Original Research Article

ENVIRONMENTAL EPIDEMIOLOGY

OPEN

Prenatal exposure to multiple organochlorine compounds and childhood body mass index

Elena Colicino[*,a], Katerina Margetaki[b,c], Damaskini Valvi[a], Nicolo Foppa Pedretti[a], Nikos Stratakis[d], Marina Vafeiadi[c], Theano Roumeliotaki[c], Soterios A. Kyrtopoulos[e], Hannu Kiviranta[f], Euripides G. Stephanou[g], Manolis Kogevinas[d,h,i], Rob McConnell[b], Kiros T. Berhane[j], Leda Chatzi[b], David V. Conti[a]

Background: Prenatal exposure to organochlorine compounds (OCs) has been associated with increased childhood body mass index (BMI); however, only a few studies have focused on longitudinal BMI trajectories, and none of them used multiple exposure mixture approaches.

Aim: To determine the association between *in-utero* exposure to eight OCs and childhood BMI measures (BMI and BMI z-score) at 4 years and their yearly change across 4–12 years of age in 279 Rhea child-mother dyads.

Methods: We applied three approaches: (1) linear mixed-effect regressions (LMR) to associate individual compounds with BMI measures; (2) Bayesian weighted quantile sum regressions (BWQSR) to provide an overall OC mixture association with BMI measures; and (3)Bayesian varying coefficient kernel machine regressions (BVCKMR) to model nonlinear and nonadditive associations.

Results: In the LMR, yearly change of BMI measures was consistently associated with a quartile increase in hexachlorobenzene (HCB) (estimate [95% Confidence or Credible interval] BMI: 0.10 [0.06, 0.14]; BMI z-score: 0.02 [0.01, 0.04]). BWQSR results showed that a quartile increase in mixture concentrations was associated with yearly increase of BMI measures (BMI: 0.10 [0.01, 0.18]; BMI z-score: 0.03 [0.003, 0.06]). In the BVCKMR, a quartile increase in dichlorodiphenyldichloroethylene concentrations was associated with higher BMI measures at 4 years (BMI: 0.33 [0.24, 0.43]; BMI z-score: 0.19 [0.15, 0.24]); whereas a quartile increase in HCB and polychlorinated biphenyls (PCB)-118 levels was positively associated with BMI measures yearly change (BMI: HCB:0.10 [0.07, 0.13], PCB-118:0.08 [0.04, 012]; BMI z-score: HCB:0.03 [0.02, 0.05], PCB-118:0.02 [0.002,04]). BVCKMR suggested that PCBs had nonlinear relationships with BMI measures, and HCB interacted with other compounds.

Conclusions: All analyses consistently demonstrated detrimental associations between prenatal OC exposures and childhood BMI measures.

Keywords: Chemical mixture; Outcome trajectories; Body mass index; Organochlorine compounds; Bayesian weighted quantile sum regressions; Bayesian varying coefficient kernel machine regressions

Introduction

The prevalence of overweight and obesity in children and adolescents has tripled since 1970, with 49% of US children and adolescents currently suffering from these conditions.[1,2] The prevalence of those conditions has also increased worldwide in recent decades.[3] More cases of overweight and obesity are beginning earlier, during childhood, and their severity and their prevalence are exacerbated during adolescence.[4] Overweight and obesity in childhood have immediate and long-term consequences on health. Among the short-term concerns, childhood obesity increases the risk of neurological, pulmonary, endocrine, and cardiometabolic disorders, including hyperlipidemia, hypertension, and abnormal glucose tolerance.[5–7] Overweight and obese children have an 80% chance of becoming obese adults, thus placing them at higher risk for chronic diseases and premature mortality in later life.[8] The prevalence of overweight and obese children show geographic variations owing to lifestyle, socioeconomic, and ethnic differences, but environmental influences have also been shown to contribute to this variation.[9–15]

Certain persistent organic pollutants, such as organochlorine compounds (OCs), are toxic lipophilic chemicals—used in agriculture, manufacturing, or industrial processes[16]—that

[a]*Department of Environmental Medicine and Public Health, Icahn School of Medicine at Mount Sinai, New York;* [b]*Department of Preventive Medicine, Keck School of Medicine, University of Southern California, Los Angeles, California;* [c]*Department of Social Medicine, Faculty of Medicine, University of Crete, Heraklion, Greece;* [d]*ISGlobal, Barcelona, Spain;* [e]*Institute of Chemical Biology, National Hellenic Research Foundation, Athens, Greece;* [f]*Department of Health Security, National Institute for Health and Welfare, Kuopio, Finland;* [g]*Environmental Chemical Processes Laboratory, Department of Chemistry, University of Crete, Heraklion, Greece;* [h]*Universitat Pompeu Fabra, Barcelona, Spain;* [i]*CIBER Epidemiología y Salud Pública, Barcelona, Spain; and* [j]*Department of Biostatistics, Mailman School of Public Health, Columbia University, New York.*

The authors declare that they have no conflicts of interest with regard to the content of this report.

During the preparation of this manuscript, E.C. was supported by the National Institute of Environmental Health Science (NIEHS): R01ES032242, 5U2CES026555-03 and P30ES023515. D.V. was supported by P30ES023515 and R21ES029328. D.V.C. was supported by P01CA196569. L.C. was supported by NIEHS (R21ES029681, R01ES030691, R01ES029944, R01ES030364, R21ES028903, and P30ES007048). N.S. has received funding from the Ministry of Science and Innovation and State Research Agency through the "Centro de Excelencia Severo Ochoa 2019-2023" Program (CEX2018-000806-S) and from IJC2020-043630-I financed by MCIN/AEI/10.13039/501100011033 and the European Union "NextGenerationEU/PRTR".

What this study adds

This is the first study using three distinct statistical methods, including two exposure mixture approaches, to show the relationship between *in-utero* exposure to organochlorine compounds (OCs) and body mass index (BMI) measures (BMI and BMI z-score) at 4 years and their yearly change from 4 to 12 years in 279 child-mother dyads from the Rhea cohort. All statistical models were consistent in showing a detrimental association between prenatal OC concentrations and yearly change in BMI measures, although only the model accommodating nonlinear and nonadditive associations consistently captured the potentially harmful role of OCs on BMI outcomes at age 4 years.

persist and accumulate in the environment for long periods, and they pass from one species to another up the food chain.[17] Although OC production and distribution were banned in Europe[19] and in the US,[20] OCs are still an environmental exposure concern. OCs are endocrine disruptors and can interfere with hormonally responsive tissue functions by dysregulating hormone signaling and cell function.[21–24] OCs affect the endocrine system and its dynamics at many stages of life, including during pregnancy.[25] Furthermore, the OCs' affinity for lipids has been proposed to trigger the onset and development of obesity. Higher prenatal exposure levels to OCs, including hexachlorobenzene (HCB), dichlorodiphenyldichloroethylene (DDE), and polychlorinated biphenyls (PCBs), have been linked to childhood body composition and obesity.[9–14,26] Our prior work has also shown that higher prenatal DDE and HCB exposures were individually associated with greater body mass index (BMI) z-score at 4 years.[27] However, only a few studies focused on longitudinal weight measures,[26] and none of them used approaches that capture joint exposure to more than one OC.

OCs rarely occur as a single compound, and prior studies that analyzed a single chemical at a time may have had limited ability to detect joint associations. To address the challenge of analyzing several compounds jointly, a few methods to handle chemical mixtures have been developed and adapted in the epidemiologic context.[28] However, no statistical approach to analyzing mixtures outperforms the others in real-data settings.[29–32] Each statistical mixture approach emphasizes a specific feature of the mixture-outcome association, suggesting that only a combination of statistical approaches can provide the full picture of the relationships among environmental exposures, singly and together, and the health outcome.[33,34]

Despite a multitude of statistical methods to analyze mixtures, only a few approaches can accommodate the associations of environmental mixtures with outcomes at baseline and longitudinally. In this study, we performed (1) linear mixed-effect regression models (LMR) and two mixture approaches that are able to handle repeated-outcome measures: (2) Bayesian weighted quantile sum regression (BWQSR),[35] and (3) Bayesian varying coefficient kernel machine regression (BVCKMR).[36] Here, we assessed the associations of prenatal exposure to OC mixture with childhood BMI measures at 4 years of age and at two subsequent ages (6 and 11–12 years), by leveraging data from the prospective Rhea cohort.[37] We hypothesized that prenatal OC exposure is associated with higher BMI measures in childhood.

Materials and methods

Study population

The Rhea cohort included 1363 mother–child pairs living in the prefecture of Heraklion, Crete, Greece.[37] Briefly, pregnant women were enrolled during the first comprehensive ultrasound examination (mean ± SD, 11.96 ± 1.49 weeks) in 2007–2008. Eligibility criteria included a good understanding of the Greek language, maternal age ≥16 years, and a singleton pregnancy. Pregnant women were further contacted at 6 months of pregnancy, at birth and their children were followed up after birth (9 months, 1, 4, 6, and 11–12 years). At each visit, we collected child anthropometric measures, dietary information, and environmental exposures via structured questionnaires and medical records. During the latest follow-up visit at 11–12 years of age, children underwent a clinical examination and provided blood samples, whereas mothers completed socio-demographic questionnaires. A total of 1110 pregnant mothers provided blood samples for analysis of prenatal OC exposures, and a total of 282 children underwent anthropometric measurements at all clinical examinations at ~4, ~6, and ~11–12 years of age (Figure 1). In the Rhea study the attrition rate was unrelated to participant characteristics, therefore, missing observations throughout the considered time frame were considered missing at random. The study was approved by the ethics committee of the University Hospital in Heraklion, Crete, Greece, and all women provided written informed consent for themselves and for their children at each visit.

Organochlorine compounds exposure

Maternal serum samples were collected at the first prenatal visit and stored in aliquots at –80°C until assayed. The OC laboratory analyses were performed by the National Institute for Health and Welfare, Environmental Health Unit, Kuopio, Finland, with an Agilent 7000B gas chromatography triple-quadrupole mass spectrometer.[38] We determined serum concentrations of HCB, DDE, and six individual PCB congeners (118, 138, 153, 156, 170, and 180). All results were reported on their molecular weight and expressed in pg/mL of serum, whereas samples below the limit of quantification (LOQ) were assigned the value 0.5×LOQ. LOQ was 6pg/mL for PCB-118 and PCB-156 and 10 pg/mL for the remaining compounds. We performed $\log_{10}$-transformations on OC concentrations and ranked them in quartiles to allow for comparison among the three statistical approaches. We identified as exposure outliers all values more distant than four standard deviations (SD) from the mean. This excluded one participant with extreme exposure values (Figure 1).

Child anthropometry

For each child, we obtained anthropometric measures once at each visit with children standing in light clothing without shoes, arms hanging freely, and with their head aligned in the Frankfort horizontal plane. For children's weight, we used a digital scale (SecaBellisima 841) to the nearest 0.1kg, and for height, we used a commercial stadiometer (Seca 213). We then computed BMI as the ratio between weight (kg) and squared height (m^2). BMI age-and-sex specific z-scores were calculated using the World Health Organization child growth reference.40 Owing to the distinctive age-specific BMI pattern—characterized by a rapid increase to a peak in the first year of life, followed by a decline to a nadir between 4 and 6 years of age, before finally rising again in adolescence and adulthood—we plotted the age-BMI relationship with both linear and locally weighted scatterplot smoothing trends in the overall population and we evaluated associations using both BMI and BMI z-score. Our data suggested an approximate linear BMI-age relationship (Figure S1; http://links.lww.com/EE/A186) therefore we included age as a linear term in all models.

Data are available upon reasonable request to the authors and after approval of the data use agreement form. Analytic code is available to the public in GitHub:

https://github.com/ElenaColicino/POPsBMIinRhea-

SDC *Supplemental digital content is available through direct URL citations in the HTML and PDF versions of this article (www.environepidem.com).*

**Corresponding author. Address: Department of Environmental Medicine and Public Health, Icahn School of Medicine at Mount Sinai, 17 E 102nd Street, NY 10029. E-mail: elena.colicino@mssm.edu*

Environmental Epidemiology (2022) 6:e201

Received: 22 July 2021; Accepted 17 February 2022

Published online 22 April 2022

DOI: 10.1097/EE9.0000000000000201

E.C. and K.M. contributed equally to the writing of this article.

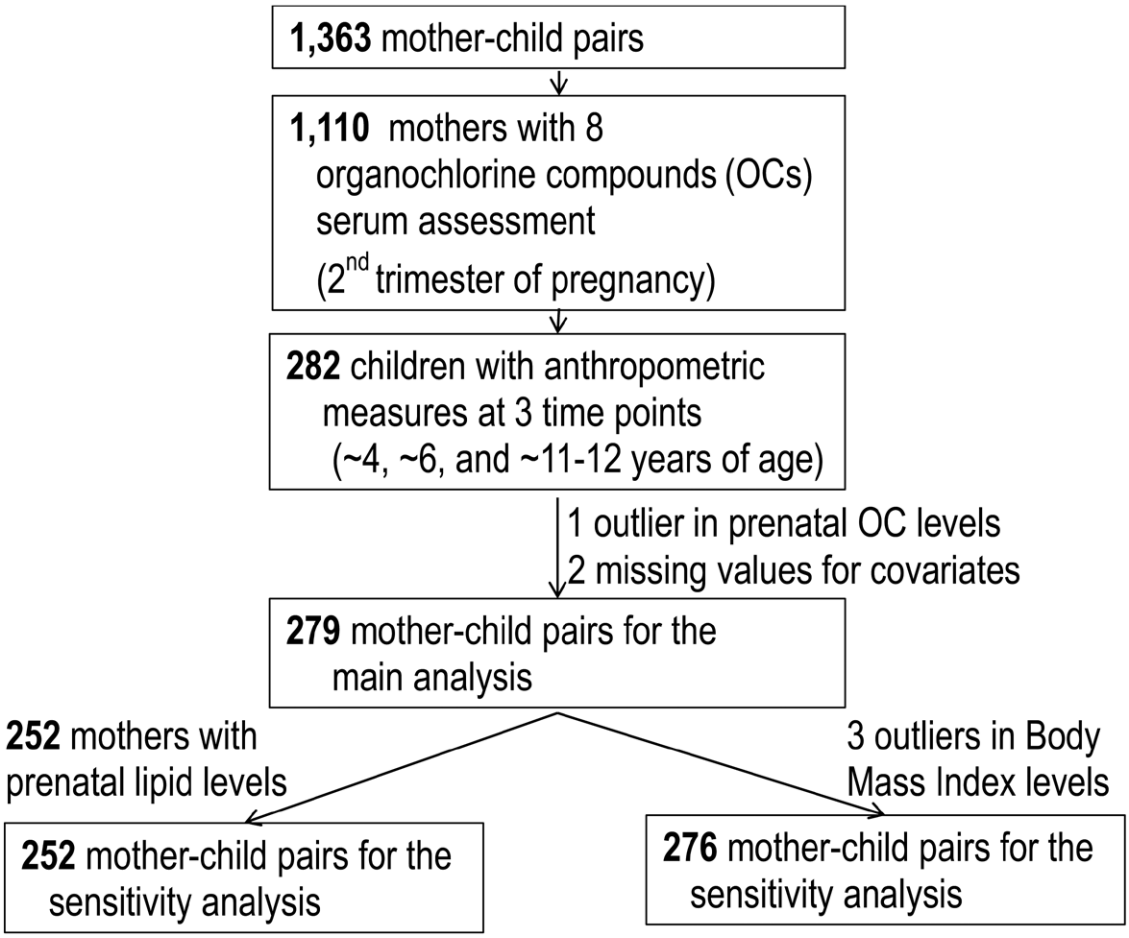

Figure 1. Overview of exclusion criteria in the Rhea study.

Covariates

We collected information about maternal and child covariates via personal interviews, together with self-administered questionnaires and a review of medical records. Maternal cholesterol and triglyceride levels were measured in plasma during the first prenatal visit by using standard enzymatic procedures on an automatic analyzer (AU5400 high-volume chemistry analyzer; Olympus America, Inc., Melville, NY). All covariates considered in this analysis were previously linked to BMI.[9–12,14,27] We constructed a directed acyclic graph (Figure S2; http://links.lww.com/EE/A186) to select the minimal set of covariates and additionally considered those variables associated with both OC exposure and BMI measures (P value < 0.1) which were shown to modify the OC-BMI coefficient estimates by more than 10% when excluded from the fully adjusted model. All main analyses were adjusted for maternal age at birth (years), maternal education (≤9 years, 9–≤12 years, or >12 years of schooling), parity (nulliparous or multiparous), and maternal BMI before pregnancy (kg/m^2), child sex (referent: male), and child's age at clinical follow-up visit (years). Of 282 children with available OC concentrations and anthropometric measures, 279 also had complete data for secondary covariates and were included in the main analysis (Figure 1).

Statistical methods

We calculated the distribution of individual OC concentrations and their correlations by using Pearson's coefficients. We then performed three statistical analyses to evaluate the relationship between OC exposures and both BMI measures (BMI and BMI z-score) at age 4 years and their yearly change across 4–12 years of age: (1) LMR evaluated individual exposure-outcome associations, (2). BWQSR identified an overall mixture effect and (3). BVCKMR modeled nonlinear and nonadditive associations. Children's age was centered at 4 years so that we could capture the association between exposure and BMI measures at 4 years and over time (between 4 and 12 years). To ease the comparisons between analyses, we reported all association estimates for a one-quartile increase in OC exposures, and we centered and scaled all continuous covariates. A summary of the advantages and limitations of each statistical approach is given in Table 1 and a detailed methods description is provided in the supplemental material; http://links.lww.com/EE/A186. We performed analyses using Stata 16, JAGs, and R version 3.6.2.

Sensitivity analyses

(1) We evaluated the robustness of our analyses by excluding children with outcome outliers. This exclusion left us with 276 mother–child pairs for the analyses on BMI and with 278 mother–child pairs for BMI z-score analyses. (2) To account for the OCs' lipid affinity, we adjusted our analyses for maternal cholesterol and triglycerides, both measured during pregnancy. Prenatal lipid levels were available for 252 mothers. (3) To examine whether associations remain robust between the two sexes and due to the well-documented sex differences in body fat composition and metabolic hormone response, we stratified the main analysis by child sex in the LMR and BWQSR models.

Results

Description of the study population

Population characteristics and OC concentrations are in Tables 2 and 3 and Table S1; http://links.lww.com/EE/A186 and

Table 1.

Summary of the characteristics of the statistical models used

Model	Advantages	Limitations	Research Question
Linear mixed-effect regression (LMR)	Estimates linear associations between individual chemicals and the outcome; easy interpretation and implementation	Does not consider correlation[a] among chemicals, thereby increasing spurious findings	What is the association of single chemical concentrations with BMI and BMI trajectories?
Bayesian weighted quantile sum regression (BWQSR)	Estimates the mixture-outcome association and the contribution of each chemical to the mixture	Does not consider nonadditive and nonlinear relationships	What is the overall association of the chemical mixture with BMI and BMI trajectories? Which chemical(s) is (are) the driver(s) for those associations?
Bayesian varying coefficient kernel machine regressions (BVCKMR)	Estimates the association between chemicals and the outcome allowing for nonadditive and nonlinear relationships	Does not estimate the overall mixture-outcome association (while the BKMR for cross-sectional data estimates the overall mixture-outcome association)	What and how (non-linear and nonadditive) is the association of single-chemical concentrations with BMI and BMI trajectories, while accounting for the correlation with all other compounds?

[a]More information in the methods section.

Table 2.

Characteristics of 279 mother-child pairs in the Rhea study

Characteristic	n (%) or Mean ± SD
Maternal characteristics	
Maternal age at delivery (y)	30.5 ± 4.4
Prepregnancy BMI (kg/m^2)	24.9 ± 4.9
Maternal level of education	
Low (<6 years)	31 (11.1)
Middle (6–12 years)	134 (47.9)
High (>12 years)	114 (41)
Parity	
Nulliparous	122 (43.7)
Multiparous	157 (56.3)
Smoking during pregnancy	
Nonsmoker	219 (81.7)
Smoker	49 (18.3)
Cholesterol (mg/dl)	212.5 ± 44.5
Triglycerides (mg/dl)	131.1 ± 52.8
Child characteristics	
Sex	
Boy	154 (55.2)
Girl	125 (44.8)
Gestational age (weeks)	38.3 ± 1.5
Birthweight (g)	3207.3 ± 430.5
Delivery mode	
Vaginal	134 (48.2)
C-section	144 (51.8)

BMI indicates body mass index; SD, standard deviation.

Table S2; http://links.lww.com/EE/A186. Pearson's correlation coefficients of maternal serum OC levels showed moderate (0.37 between DDE and PCB-156) to high correlation among chemicals (0.96 between PCB-180 and PCB-170) (Figure S3; http://links.lww.com/EE/A186).

Association between prenatal OCs and BMI

Linear mixed-effect model regressions for individual chemicals

After correcting for multiple comparison testing by using 5% false discovery rate (FDR), we found a borderline association between prenatal concentrations of DDE and BMI (kg/m^2) at 4 years of age (Figure 2A, Table S3; http://links.lww.com/EE/A186) and a significant and positive association between a quartile increase in DDE levels and BMI z-score at 4 years (Estimate [Est.]:0.14, 95% confidence interval [CI] = 0.05;0.23, q-value = 0.03) (Figure 2C, Table S3; http://links.lww.com/EE/A186).

In the period between 4 and 12 years of age, the yearly change in BMI was positively associated with a quartile increase in individual prenatal level of HCB (Est.:0.10, 95% CI = 0.06, 0.14, q-value < 0.001), PCB-118 (Est.:0.05, 95% CI = 0.02, 0.09, q-value = 0.02), and PCB-138 (Est.:0.06, 95% CI = 0.01, 0.10, q-value = 0.03) (Figure 2B; Table S3; http://links.lww.com/EE/A186). HCB findings were also consistent in the association with BMI z-score yearly change (Est.:0.02, 95% CI = 0.01, 0.04, q-value = 0.004) (Figure 2D, Table S3; http://links.lww.com/EE/A186).

A few compounds showed significant pairwise interactions in their associations with BMI measures at 4 years of age and yearly change in BMI after correcting for multiple comparisons. All significant interactions included HCB. The HCB-PCB-153 and HCB-PCB-156 interactions were consistently associated with both BMI and BMI z-score at 4 years (Tables S4; http://links.lww.com/EE/A186 and Tables S5; http://links.lww.com/EE/A186). Pairwise interactions between HCB and other PCBs were only significant in their association with BMI yearly change.

BWQS for exposure mixtures

Results from the BWQSR showed that a one-quartile increase in the overall OC mixture level was not associated with BMI measures at age 4 years (Figure 3A–C; Table S6; http://links.lww.com/EE/A186), but it was associated with a 0.10 kg/m^2 BMI increase (95% credible interval [CrI]: 0.01, 0.18) and a 0.03 BMI z-score increase (95% CrI: 0.003, 0.06) every year from age 4 to 12 years (Figures 3B–D; Table S6; http://links.lww.com/EE/A186). The contribution of each OC to the mixture was balanced across all compounds, with weights ranging from 0.18 or 0.16 (for DDE, respectively for BMI and BMI z-score) to 0.10 (for PCB-118), in comparison to the prior expected weight of 0.125 for each compound.

BVCKMR to account for co-exposure and nonadditive, nonlinear relationships

BVCKMR findings showed that a one-quartile increase in DDE, PCB-118, and PCB-156 levels was associated with higher BMI at 4 years of age (Est. [95% CrI]: DDE: 0.33 [0.24, 0.43], PCB-118: 0.18 [0.00, 0.37], PCB-156: 0.32 [0.02, 0.62]) (Figure 4A, Table S7; http://links.lww.com/EE/A186). Results on DDE were also consistent in the association with BMI z-score at 4 years (Est. [95% CrI]: 0.19 [0.15, 0.24] (Figure 4C, Table S7; http://links.lww.com/EE/A186). In addition, a quartile increase in HCB and PCB-118 levels was positively associated with yearly change of BMI measures across 4–12 years of age (HCB: BMI: 0.10 [0.07, 0.13]; BMI z-score: 0.03 [0.02, 0.05]; PCB-118: BMI: 0.08 [0.04, 0.12]; BMI z-score: 0.02 [0.002, 0.04] Figures 4B–D, Table S7; http://links.lww.com/EE/A186). A quartile increase in DDE exposure was associated with a decrease in yearly change of BMI measures (BMI: –0.03 [–0.05, 0.00];

Table 3.

Levels of persistent organic pollutants in 2nd trimester maternal serum (n = 279)

Chemical	GM (95% CI) (pg/mL)	Min	Max	Percentile			LOQ (pg/mL)	n < LOQ
				25th	50th	75th		
HCB	93.1 (89.8, 96.5)	28.6	703	64.5	87.5	131	10	0
DDE	2210 (2210, 2220)	210	21600	1290	2150	3670	10	0
PCB-118	18.9 (15.5, 22.2)	3.0	78.9	13.7	19.3	27.3	6	4
PCB-153	136 (133, 140)	31.5	620	94.9	138	198	10	0
PCB-138	73.4 (70.0, 76.8)	13.1	282	50.4	73.1	111	6	113
PCB-156	6.5 (2.5, 10.5)	3.0	40.7	3.0	7.0	11.6	10	0
PCB-180	75.5 (72.0, 78.9)	14.3	531	51.6	75.9	113	10	0
PCB-170	37.9 (34.2, 41.5)	5.0	272	26.4	39.1	57.8	10	5

CI indicates confidence interval; DDE, dichlorodiphenyldichloroethylene; GM, geometric mean; HCB, hexachlorobenzene; LOQ, limit of quantification; Max, maximum; Min, minimum; PCB, polychlorinated biphenyl congeners (118, 153, 138, 156, 170, and 180).

BMI z-score: −0.02 [−0.03, −0.01]). BVCKMR findings also showed a negative association between PCB-138 exposure and BMI z-score yearly change (PCB-138:−0.10 [−0.19, −0.01]) (Figure 4D, Table S7; http://links.lww.com/EE/A186).

Driven by the significant pairwise interaction terms obtained from LMRs, we further evaluated the interactions between HCB and all other compounds based on the exposure-response surface estimated by BVCKMR (Figure S4; http://links.lww.com/EE/A186 and Figures S5; http://links.lww.com/EE/A186). We used heatmaps and cross-sectional plots to reduce dimensionality and to graphically depict the exposure-response relationship. Results were consistent by BMI and BMI z-score. The heatmaps suggested that PCBs (118, 153, 138, 156, 180, and 170) had nonlinear relationships with BMI measures at 4 years and with yearly changes in BMI measures, and HCB interacted with other compounds (DDE, PCB-156, PCB-153, and PCB-180) (Figures S4; http://links.lww.com/EE/A186 and Figures S5; http://links.lww.com/EE/A186). HCB concentrations magnified

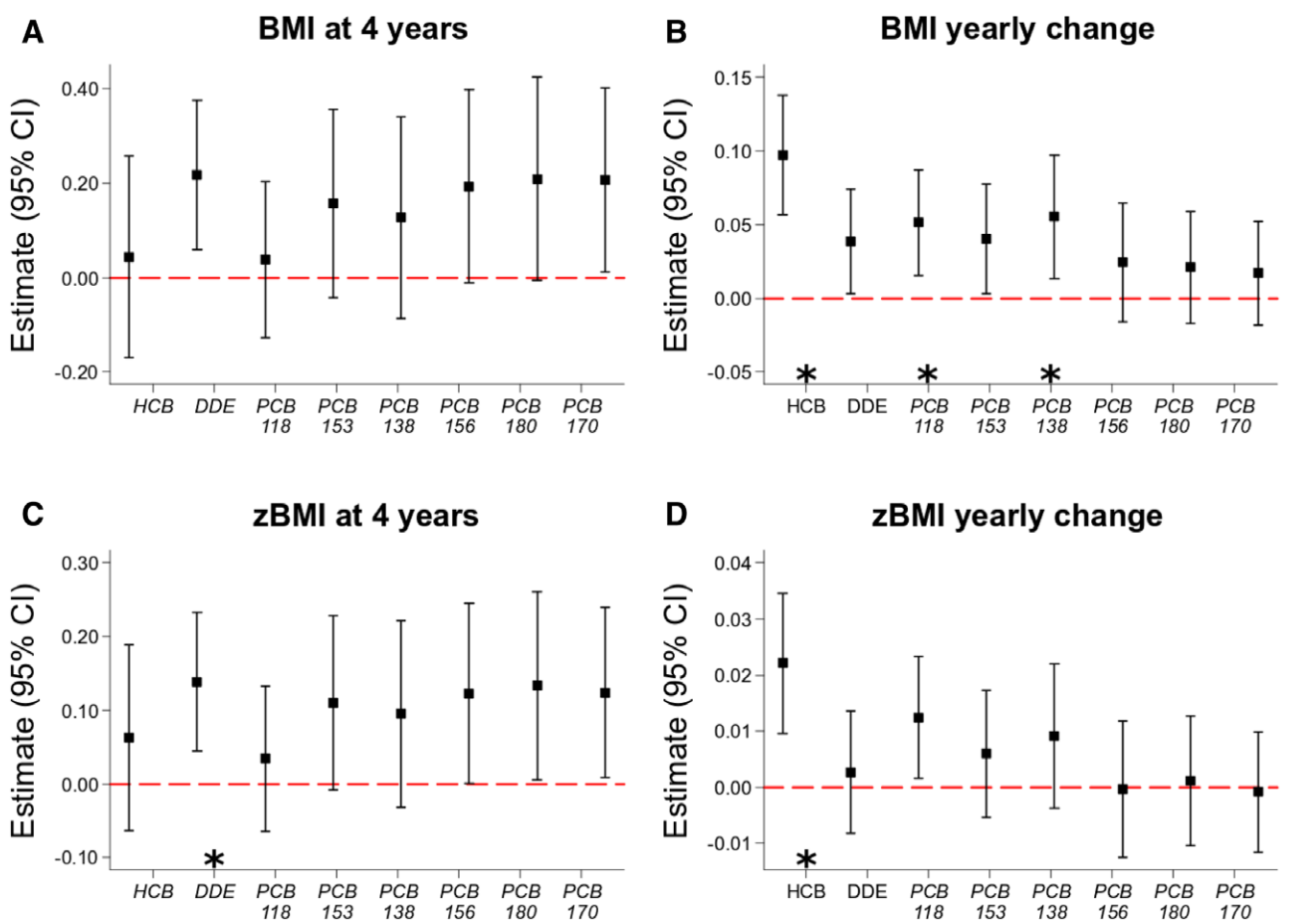

Figure 2. Results of the linear mixed-effect regression[a] in n = 279 mother-child pairs from the Rhea study. Coefficient estimates and 95% confidence intervals (CI) for the relationship[a] between a one-quartile increase in the individual exposure to organochlorine compounds (OC) and childhood body mass index (BMI) at 4 years (A), yearly change in BMI from 4 to 12 years of age (B), BMI z-score (z-BMI) at 4 years (C) and yearly change in BMI z-score from 4 to 12 years of age (D). DDE indicates dichlorodiphenyldichloroethylene; HCB, hexachlorobenzene; PCB, polychlorinated biphenyl congeners (118, 153, 138, 156, 170, and 180). [a]Adjusted for maternal age at birth (years), maternal education at recruitment (≤6 years, >6–≤12 years, or >12 years), parity (nulliparous or multiparous), and maternal BMI before pregnancy (kg/m^2), child sex (M/F), and child age at clinical follow-up visit (years). *Statistically significant after correcting for multiple testing (Table S1).

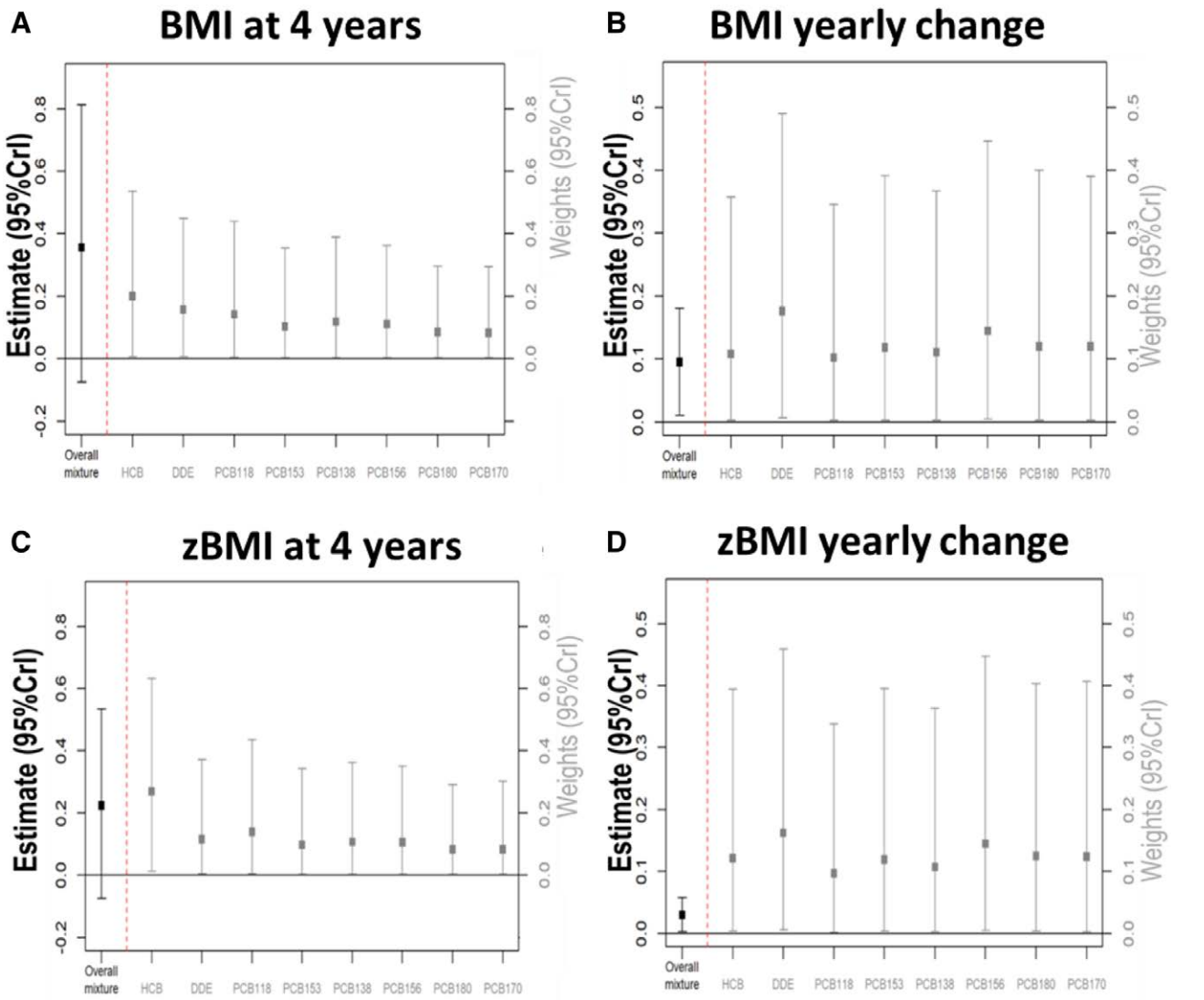

Figure 3. Results of the Bayesian weighted quantile sum regression* in n = 279 mother-child pairs from the Rhea study. Coefficient estimates and 95% credible intervals (CrI) for the relationship* between a one-quartile increase in the overall mixture exposure to organochlorine compounds and both childhood body mass index (BMI) at 4 years (A), yearly change in BMI from 4 to 12 years of age (B), BMI z-score (z-BMI) at 4 years (C) and yearly change in BMI z-score from 4 to 12 years of age (D). The weights and 95% CrI identifying the individual contribution of each OC to the mixture are shown in gray. DDE indicates dichlorodiphenyldichloroethylene; HCB, hexachlorobenzene; PCB, polychlorinated biphenyl congeners (118, 153, 138, 156, 170, and 180).DDE indicates dichlorodiphenyldichloroethylene; HCB, hexachlorobenzene; PCB, polychlorinated biphenyl congeners (118, 153, 138, 156, 170, and 180)*Adjusted for maternal age at birth (years), maternal education at recruitment (≤6 years, >6–≤12 years, or >12 years), parity (nulliparous or multiparous) and maternal BMI before pregnancy (kg/m^2), child sex (M/F), and child age at clinical follow-up visit (years).

the associations of DDE, PCB-156, and PCB-153 with BMI measures at 4 years and led to the increased yearly change in BMI measures, with high levels of DDE, PCB-153, and PCB-180, holding all other exposures at their median value.

Sensitivity analysis

(1) There was no major departure from the overall conclusions of the primary analysis after excluding suspected outliers of BMI (n = 3) or BMI z-score (n = 1). However, in the LMR the OC-BMI measures associations at 4 years of age were not statistically significant (Figures S6–S8; http://links.lww.com/EE/A186). (2) After adjusting for lipids, our results were similar to those of the main analyses in the LMR and in the BWQSR (Figures S9; http://links.lww.com/EE/A186 and Figures S10; http://links.lww.com/EE/A186), although we found novel positive associations between PCB-153 with yearly change in BMI measures, using the BVCKMR (Figure S11; http://links.lww.com/EE/A186). PCB-170 exposure was also associated with yearly change in BMI in the BVCKMR model (3) Results on sex-stratified analysis showed heterogeneity in the OC-BMI and OC-BMI z-score associations between boys and girls. Boys showed significant and more exacerbated associations between prenatal exposure to OCs and yearly change in BMI and BMI z-score than girls. BWQSR results supported a stronger overall mixture association with yearly change in BMI measures in boys (Est.[95% CrI]: BMI: 0.15 [0.05, 0.26]; BMI z-score: 0.04 [0.00, 0.07]) than in girls (Est. [95% CrI]: BMI: 0.02 [–0.08, 0.13]; BMI z-score: -0.01[–0.04, 0.03]) (Figures S12–S15; http://links.lww.com/EE/A186).

Discussion

This is the first study to show the relationship between *in-utero* exposure to OCs and childhood BMI measures at 4 years and yearly thereafter by using three distinct analytical models which

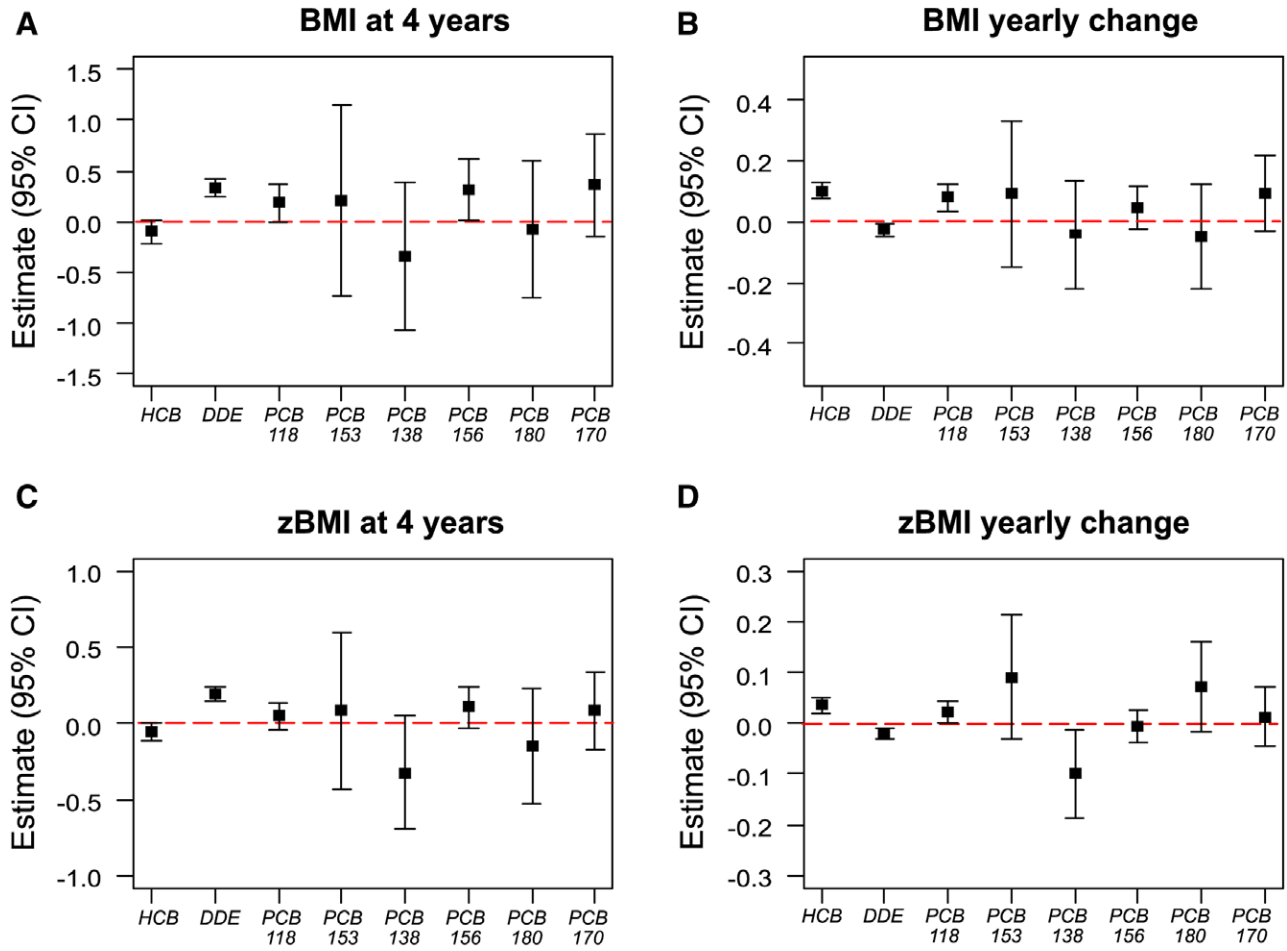

Figure 4. Results of the Bayesian varying coefficient kernel machine regression* in n = 279 mother-child pairs from the Rhea study. Coefficient estimates and 95% credible intervals (CrI) for the relationship* between a one-quartile increase (from 50th percentile to 75th percentile) in the exposure to a single organochlorine compound and childhood body mass index (BMI) at 4 years (A) and yearly change in BMI from 4 to 12 years of age (B) BMI z-score (z-BMI) at 4 years (C) and yearly change in BMI z-score from 4 to 12 years of age (D). DDE indicates dichlorodiphenyldichloroethylene; HCB, hexachlorobenzene; PCB, polychlorinated biphenyl congeners (118, 153, 138, 156, 170, and 180)*Adjusted for maternal age at birth (years), maternal education at recruitment (≤6 years, >6–≤12 years, or >12 years), parity (nulliparous or multiparous) and maternal BMI before pregnancy (kg/m^2), child sex (M/F), and child age at clinical follow-up visit (years)

can provide strong evidence for robust associations. We found that prenatal exposure to OCs was associated with higher BMI and BMI z-score at 4 years and with increased yearly change in BMI measures between 4 and 12 years of age. All models were consistent in showing harmful associations between prenatal OC concentration and yearly change in BMI measurements, although only the model accommodating nonlinear and nonadditive associations consistently captured the potentially harmful role of DDE, PCB-118, and PCB-156 on BMI measures at age 4 years.

The linear mixed-effect model regression showed positive associations between the individual OCs and yearly change in BMI measures, and the BWQSR showed an overall positive association between the OC mixture and yearly change in BMI measures, although there was no difference in relative contribution to the mixture among OC compounds with BWQSR. BVCKMR confirmed a positive association between both HCB and PCB-118 levels with yearly change in BMI measures from 4-12 years of age and showed a negative association between DDE exposure and yearly change in BMI measures and between PCB-138 and BMI z-score yearly change. BVCKMR also suggested nonlinear associations of PCBs with childhood BMI measures, both at 4 years and over time, and interactions between HCB and other PCB compounds. Results were consistent across sensitivity analyses.

Although the three approaches are based on models that have different assumptions and characteristics, they showed remarkably similar results with only a few minor discrepancies. The LMR provided the canonical association between individual OC exposures and both BMI measures at 4 years and their yearly changes across 4–12 years of age. However, that approach precludes evaluating potential nonlinear and synergistic associations.[36] In addition, the LMR approach could not accommodate the presence of multiple correlated OCs, and so a multiple testing correction, such as FDR, had to be considered to reduce false-positive findings.[41] The BWQSR and BVCKMR are mixture approaches, and both of them incorporate the correlation structure of OC exposures by modeling them jointly, thus minimizing the issue of false positives and standard error inflation of the classical linear framework.[42,43]

The BWQSR suggested associations between OC mixture and BMI outcomes, assuming additivity among OCs and a linear relationship between OC exposures and outcomes.[35] Because of these assumptions, BWQSR results were similar to those from the linear models. The BWQSR assumed a Dirichlet density distribution with equal parameters for all OC compounds as prior for the weights,[35] and for this reason, a strong correlation structure among OCs might have led to balanced weights. Future studies may want to consider a more informative prior for the weights and also provide the Bayes factors to formally compare estimated weights with those under the null for all OCs to the mixture. A major advantage of BWQSR is that it provides an overall mixture-outcome association, thus complementing results from the BVCKMR.

BVCKMR, in contrast to the previous models, estimates interactions among OCs and nonlinear OC exposure–BMI measures associations via quadratic kernel functions. Because of these flexible characteristics, this approach estimates more parameters, thus requiring more computational time and data than previous alternatives, and relies on model form and assumptions that can be sensitive to outliers.[36] Although this approach had very different assumptions from previous models, the results were consistent regarding the positive associations between both HCB and PCB-118 levels and yearly change in BMI measures from 4–12 years of age. It also provided a different perspective than the other approaches, suggesting nonlinear exposure-response associations with PCBs and synergistic PCB-HCB relationships. Pairwise interactions in linear models also supported the synergistic relationships. BVCMKR and LMR also showed some significant results with DDE, PCB-118, and PCB-156 with BMI measures at 4 years.

All our findings were consistent with previous literature reporting positive associations between *in-utero* exposure of individual OCs and increases in childhood BMI measures. Prior studies, also from our group, consistently identified an increase in childhood BMI measures associated with an increase in individual levels of exposure to HCB, DDE,[10,12,14,27,44–46] or PCBs,[10,46] including a previous multiple-pollutant approach.[9] Prior results showed strong associations between childhood BMI and HCB or DDE, and previous analyses have shown nonmonotonic associations for those chemicals.[10,47,48] Here we confirmed the detrimental role of elevated HCB and DDE levels, and we leveraged a kernel machine regression to confirm similar nonlinear chemical-response associations. Prior associations of BMI and prenatal exposure to PCBs were inconclusive, and some of the studies reported null results,[11,46] although others showed nonmonotonic associations.[10] Based on those findings, many authors suggested that the BMI associations with PCBs were positively confounded by other OC compounds and potentially masked by the strong correlation structure and dose-response associations.[10,11,27] In contrast, we found nonlinear associations between PCBs and childhood BMI measures while accounting for their correlation, and interactions among PCBs and HCB.

The putative biological mechanisms that relate intrauterine OC exposures to elevated childhood BMI involve the capacity of OCs to alter the endocrine system by modulating preadipocyte proliferation,[49] which increases in response to exposure to OC environmental levels (PCB-153 and DDE). Those associations were similar across different types of cells.[50] *In-vitro* studies have also shown that exposure of mature adipocytes to DDE led to higher levels of leptin, a hormone that regulates the cell's energy balance.[51] Results were also consistent with another study showing prenatal exposure to both HCB and DDE levels and child leptin levels.[52]

OCs, similar to other endocrine-disrupting chemicals, alter thyroid function and metabolism,[53] thereby impacting weight and its homeostasis.[54] HCB, DDE, and PCBs have been shown to have estrogenic, antiestrogenic, and antiandrogenic effects.[55] Specifically, HCB has a role as both androgen receptor and estrogen-related receptor antagonist,[56,57] whereas DDE promotes estrogenic activity at higher levels than in control groups, and lower DDE concentrations show antiandrogenic and antiprogesterogenic activity,[56] which mimics the roles of estrogen receptor agonists and androgen receptor antagonists. PCB congeners show estrogenic, antiestrogenic, and/or antiandrogenic effects at very low and high concentrations, thus suggesting that PCBs have a nonmonotonic effect.[12,55,58,59] Our findings concur with a breadth of existing data from *in-vivo* and *in-vitro* studies that show obesogenic effects of OC exposures. Based on our results, future experimental and epidemiologic studies should also consider potential OC interactions, especially synergistic effects between HCB and other OCs.

Several strengths should be noted in our article. The results of the three analytical approaches have different assumptions and characteristics, and they suggested similar conclusions. All methods captured the positive associations between maternal OC exposure and childhood BMI measures. The methods complement each other, providing results from the classical linear framework, and results from two mixture-based approaches. Previous literature also supported a few discrepancies in the findings, suggesting that a combination of methods provides a more complete picture of the relationships between prenatal OC exposures and childhood BMI measures. To make comparisons across methods and to limit the effect of exposure outliers, we centered and scaled all continuous covariates, and we reported all results as an increment in the quartile of the exposures. We leveraged the Rhea study, a well-established prospective cohort with prenatal exposure information and longitudinal child anthropometric measures.[37] OC levels in the Rhea study were similar to or lower than concentrations in other populations. For example, exposure to HCB (median, 0.09 μg/L) and DDE (median, 2.15 μg/L) was lower than the median exposure in previous American and European studies (HCB: 0.24 μg/L,[10] 0.68 μg/L,[45] DDE: 24.59 μg/L,[60] 1.06 μg/L[10]). PCB concentrations were also lower than reported levels in other studies.[10,60] In our sensitivity analyses, results with lipid adjustment were largely consistent with the main analyses. Results on sex-stratified analysis also were consistent with previous findings showing differences in the magnitude and sensitivity of the effects of OCs by sex because of their differences in the natural androgen–estrogen balance during critical windows of fetal development.[61] In addition, our sex-stratified results were in line with well-documented sex differences in body fat composition, fat distribution, energy homeostasis, and metabolic hormone response, showing a stronger association between prenatal exposure to OCs and BMI in boys than in girls.[61] Larger studies with sufficient power to detect interactions and to avoid small data bias should also focus on determining sex-stratified associations with BKMR.

In our analyses, we did not consider birthweight. Indeed, birthweight can be considered an intermediary because of its adverse relationship with prenatal intrauterine exposure to OCs.[46,62] Owing to missing information about stillbirths and miscarriages, we also did not consider any live-birth bias, which assumes that children with higher exposure are more likely to be born still and with altered birth weight,[63] and larger studies should investigate the OC role on stillbirths.

Although we hypothesized missing at random in our study, the loss to follow-up may bias our results and further studies should consider weighting techniques; however, it has been suggested that attrition similar or even larger than that of the Rhea study would not impact the qualitative conclusions in terms of direction and magnitude.[64] Further studies should also assess the OC role on other measures of adiposity, such as percent body fat and fat mass index, or the contribution of obesity-related factors, such as physical activity/sedentary life, and pubertal timing, to those associations since we had no longitudinal data regarding such factors. In addition, we had no information about cumulative OC levels during the prenatal period or OC levels in the postnatal period, thus we were not able to rule out the influential windows of exposure during early life or whether the observed associations for prenatal OC exposure are partially owing to correlations with postnatal exposure levels. However, prior studies have shown that effect estimates for the association between prenatal OC exposures and child weight or BMI do not substantially change after adjustment for postnatal OC exposure.[10,12] Finally, exposure to HCB, DDE, and PCBs was lower than levels in other studies,[4,5,10,60] therefore, our results should be cautiously generalized to other populations with different exposure ranges.

Conclusions

We showed that *in-utero* exposure to OC concentrations was associated with higher BMI measures at 4 years and with steeper

yearly changes in BMI measures from 4 to 12 years of age. Results were similar across three distinct statistical approaches, despite differences in the models' assumptions and characteristics. All findings taken together provide a more comprehensive characterization of the associations between prenatal OC exposures and childhood BMI measures, suggesting long-term consequences for those exposures.

Bibliography

1. Hedley AA, Ogden CL, Johnson CL, Carroll MD, Curtin LR, Flegal KM. Prevalence of overweight and obesity among US children, adolescents, and adults, 1999-2002. *JAMA*. 2004;291:2847–2850.
2. Dong GH, Qian Z, Liu MM, et al. Ambient air pollution and the prevalence of obesity in Chinese children: the seven northeastern cities study. *Obesity*. 2014;22:795–800.
3. Global action plan on physical activity 2018–2030: more active people for a healthier world. Geneva: World Health Organization; 2018. Licence: CC BY-NC-SA 3.0 IGO.
4. Hughes AR, Sherriff A, Lawlor DA, Ness AR, Reilly JJ. Incidence of obesity during childhood and adolescence in a large contemporary cohort. *Prev Med*. 2011;52:300–304.
5. Must A, Strauss RS. Risks and consequences of childhood and adolescent obesity. *Int J Obes Relat Metab Disord*. 1999;23 Suppl 2:S2–11.
6. Dietz WH. Health consequences of obesity in youth: childhood predictors of adult disease. *Pediatrics*. 1998;101(3 Pt 2):518–525.
7. Puhl RM, Latner JD. Stigma, obesity, and the health of the nation's children. *Psychol Bull*. 2007;133:557–580.
8. Reilly JJ, Kelly J. Long-term impact of overweight and obesity in childhood and adolescence on morbidity and premature mortality in adulthood: systematic review. *Int J Obes (Lond)*. 2011;35:891–898.
9. Agay-Shay K, Martinez D, Valvi D, et al. Exposure to endocrine-disrupting chemicals during pregnancy and weight at 7 years of age: a multi-pollutant approach. *Environ Health Perspect*. 2015;123:1030–1037.
10. Valvi D, Mendez MA, Martinez D, et al. Prenatal concentrations of polychlorinated biphenyls, DDE, and DDT and overweight in children: a prospective birth cohort study. *Environ Health Perspect*. 2012;120:451–457.
11. Karlsen M, Grandjean P, Weihe P, Steuerwald U, Oulhote Y, Valvi D. Early-life exposures to persistent organic pollutants in relation to overweight in preschool children. *Reprod Toxicol*. 2017;68:145–153.
12. Valvi D, Mendez MA, Garcia-Esteban R, et al. Prenatal exposure to persistent organic pollutants and rapid weight gain and overweight in infancy. *Obesity (Silver Spring)*. 2014;22:488–496.
13. Vrijheid M, Casas M, Gascon M, Valvi D, Nieuwenhuijsen M. Environmental pollutants and child health-A review of recent concerns. *Int J Hyg Environ Health*. 2016;219:331–342.
14. Warner M, Aguilar Schall R, Harley KG, Bradman A, Barr D, Eskenazi B. In utero DDT and DDE exposure and obesity status of 7-year-old Mexican-American children in the CHAMACOS cohort. *Environ Health Perspect*. 2013;121:631–636.
15. Wallwork RS, Colicino E, Zhong J, et al. Ambient fine particulate matter, outdoor temperature, and risk of metabolic syndrome. *Am J Epidemiol*. 2017;185:30–39.
16. Guo W, Pan B, Sakkiah S, et al. Persistent organic pollutants in food: contamination sources, health effects and detection methods. *Int J Environ Res Public Health*. 2019;16:E4361.
17. Passuello A, Mari M, Nadal M, Schuhmacher M, Domingo JL. POP accumulation in the food chain: integrated risk model for sewage sludge application in agricultural soils. *Environ Int*. 2010;36:577–583.
18. Herbstman JB, Sjödin A, Kurzon M, et al. Prenatal exposure to PBDEs and neurodevelopment. *Environ Health Perspect*. 2010;118:712–719.
19. Programme UE. Stockholm Convention on Persistent Organic Pollutants. Available at: http://chm.pops.int. Accessed 6 October 2021.
20. Congress U. Safe Chemicals Act of 2011. In: Congress U, ed. *The Official Start of TSCA Reform Efforts in the New Congress Keller and Heckman LLP*.
21. Lara LA, Duarte AA, Reis RM, Vieira CS, Rosa-e-Silva AC. Endocrine disrupters: potential risk factors affecting sexual function in both men and women. *J Sex Med*. 2012;9:941–942.
22. Meeker JD. Exposure to environmental endocrine disruptors and child development. *Arch Pediatr Adolesc Med*. 2012;166:952–958.
23. Pombo M, Castro-Feijoo L. Endocrine disrupters. *J Pediatr Endocrinol Metab* 2005;18(supplement):1145–1156.
24. Rylander L, Rignell-Hydbom A, Tinnerberg H, Jönsson BA. Trends in human concentrations of endocrine disruptors: possible reasons and consequences. *J Epidemiol Community Health*. 2014;68:4–5.
25. Mahalingaiah S, Missmer SA, Maity A, et al. Association of hexachlorobenzene (HCB), dichlorodiphenyltrichloroethane (DDT), and dichlorodiphenyldichloroethylene (DDE) with *in vitro* fertilization (IVF) outcomes. *Environ Health Perspect*. 2012;120:316–320.
26. Güil-Oumrait N, Valvi D, Garcia-Esteban R, et al. Prenatal exposure to persistent organic pollutants and markers of obesity and cardiometabolic risk in Spanish adolescents. *Environ Int*. 2021;151:106469.
27. Vafeiadi M, Georgiou V, Chalkiadaki G, et al. Association of prenatal exposure to persistent organic pollutants with obesity and cardiometabolic traits in early childhood: the Rhea mother-child cohort (Crete, Greece). *Environ Health Perspect*. 2015;123:1015–1021.
28. Lazarevic N, Barnett AG, Sly PD, Knibbs LD. Statistical methodology in studies of prenatal exposure to mixtures of endocrine-disrupting chemicals: a review of existing approaches and new alternatives. *Environ Health Perspect*. 2019;127:26001.
29. Gibson EA, Nunez Y, Abuawad A, et al. An overview of methods to address distinct research questions on environmental mixtures: an application to persistent organic pollutants and leukocyte telomere length. *Environ Health*. 2019;18:76.
30. Zhang Y, Dong T, Hu W, et al. Association between exposure to a mixture of phenols, pesticides, and phthalates and obesity: comparison of three statistical models. *Environ Int*. 2019;123:325–336.
31. Chiu YH, Bellavia A, James-Todd T, et al.; EARTH Study Team. Evaluating effects of prenatal exposure to phthalate mixtures on birth weight: a comparison of three statistical approaches. *Environ Int*. 2018;113:231–239.
32. Mazzella M, Sumner SJ, Gao S, et al.; CHEAR Metabolomics Analysis Team. Quantitative methods for metabolomic analyses evaluated in the Children's Health Exposure Analysis Resource (CHEAR). *J Expo Sci Environ Epidemiol*. 2020;30:16–27.
33. Taylor KW, Joubert BR, Braun JM, et al. Statistical approaches for assessing health effects of environmental chemical mixtures in epidemiology: lessons from an innovative workshop. *Environ Health Perspect*. 2016;124:A227–A229.
34. Tanner E, Lee A, Colicino E. Environmental mixtures and children's health: identifying appropriate statistical approaches. *Curr Opin Pediatr*. 2020;32:315–320.
35. Colicino E, Pedretti NF, Busgang SA, Gennings C. Per- and poly-fluoroalkyl substances and bone mineral density: results from the Bayesian weighted quantile sum regression. *Environ Epidemiol*. 2020;4:e092.
36. Liu SH, Bobb JF, Claus Henn B, et al. Bayesian varying coefficient kernel machine regression to assess neurodevelopmental trajectories associated with exposure to complex mixtures. *Stat Med*. 2018;37:4680–4694.
37. Chatzi L, Leventakou V, Vafeiadi M, et al. Cohort profile: the mother-child cohort in Crete, Greece (Rhea Study). *Int J Epidemiol*. 2017;46:1392–1393k.
38. Koponen J, Rantakokko P, Airaksinen R, Kiviranta H. Determination of selected perfluorinated alkyl acids and persistent organic pollutants from a small volume human serum sample relevant for epidemiological studies. *J Chromatogr A*. 2013;1309:48–55.
39. Schisterman EF, Whitcomb BW, Louis GM, Louis TA. Lipid adjustment in the analysis of environmental contaminants and human health risks. *Environ Health Perspect*. 2005;113:853–857.
40. de Onis M, Onyango AW, Borghi E, Siyam A, Nishida C, Siekmann J. Development of a WHO growth reference for school-aged children and adolescents. *Bull World Health Organ*. 2007;85:660–667.
41. Benjamini Y, Hochberg Y. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J Royal Stat Soc*. 1995;57:289–300.
42. Yorita Christensen KL, White P. A methodological approach to assessing the health impact of environmental chemical mixtures: PCBs and hypertension in the National Health and Nutrition Examination Survey. *Int J Environ Res Public Health*. 2011;8:4220–4237.
43. Becker J-M, Ringle CM, Sarstedt M, Volckner F. How collinearity affects mixture regression results. *Mark Lett*. 2015;26:643–659.
44. Mendez MA, Garcia-Esteban R, Guxens M, et al. Prenatal organochlorine compound exposure, rapid weight gain, and overweight in infancy. *Environ Health Perspect*. 2011;119:272–278.
45. Smink A, Ribas-Fito N, Garcia R, et al. Exposure to hexachlorobenzene during pregnancy increases the risk of overweight in children aged 6 years. *Acta Paediatr*. 2008;97:1465–1469.
46. Verhulst SL, Nelen V, Hond ED, et al. Intrauterine exposure to environmental pollutants and body mass index during the first 3 years of life. *Environ Health Perspect*. 2009;117:122–126.
47. Lee DH, Steffes MW, Sjödin A, Jones RS, Needham LL, Jacobs DR Jr. Low dose of some persistent organic pollutants predicts type 2 diabetes: a nested case-control study. *Environ Health Perspect*. 2010;118:1235–1242.

48. Lee DH, Steffes MW, Sjödin A, Jones RS, Needham LL, Jacobs DR Jr. Low dose organochlorine pesticides and polychlorinated biphenyls predict obesity, dyslipidemia, and insulin resistance among people free of diabetes. *PLoS One*. 2011;6:e15977.
49. Moreno-Aliaga MJ, Matsumura F. Effects of 1,1,1-trichloro-2,2-bis(p-chlorophenyl)-ethane (p,p'-DDT) on 3T3-L1 and 3T3-F442A adipocyte differentiation. *Biochem Pharmacol*. 2002;63:997–1007.
50. Chapados NA, Casimiro C, Robidoux MA, et al. Increased proliferative effect of organochlorine compounds on human preadipocytes. *Mol Cell Biochem*. 2012;365:275–278.
51. Howell G 3rd, Mangum L. Exposure to bioaccumulative organochlorine compounds alters adipogenesis, fatty acid uptake, and adipokine production in NIH3T3-L1 cells. *Toxicol In Vitro*. 2011;25:394–402.
52. Yu WH, Kimura M, Walczewska A, Karanth S, McCann SM. Role of leptin in hypothalamic-pituitary function. *Proc Natl Acad Sci U S A*. 1997;94:1023–1028.
53. Diamanti-Kandarakis E, Bourguignon JP, Giudice LC, et al. Endocrine-disrupting chemicals: an Endocrine Society scientific statement. *Endocr Rev*. 2009;30:293–342.
54. Grün F, Blumberg B. Endocrine disrupters as obesogens. *Mol Cell Endocrinol*. 2009;304:19–29.
55. Bonefeld-Jørgensen EC, Andersen HR, Rasmussen TH, Vinggaard AM. Effect of highly bioaccumulated polychlorinated biphenyl congeners on estrogen and androgen receptor activity. *Toxicology*. 2001;158:141–153.
56. Li J, Li N, Ma M, Giesy JP, Wang Z. *In vitro* profiling of the endocrine disrupting potency of organochlorine pesticides. *Toxicol Lett*. 2008;183:65–71.
57. Tang-Péronard JL, Heitmann BL, Andersen HR, et al. Association between prenatal polychlorinated biphenyl exposure and obesity development at ages 5 and 7 y: a prospective cohort study of 656 children from the Faroe Islands. *Am J Clin Nutr*. 2014;99:5–13.
58. Boas M, Feldt-Rasmussen U, Skakkebaek NE, Main KM. Environmental chemicals and thyroid function. *Eur J Endocrinol*. 2006;154:599–611.
59. Flor S, He X, Lehmler HJ, Ludewig G. Estrogenicity and androgenicity screening of PCB sulfate monoesters in human breast cancer MCF-7 cells. *Environ Sci Pollut Res Int*. 2016;23:2186–2200.
60. Cupul-Uicab LA, Klebanoff MA, Brock JW, Longnecker MP. Prenatal exposure to persistent organochlorines and childhood obesity in the US collaborative perinatal project. *Environ Health Perspect*. 2013;121:1103–1109.
61. Heindel JJ, Blumberg B, Cave M, et al. Metabolism disrupting chemicals and metabolic disorders. *Reprod Toxicol*. 2017;68:3–33.
62. Wojtyniak BJ, Rabczenko D, Jönsson BA, et al.; INUENDO research group. Association of maternal serum concentrations of 2,2', 4,4'5,5'-hexachlorobiphenyl (CB-153) and 1,1-dichloro-2,2-bis (p-chlorophenyl)-ethylene (p,p'-DDE) levels with birth weight, gestational age and preterm births in Inuit and European populations. *Environ Health*. 2010;9:56.
63. Raz R, Kioumourtzoglou MA, Weisskopf MG. Live-birth bias and observed associations between air pollution and autism. *Am J Epidemiol*. 2018;187:2292–2296.
64. Howe LD, Tilling K, Galobardes B, Lawlor DA. Loss to follow-up in cohort studies: bias in estimates of socioeconomic inequalities. *Epidemiology*. 2013;24:1–9.