Brief No. 94. Hyattsville, MD: National Center for Health Statistics; 2012
2. Akinbami LJ. The state of childhood asthma, United States, 1980–2005. Advance data. *Vital Health Stat.* 2006;(381):1–24.
3. Moorman JE, Rudd RA, Johnson CA, et al. National Surveillance for Asthma—United States, 1980–2004. *Morb Mort Wkly Rep Surveill Summ.* 2007;56:1–54.
4. Brim SN, Rudd RA, Funk RH, Callahan DB. Asthma prevalence among US children in underrepresented minority populations: American Indian/Alaska Native, Chinese, Filipino, and Asian Indian. *Pediatrics.* 2008;122:e217–e222.
5. Moorman JE, Garbe PL, Sondik EJ. Status of childhood asthma in the United States, 1980–2007. *Pediatrics.* 2009;123(Suppl 3):S131–S145.
6. Moorman JE, Zahran H, Truman BI, Molla MT. Current asthma prevalence—United States, 2006–2008. In: CDC Health Disparities and Inequalities Report—United States, 2011. *Morb Mort Wkly Rep Surveill Summ.* 2011;60(Suppl):84–86.
7. Institute of Medicine, Committee on the Assessment of Asthma and Indoor Air. *Clearing the Air: Asthma and Indoor Air Exposures*. Washington, DC: National Academies Press; 2000.
8. Landrigan PJ, Claudio L, Markowitz SB, et al. Pesticides and inner-city children: exposures, risks, and prevention. *Environ Health Perspect.* 1999;107(Suppl 3):431–437.

9. Thier A, Enck J, Klossner C. Plagued by pesticides: an analysis of New York State's 1997 pesticide use and sales data. Available at: <http://www.eany.org/news/reports/172-plagued-by-pesticides>. Published 1998. Accessed May 2012.
10. Horton MK, Jacobson JB, McKelvey W, et al. Characterization of residential pest control products used in inner city communities in New York City. *J Expo Sci Environ Epidemiol*. 2011;21:291–301.
11. Whyatt RM, Barr DB, Camann DE, et al. Contemporary-use pesticides in personal air samples during pregnancy and blood samples at delivery among urban minority mothers and newborns. *Environ Health Perspect*. 2003;111:749–756.
12. Williams MK, Rundle A, Holmes D, et al. Changes in pest infestation levels, self-reported pesticide use, and permethrin exposure during pregnancy after the 2000–2001 U.S. Environmental Protection Agency restriction of organophosphates. *Environ Health Perspect*. 2008;116:1681–1688.
13. Castorina R, Bradman A, McKone TE, Barr DB, Harnly ME, Eskenazi B. Cumulative organophosphate pesticide exposure and risk assessment among pregnant women living in an agricultural community: a case study from the CHAMACOS cohort. *Environ Health Perspect*. 2003;111:1640–1648.
14. Fenske RA, Bradman A, Whyatt RM, Wolff MS, Barr DB. Lessons learned for the assessment of children's pesticide exposure: critical sampling and analytical issues for future studies. *Environ Health Perspect*. 2005;113:1455–1462.
15. Fryer AD, Lein PJ, Howard AS, Yost BL, Beckles RA, Jett DA. Mechanisms of organophosphate insecticide-induced airway hyperreactivity. *Am J Physiol Lung Cell Mol Physiol*. 2004;286:L963–L969.
16. Sunyer J, Garcia-Esteban R, Alvarez M, et al. DDE in mothers' blood during pregnancy and lower respiratory tract infections in their infants. *Epidemiology*. 2010;21:729–735.
17. Sunyer J, Torrent M, Garcia-Esteban R, et al. Early exposure to dichlorodiphenyldichloroethylene, breastfeeding and asthma at age six. *Clin Exp Allergy*. 2006;36:1236–1241.
18. Sunyer J, Torrent M, Munoz-Ortiz L, et al. Prenatal dichlorodiphenyldichloroethylene (DDE) and asthma in children. *Environ Health Perspect*. 2005;113:1787–1790.
19. Gascon M, Vrijheid M, Martinez D, et al.; Infancia y Medio Ambiente Environment and Childhood Project. Pre-natal exposure to dichlorodiphenyldichloroethylene and infant lower respiratory tract infections and wheeze. *Eur Respir J*. 2012;39:1188–1196.
20. Weselak M, Arbuckle TE, Wigle DT, Krewski D. In utero pesticide exposure and childhood morbidity. *Environ Res*. 2007;103:79–86.
21. Valcin M, Henneberger PK, Kullman GJ, et al. Chronic bronchitis among nonsmoking farm women in the agricultural health study. *J Occup Environ Med*. 2007;49:574–583.
22. Hoppin JA, Valcin M, Henneberger PK, et al. Pesticide use and chronic bronchitis among farmers in the agricultural health study. *Am J Ind Med*. 2007;50:969–979.
23. Chakraborty S, Mukherjee S, Ray MR, Roychoudhury S, Siddique S, Lahiri T. Chronic exposures to cholinesterase-inhibiting pesticides adversely affect respiratory health of agricultural workers in India. *J Occup Health*. 2009;51:488–497.
24. Centers for Disease Control and Prevention (CDC). *Third National Report on Human Exposure to Environmental Chemicals*. National Center for Environmental Health Publication No. 05–0570. Atlanta, GA: National Center for Environmental Health (NCEH); 2005.
25. Zambrana RE, Carter-Pokras O. Improving health insurance coverage for Latino children: a review of barriers, challenges and state strategies. *J Natl Med Assoc*. 2004;96:508–523.
26. Akinbami LJ, Rhodes JC, Lara M. Racial and ethnic differences in asthma diagnosis among children who wheeze. *Pediatrics*. 2005;115:1254–1260.
27. Eggleston PA, Diette G, Lipsett M, et al. Lessons learned for the study of childhood asthma from the centers for children's environmental health and disease prevention research. *Environ Health Perspect*. 2005;113:1430–1436.
28. Boeniger MF, Lowry LK, Rosenberg J. Interpretation of urine results used to assess chemical exposure with emphasis on creatinine adjustments: a review. *Am Ind Hyg Assoc J*. 1993;54:615–627.
29. Barr DB, Bravo R, Weerasekera G, et al. Concentrations of dialkyl phosphate metabolites of organophosphorus pesticides in the U.S. population. *Environ Health Perspect*. 2004;112:186–200.
30. US Census Bureau, Population Division, Fertility & Family Statistics Branch. Current Population Survey: Definitions and Explanations, How the Census Bureau Measures Poverty. Available at: <http://www.census.gov/hhes/www/poverty/about/overview/measure.html>. Published 2004. Accessed July 2013.
31. Thompson ML, Myers JE, Kriebel D. Prevalence odds ratio or prevalence ratio in the analysis of cross sectional data: what is to be done? *Occup Environ Med*. 1998;55:272–277.
32. Greenland S. Dose-response and trend analysis in epidemiology: alternatives to categorical analysis. *Epidemiology*. 1995;6:356–365.
33. Benowitz NL, Bernert JT, Caraballo RS, Holiday DB, Wang J. Optimal serum cotinine levels for distinguishing cigarette smokers and nonsmokers within different racial/ethnic groups in the United States between 1999 and 2004. *Am J Epidemiol*. 2009;169:236–248.
34. StataCorp. Stata Statistical Software: Release 10. 2007; College Station, TX: StataCorp.
35. Centers for Disease Control and Prevention (CDC). *Fourth National Report on Human Exposure to Environmental Chemicals*. Atlanta, GA: National Center for Environmental Health; 2009.
36. US Commission for Environmental Cooperation. North American Regional Action Plan on DDT. North American Working Group for the Sound Management of Chemicals. Task Force on DDT and Chlordane, June 1997. Available at: <http://www.cec.org/Page.asp?PageID=924&ContentID=1262>. Accessed May 2012.
37. Proskocil BJ, Bruun DA, Lorton JK, et al. Antigen sensitization influences organophosphorus pesticide-induced airway hyperreactivity. *Environ Health Perspect*. 2008;116:381–388.
38. Duramad P, Harley K, Lipsett M, et al. Early environmental exposures and intracellular Th1/Th2 cytokine profiles in 24-month-old children living in an agricultural area. *Environ Health Perspect*. 2006;114:1916–1922.
39. Karmaus W, Kuehr J, Kruse H. Infections and atopic disorders in childhood and organochlorine exposure. *Arch Environ Health*. 2001;56:485–492.
40. Glynn A, Thuvander A, Aune M, et al. Immune cell counts and risks of respiratory infections among infants exposed pre- and postnatally to organochlorine compounds: a prospective study. *Environ Health*. 2008; 62.
41. Freeman NC, Schneider D, McGarvey P. The relationship of health insurance to the diagnosis and management of asthma and respiratory problems in children in a predominantly Hispanic urban community. *Am J Public Health*. 2003;93:1316–1320.
42. Quinn K, Shalowitz MU, Berry CA. Racial and ethnic disparities in diagnosed and possible undiagnosed asthma among public-school children in Chicago. *Am J Public Health*. 2006;96:1599–1603.
43. Canino G, Koinis-Mitchell D, Ortega AN, McQuaid EL, Fritz GK, Alegria M. Asthma disparities in the prevalence, morbidity, and treatment of Latino children. *Soc Sci Med*. 2006;63:2926–2937.
44. Eldeirawi KM, Persky VW. Associations of acculturation and country of birth with asthma and wheezing in Mexican American youths. *J Asthma*. 2006;43:279–286.
45. Eldeirawi K, McConnell R, Furner S, et al. Associations of doctor-diagnosed asthma with immigration status, age at immigration, and length of residence in the United States in a sample of Mexican American School Children in Chicago. *J Asthma*. 2009;46:796–802.

46. Eldeirawi KM, Persky VW. Associations of physician-diagnosed asthma with country of residence in the first year of life and other immigration-related factors: Chicago asthma school study. *Ann Allergy Asthma Immunol.* 2007;99:236–243.
47. Subramanian SV, Jun HJ, Kawachi I, Wright RJ. Contribution of race/ethnicity and country of origin to variations in lifetime reported asthma: evidence for a nativity advantage. *Am J Public Health.* 2009;99:690–697.
48. Joseph SP, Borrell LN, Shapiro A. Self-reported lifetime asthma and nativity status in U.S. children and adolescents: results from the National Health and Nutrition Examination Survey 1999–2004. *J Health Care Poor Underserved.* 2010;21(2 Suppl): 125–139.
49. Mosnaim GS, Sadowski LS, Durazo-Arvizu RA, et al. Parental language and asthma among urban Hispanic children. *J Allergy Clin Immunol.* 2007;120:1160–1165.
50. Yeatts K, Davis KJ, Sotir M, Herget C, Shy C. Who gets diagnosed with asthma? Frequent wheeze among adolescents with and without a diagnosis of asthma. *Pediatrics.* 2003;111(5 Pt 1): 1046–1054.
51. Strachan DP. Family size, infection and atopy: the first decade of the “hygiene hypothesis”. *Thorax.* 2000;55(Suppl 1):S2–S10.
52. Eder W, von Mutius E. Hygiene hypothesis and endotoxin: what is the evidence? *Curr Opin Allergy Clin Immunol.* 2004;4:113–117.
53. Von Mutius E. Allergies, infections and the hygiene hypothesis—the epidemiological evidence. *Immunobiology.* 2007;212:433–439.

Appendix A. Adjusted Asthma Outcome Prevalence Rate Ratios by Creatinine-Corrected Pesticide Exposure Percentiles Among US-Born Mexican Americans

1. Dialkyl phosphate pesticide (DAP) exposure among 6–11-year-old US-born Mexican Americans; NHANES 1999–2008

Percentiles	Ranges	Mexican American (<i>N</i> = 432)	
		<i>n</i>	(%)
<25th percentile	2–31	101	(23)
25th–50th percentile	32–84	123	(28)
50th–75th percentile	85–216	109	(25)
>75th percentile	217–6,193	99	(23)
		Cases ^a	ADJ PRR ^b
			(95% CI)
Ever Asthma			
<25th percentile		14	1.0 Referent
25th–50th percentile		11	0.87 (0.40–1.90)
50th–75th percentile		12	1.01 (0.40–2.53)
>75th percentile		14	1.51 (0.65–3.47)
Current Asthma			
<25th percentile		6	1.1 Referent
25th–50th percentile		8	1.10 (0.34–3.57)
50th–75th percentile		5	0.67 (0.16–2.83)
>75th percentile		9	1.76 (0.53–5.87)
Current Wheeze			
<25th percentile		7	1.0 Referent
25th–50th percentile		9	0.81 (0.27–2.41)
50th–75th percentile		7	0.69 (0.22–2.14)
>75th percentile		9	1.55 (0.53–4.51)

2. Dialkyl phosphate pesticide (DAP) exposure among 12–15-year-old US-born Mexican Americans; NHANES 1999–2008

Percentiles	Ranges	Mexican American (<i>N</i> = 362)	
		<i>n</i>	(%)
<25th percentile	2–21	91	(25)
25th–50th percentile	22–55	103	(28)
50th–75th percentile	56–146	91	(25)
>75th percentile	147–5,028	77	(21)
		Cases ^a	ADJ PRR ^c
			(95% CI)
Ever Asthma			
<25th percentile		18	1.0 Referent
25th–50th percentile		13	0.83 (0.38–1.84)
50th–75th percentile		14	0.77 (0.45–1.34)
>75th percentile		10	0.54 (0.23–1.24)
Current Asthma			
<25th percentile		11	1.0 Referent†
25th–50th percentile		9	0.95 (0.37–2.49)
50th–75th percentile		9	0.72 (0.32–1.61)
>75th percentile		6	0.38 (0.12–1.21)§
Current Wheeze			
<25th percentile		10	1.0 Referent
25th–50th percentile		10	1.17 (0.45–3.04)
50th–75th percentile		10	0.75 (0.31–1.83)
>75th percentile		9	0.69 (0.25–1.96)

3. Dichlorodiphenyldichloroethylene (*p,p'*-DDE) exposure among 12–15-year-old US-born Mexican Americans; NHANES 1999–2004

Percentiles	Ranges	Mexican American (<i>N</i> = 278)	
		<i>n</i>	(%)
<40th percentile	20–91	36	(14)
40th–80th percentile	92–182	83	(32)
>80th percentile	184–3,230	159	(61)

(Continued on next page)

Appendix A. Adjusted Asthma Outcome Prevalence Rate Ratios by Creatinine-Corrected Pesticide Exposure Percentiles Among US-Born Mexican Americans (*Continued*)

	Cases ^a	ADJ PRR ^b	(95% CI)
Ever Asthma			
<40th percentile	8	1.0	Referent
40th-80th percentile	8	0.63	(0.17–2.37)
>80th percentile	23	0.52	(0.18–1.50)
Current Asthma			
<40th percentile	6	1.0	Referent†
40th-80th percentile	5	0.50	(0.09–2.70)
>80th percentile	15	0.40	(0.15–1.09) [§]
Current Wheeze			
<40th percentile	5	1.0	Referent†
40th-80th percentile	7	0.64	(0.13–3.17)
>80th percentile	14	0.40	(0.14–1.17) [§]

^aParent-reported asthma outcomes.^bPrevalence rate ratios adjusted for body mass index.^cPrevalence rate ratios adjusted for sex, body mass index, and cotinine.[§].025 < *p* ≤ .10. Marginally statistically significant.□.025 < trend *p* ≤ .10. Marginally statistically significant.

Appendix B. Unadjusted and Adjusted Asthma Outcome Prevalence Rate Ratios by Creatinine-Corrected Summed Dimethyl Alkylphosphate (DMAP) and Diethyl Alkylphosphate (DEAP) Pesticide Exposure Percentiles of Non-Hispanic White, Non-Hispanic Black, and Mexican Americans; NHANES 1999–2008

Percentiles	Ranges	N	Ever Asthma				Prevalence rate ratio (95% CI)			
			Current Asthma		Current Wheeze					
			Cases ^a	UNADJ PRR ^b	ADJ ^c PRR	Cases ^a	UNADJ PRR ^b	ADJ ^c PRR		
1. 6–11 years of age										
DMAP creatinine corrected (nmol/L) (N = 1,486)										
<25th percentile	2–17	382	62	1.00 Referent	1.00 Referent	38	1.00 Referent	1.00 Referent	39	1.00 Referent
25th–50th percentile	17–58	381	55	0.92 (0.49–1.73)	0.99 (0.54–1.80)	34	1.22 (0.61–2.46)	1.34 (0.70–2.58)	39	1.31 (0.75–2.31)
50th–75th percentile	59–167	354	53	1.34 (0.67–2.70)	1.47 (0.77–2.78)	34	1.53 (0.70–3.32)	1.69 (0.81–3.51)	36	1.30 (0.70–2.39)
>75th percentile	168–6,115	367	62	1.03 (0.54–1.95)	1.13 (0.63–2.05)	40	1.19 (0.53–2.65)	1.32 (0.61–2.82)	47	1.59 (0.92–2.74)
DEAP creatinine corrected (nmol/L) (N = 2,404)										
<25th percentile	0.5–4	564	54	1.00 Referent	1.00 Referent	30	1.00 Referent	1.00 Referent	35	1.00 Referent
25th–50th percentile	5–11	575	53	1.23 (0.74–2.07)	1.28 (0.78–2.12)	32	1.57 (0.79–3.13)	1.65 (0.83–3.25)	32	0.90 (0.44–1.83)
50th–75th percentile	12–39	676	65	1.06 (0.58–1.91)	1.11 (0.64–1.94)	43	1.54 (0.77–3.07)	1.59 (0.81–3.12)	50	1.11 (0.62–1.99)
>75th percentile	40–5,379	589	60	1.07 (0.60–1.92)	1.18 (0.69–2.03)	41	1.62 (0.82–3.20)	1.79 (0.93–3.44) [§]	44	1.02 (0.51–2.03)
2. 12–15 years of age										
DMAP creatinine corrected (nmol/L) (N = 1,293)										
<25th percentile	2–11	354	58	1.00 Referent	1.00 Referent	40	1.00 Referent	1.00 Referent	36	1.00 Referent
25th–50th percentile	12–36	345	50	0.87 (0.54–1.40)	0.89 (0.55–1.44)	30	0.74 (0.42–1.30)	0.73 (0.41–1.30)	33	1.01 (0.52–1.96)
50th–75th percentile	37–110	315	55	1.36 (0.84–2.21)	1.45 (0.88–2.39)	38	1.30 (0.73–2.31)	1.38 (0.80–2.40)	32	0.87 (0.43–1.76)
>75th percentile	111–7,875	279	48	1.22 (0.74–2.03)	1.35 (0.79–2.30)	31	1.03 (0.53–1.98)	1.12 (0.59–2.11)	36	1.05 (0.53–2.08)
DEAP creatinine corrected (nmol/L) (N = 1,293)										
<25th percentile	0.5–3	331	57	1.00 Referent	1.00 Referent	38	1.00 Referent	1.00 Referent	34	1.00 Referent
25th–50th percentile	4–9	300	58	0.89 (0.56–1.40)	0.90 (0.59–1.39)	38	1.11 (0.57–2.17)	1.12 (0.60–2.09)	37	1.32 (0.72–2.41)
50th–75th percentile	10–31	363	41	0.61 (0.37–1.01) [§]	0.62 (0.37–1.03) [§]	24	0.57 (0.30–1.11) [§]	0.57 (0.30–1.09) [§]	28	0.64 (0.32–1.29)
>75th percentile	32–4,414	299	55	1.05 (0.66–1.68)	1.07 (0.67–1.71)	39	1.29 (0.70–2.37)	1.28 (0.72–2.29)	38	1.49 (0.80–2.78)

^aParent-reported asthma outcomes.

^bPrevalence rate ratios.

^cPrevalence rate ratios adjusted for sex, race/ethnicity, mass index, poverty income ratio, prenatal smoking, and cotinine.

[§].025 < p ≤ .10. Marginally statistically significant.

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