

Arachidonic acid status during pregnancy is associated with polychlorinated biphenyl exposure¹⁻³

Philippe Grandjean and Pál Weihe

ABSTRACT

Background: Seafood is an important source of long-chain polyunsaturated fatty acids (LCPs), which are essential for normal growth and development. However, the nutritional benefits could be limited by polychlorinated biphenyl (PCB) contamination. In particular, inhibition of desaturase activities by PCBs may affect the maintenance of arachidonic acid (AA) status during development.

Objective: The aim was to evaluate AA status in a birth cohort from a fishing community with a high seafood intake and a wide range of PCB exposures.

Design: We measured LCP concentrations in paired mother and umbilical cord serum samples obtained from 182 consecutive births in the Faroe Islands, where PCB-contaminated whale blubber forms part of the diet. PCB exposure was determined from maternal concentrations.

Results: Serum phospholipid AA concentrations averaged 9.14% and 16.5% (by wt) in maternal and cord serum, respectively. After adjustment for gestational age and concentrations of linoleic, α -linolenic, and eicosapentaenoic acids, a decrease in AA concentrations of 0.17% (by wt) (95% CI: 0.03%, 0.31%) and 0.31% (by wt) (95% CI: 0.10%, 0.52%) was seen in maternal and cord serum, respectively, for each doubling of PCB exposure.

Conclusions: Increased PCB exposure was associated with a modest decrease in serum AA concentrations, which is in accordance with the experimental evidence of desaturase inhibition by PCBs. Such interference with LCP utilization could attenuate the beneficial effects of the essential lipids contained in seafood. Because AA is of key importance for growth and development, these results suggest that this possible mechanism for PCB toxicity deserves to be explored. *Am J Clin Nutr* 2003;77:715-9.

KEY WORDS Arachidonic acid, fatty acids, fatty acid desaturases, maternal-fetal exchange, polychlorinated biphenyls, long-chain polyunsaturated fatty acids, Faroe Islands

INTRODUCTION

Seafood is an important source of nutrients, including long-chain polyunsaturated fatty acids (LCPs) (1). Arachidonic acid (AA; 20:4n-6) is of particular importance for growth and development (2, 3). Serum AA concentrations are usually maintained even at high seafood intakes (4-6), but an increased supply of eicosapentaenoic acid (EPA; 20:5n-3) during development is thought to reduce AA concentrations (7, 8).

Some types of seafood may contain toxic contaminants, such as polychlorinated biphenyls (PCBs) (9) that are known to cause developmental toxicity (10, 11). Experimental studies suggest that PCBs may affect the utilization of LCPs by inhibition of Δ^5 - and Δ^6 -desaturation (12, 13). Such enzyme inhibition would lead to deficient formation of AA from its precursor, linoleic acid (LA; 18:2n-6). Most of the fetal supply of AA is thought to originate from the maternal circulation (14), but fetal hepatic metabolism also plays a role (15).

Therefore, the question emerged whether toxic pollutants may have an effect on the benefits provided by essential LCPs in seafood. To address this concern, we studied LCP profiles in paired mother and umbilical cord serum samples obtained from consecutive births in a fishing community with a high intake of marine lipids and increased exposures to PCBs from maternal intake of whale blubber (16, 17).

SUBJECTS AND METHODS

Subjects

A cohort of 182 spontaneous, singleton, term births was generated from consecutive births during a 12-mo period in 1994-1995 at the National Hospital in Tórshavn, Faroe Islands, Denmark (16, 17). This cohort represented 64% of the 293 births that occurred during that period; incomplete sampling occurred mainly because of surgical intervention or logistic problems in the busy ward. In addition, 4 infants were excluded because of birth before 36 wk of gestation, and 2 infants were excluded because of congenital disease. Relevant obstetric data were obtained by standardized procedures; all birth weights were ≥ 2500 g. The study was designed in accordance with the latest version of the

¹ From the Institute of Public Health, University of Southern Denmark, Odense (PG and PW); the Departments of Environmental Health and Neurology, Boston University Schools of Medicine and Public Health, Boston (PG); and the Faroese Hospital System, Tórshavn, Faroe Islands, Denmark (PW).

² Supported by the European Commission (Environment and Climate Research Programme, EV 5V-CT940 472), the National Institute of Environmental Health Sciences (ES06894), and the Danish Medical Research Council.

³ Address reprint requests to P Grandjean, Institute of Public Health, University of Southern Denmark, Winslowparken 17, DK-5000 Odense, Denmark. E-mail: pgrandjean@health.sdu.dk.

Received May 16, 2002.

Accepted for publication July 30, 2002.

TABLE 1

Relative concentrations of long-chain polyunsaturated fatty acids in phospholipids in maternal ($n = 173$) and umbilical cord sera ($n = 154$) from mother and infant pairs and the association of those concentrations with polychlorinated biphenyl concentrations in maternal serum after logarithmic transformation

Fatty acid	Maternal serum			Cord serum		
	$\bar{x} \pm SD$	r	P	$\bar{x} \pm SD$	r	P
	% by wt			% by wt		
18:2n-6	19.6 ± 2.36	-0.17	0.023	7.51 ± 1.46	0.14	0.09
18:3n-3	0.32 ± 0.10	0.03	0.68	0.09 ± 0.17	-0.06	0.47
20:3n-6	3.83 ± 0.69	-0.06	0.41	6.06 ± 0.92	0.13	0.10
20:4n-6	9.14 ± 1.31	-0.07	0.38	16.5 ± 1.63	-0.32	<0.001
20:5n-3	1.16 ± 0.62	0.42	<0.001	1.14 ± 0.30	0.36	<0.001
20:3n-9	0.21 ± 0.10	0.06	0.42	0.68 ± 0.30	-0.09	0.27
22:6n-3	8.80 ± 1.67	0.24	0.002	8.95 ± 1.65	0.13	0.11
22:4n-6	0.44 ± 0.10	-0.39	<0.001	0.77 ± 0.14	-0.24	0.002
22:5n-6	0.46 ± 0.19	-0.34	<0.001	0.76 ± 0.26	-0.28	0.001

Helsinki Declaration and was approved by the Faroese Committee on Research Ethics. All of the mothers provided written informed consent.

Blood analyses

Maternal serum was obtained from 173 of the participating women in connection with the last antenatal consultation at 34 wk of gestation. Blood from the umbilical cord was taken by the midwife in heparinized syringes with polytetrafluoroethylene-lined pistons; cord serum was available from 154 births. Milk for supplementary PCB analysis was obtained at days 4 and 5 after parturition.

Both maternal and cord serum samples were analyzed for LCPs in the phospholipid fraction (18). The analysis included essential LCPs and their elongation and desaturation products: LA, α -linolenic acid (18:3n-3), AA, EPA, docosahexaenoic acid (DHA; 22:6n-3), the 2 eicosatrienoic acids (20:3n-6 and 20:3n-9), docosatetraenoic acid (22:4n-6), and docosapentaenoic acid (DPA; 22:5n-6). A human control serum stored at -80°C was used to secure a day-to-day precision for each LCP within a CV of 5%. Results were computed as relative percentages by wt in the phospholipid fraction.

Maternal serum was analyzed for the main PCB congeners (16, 19). Because the 3 most prevalent PCB congeners (138, 153, and 180) constitute 50% of total PCBs, the sum of these 3 congeners was multiplied by 2.0 to obtain an approximate total PCB measure (20). Recovery of the PCB congeners averaged $\approx 70\%$. When expressed in relation to the lipid concentration, PCB concentrations were similar in serum and milk [according to standard laboratory procedures, previously published results (16) for milk analyses were adjusted whereas the serum data were not]. A milk result was available from 168 mothers and was used for 8 mothers for whom a serum PCB analysis was missing.

Statistical analyses

PCB concentrations were logarithmically transformed because of skewed distributions. Parametric methods were applied throughout. Multiple regression analysis was used to determine the relative importance of relevant predictors of the outcome variables. The covariates examined for possible confounding were smoking during pregnancy, diabetes, parity, length of gestation, birth weight, and the infant's sex. Covariates were kept in the final regression equation if statistically significant ($P < 0.1$) after backward elimination. The robustness of the final regression equation

was tested by inclusion of removed covariates one by one. All calculations were performed with SPSS for WINDOWS version 10.0 (SPSS Inc, Chicago).

RESULTS

Total LCP concentrations in maternal serum were, on average, ≈ 3 -fold those in cord serum (1.92 compared with 0.63 mg/L). AA concentrations in the paired serum samples were highly associated ($r = 0.49$, $P < 0.001$; $n = 151$), but the relative concentration of AA in the phospholipid phase was higher in cord serum than in maternal serum (Table 1). Detailed results were reported previously (17) and appear quite comparable to results obtained with similar methodologies from other populations without LCP deficiencies (2, 21–23). However, as expected for a population with a high intake of marine fats (4–6), EPA and DHA concentrations were comparatively high.

Total PCB concentrations ($n = 181$) spanned 2 orders of magnitude, with a geometric average of 1.12 $\mu\text{g/g}$ lipid. Simple correlations without adjustment for confounders showed a positive association between PCB concentrations and EPA and DHA concentrations as a probable reflection of their common dietary source (Table 1). However, as a departure from this expected pattern, AA concentrations in cord serum were strongly and negatively associated with PCB concentrations. A weaker and nonsignificant tendency (before confounder adjustment) was apparent for maternal serum. Also, an inverse association was seen between PCB concentrations and concentrations of the elongation and desaturation products docosatetraenoic acid and DPA.

Multiple regression analyses were carried out with AA concentrations in maternal and cord sera as the dependent variables. Maternal serum concentrations of LA, α -linolenic acid, and EPA were found to be the main determinants of the 2 sets of AA concentrations and were therefore included in all subsequent regression equations. The maternal serum AA concentration was used as an additional predictor of the cord serum AA concentration. Likewise, gestational age was included because it was positively associated with cord serum AA concentrations. Other obstetric covariates failed to show any clear associations and were therefore not considered further. Although AA concentrations were positively associated with maternal serum concentrations of the elongation and desaturation products docosatetraenoic acid and DPA and were negatively associated with 20:3n-9, addition of these LCPs to the models changed the PCB regression coefficients only negligibly.

TABLE 2

Differences between the arachidonic acid (AA) concentrations (in percentages by wt of phospholipids) in maternal and cord sera of subjects in the higher 2 tertiles of serum polychlorinated biphenyl (PCB) concentrations and those in maternal and cord sera of subjects in the lowest tertile ($<0.74 \mu\text{g/g lipid}$)¹

	0.74–1.49 $\mu\text{g PCB/g lipid}$	$>1.50 \mu\text{g PCB/g lipid}$
	% by wt	
Maternal serum ²	-0.22 ± 0.21	-0.43 ± 0.21
Cord serum ³	-0.50 ± 0.29	-0.68 ± 0.29

¹ $\bar{x} \pm \text{SD}$. Adjustment was made for maternal serum concentrations of linoleic, α -linolenic, and eicosapentaenoic acids. Cord serum concentrations were also adjusted for gestational age and maternal serum AA concentrations.

² P for trend = 0.019.

³ P for trend = 0.009.

The adjusted regression coefficients for the logarithmic transformation of the PCB concentrations were -0.53 and -1.02 for maternal and cord sera, respectively. The effect of doubling the PCB exposure may then be calculated by multiplying these coefficients by 0.301: the predicted decrease in the AA concentration (percentage by wt in phospholipids) was 0.17% (95% CI: 0.03%, 0.31%) and 0.31% (95% CI: 0.10%, 0.52%) for maternal and cord sera, respectively. The results for tertile groups of PCB exposure are shown in **Table 2**, and the association between PCB exposure and the confounder-adjusted AA concentrations in cord serum is shown in **Figure 1**. One observation of a low AA concentration (9.3% by wt) at a high PCB concentration (9.6 $\mu\text{g/g lipid}$) was clearly an influential data point. Exclusion of this data point caused an attenuation of the adjusted PCB regression coefficient to -0.86 ($P = 0.013$) as a predictor of cord serum AA concentration.

Because AA is formed by the action of Δ^5 -desaturation from 20:3n-6, which is formed from LA by means of Δ^6 -desaturation (2, 24), we calculated the ratios of the cord serum AA concentration to the cord serum concentrations of each of its 2 precursors. Both ratios were significantly and negatively associated with PCB concentrations ($r = -0.19$, $P = 0.02$ and $r = -0.18$, $P = 0.03$, respectively). This result is also in accordance with the regression analysis, which included adjustment for LA.

LCP status may be evaluated from the ratio of an essential LCP to the most closely related LCP that is synthesized by the body under insufficiency, eg, the ratio of DHA to DPA (2). This ratio increased significantly with the PCB concentration ($r = 0.23$, $P = 0.005$). Thus, in this population with a high intake of marine food, essential LCPs appear to be supplied in ample amounts along with PCBs, thus effectively ruling out any deficient intake as a matter of concern.

The total PCB measure used as the exposure biomarker in this study represents several related congeners. However, the further search for an etiologic agent responsible for the decreased AA concentrations was compromised by the strong collinearity between these persistent organochlorine compounds (16). Accordingly, none of the individual congener concentrations determined had an association with AA concentrations that differed significantly from that of the total PCB concentration.

DISCUSSION

The AA concentration in serum phospholipids represents the combined result of LCP intake and the formation, breakdown,

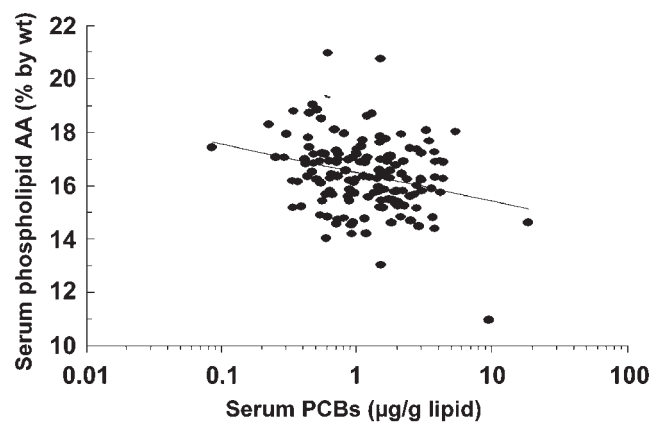


FIGURE 1. Association between polychlorinated biphenyl (PCB) exposure (total PCBs in maternal serum lipids) and concentrations of arachidonic acid (AA) in umbilical cord serum phospholipids adjusted to average gestational age and average maternal serum concentrations of linoleic, α -linolenic, arachidonic, and eicosapentaenoic acids.


and distribution of AA and its precursors in the body, which in the present study also involved transplacental transport. With ample supplies of essential LCPs, adults maintain serum AA concentrations within a relatively narrow range (4–6, 25). The significant association between maternal and cord serum AA concentrations (22), also replicated in the present study, suggests a substantial passage of AA or its precursors through the placenta. However, fetal growth results in high nutrient demands, as indicated by LCP concentrations in cord serum that average only one-third of those in maternal serum. Some LCP concentrations in maternal serum may even decrease during pregnancy (21), but the effect of this variable was minimized in the present study by taking the maternal blood samples at a specified time during pregnancy. Likewise, although only term births were included in the present study, gestational age was taken into account with regard to the cord serum LCP concentrations. Neither birth weight nor gestational age was associated with PCB concentrations (17), thus limiting the effect of these covariates on the main topic of the study.

LA is the main dietary precursor of AA, and a negative association between the 2 in maternal serum suggests that AA concentrations in the phospholipid fraction of the serum may be maintained at the expense of LA. However, unlike evidence from another study (23), this pattern was less obvious in the present study and was not significant in cord serum. In generating AA from LA, both Δ^5 - and Δ^6 -desaturation are necessary; the enzymes responsible for this desaturation are expressed in the fetal liver (15) to help maintain AA supplies during development (14). The enzymes are also involved in producing EPA from α -linolenic acid, and because of negative feedback, a high intake of EPA could decrease AA status (7, 8). These considerations were amply reflected in the regression analyses in which adjustments for LA and EPA were included as important covariates.

The coplanar PCB congener 126 has been shown to cause a decrease in AA concentrations in the liver of rats and an inhibition of both Δ^5 - and Δ^6 -desaturation in liver homogenates (12, 13). This experimental evidence therefore suggests a relevant toxic mechanism that has not previously been considered in humans.

However, in human serum, this particular congener occurs at very low concentrations (20) and was not included in the PCB analysis in the present study. As expected, all PCB congeners determined showed substantial collinearity (16, 17), and the results therefore do not provide evidence on the specific PCB congener or congeners that may affect serum AA concentrations. In addition, an observational study can provide only weak support for causal associations. However, of possible relevance is the report that tissue AA concentrations and accumulated PCBs are also strongly and negatively associated in dolphins (26). In concert, this evidence suggests that desaturation inhibition is a conceivable toxic mechanism for certain PCB congeners. Still, other types of interference with LCP metabolism should not be disregarded, because PCBs are thought to induce lipid peroxidation (27) and may affect AA release by phospholipase A2 (EC 3.1.1.4) activation (28).

These findings may be of clinical and toxicologic relevance, because AA can act as a second messenger, and a decreased AA supply may constitute a possible mechanism for the neurotoxicity, growth retardation, or other adverse effects known to result from developmental PCB exposure (10, 28). The fact that similar effects may be caused by LCP deficiencies (3, 29, 30) suggests that the possible interaction between PCBs and AA should be explored further.

In addition, LCP status may contribute to the known discrepancies in the findings of epidemiologic studies of PCB toxicity in populations with or without high intakes of seafood (10, 11, 31, 32). Thus, the 2 populations with the weakest indications of developmental PCB toxicity (11, 32) also had the largest seafood intake. The possible interaction between PCBs and LCP metabolism should therefore lead to a reexamination of data on AA status in regard to high intakes of marine food. 

We are indebted to members of the Faroese health care system for assistance in generating this cohort and in conducting the examinations of the infants. The cohort was formed as part of the European study Neonatal PCB—Exposure and Neurodevelopmental Deficit coordinated by G Winneke, Düsseldorf, Germany. JW Brock, B Heinzow, and K Bjerve made results available on serum PCBs, milk PCBs, and serum LCPs, respectively.

PW and PG planned the cohort study, PW supervised the clinical work, and PG evaluated the results and wrote the manuscript, which was then revised and approved by both authors. Neither of the authors have any conflicts of interest in regard to this study.

REFERENCES

- Hearn TL, Sgoutas SA, Hearn JA, Sgoutas DS. Polyunsaturated fatty acids and fat in fish flesh for selecting species for health benefits. *J Food Sci* 1987;52:1209–11.
- Hornstra G. Essential fatty acids in mothers and their neonates. *Am J Clin Nutr* 2000;71(suppl):1262S–9S.
- Carlson SE, Werkman SH, Peoples JM, Cooke RJ, Tolley EA. Arachidonic acid status correlates with first year growth in preterm infants. *Proc Natl Acad Sci U S A* 1993;90:1073–7.
- Bønaa KH, Bjerve KS, Nordøy A. Habitual fish consumption, plasma phospholipid fatty acids, and serum lipids: the Tromsø study. *Am J Clin Nutr* 1992;55:1126–34.
- Sanjurjo P, Matorras R, Perteagudo L. Influence of fatty fish intake during pregnancy in the polyunsaturated fatty acids of erythrocyte phospholipids in the mother at labor and newborn infant. *Acta Obstet Gynecol Scand* 1995;74:594–8.
- Andersen LF, Solvoll K, Drevon CA. Very-long-chain n–3 fatty acids as biomarkers for intake of fish and n–3 fatty acid concentrates. *Am J Clin Nutr* 1996;64:305–11.
- Carlson SE, Cooke RJ, Werkman SH, Tolley EA. First year growth of preterm infants fed standard compared to marine oil n–3 supplemented formula. *Lipids* 1992;27:901–7.
- Barham JB, Edens MB, Fonteh AN, Johnson MM, Easter L, Chilton FH. Addition of eicosapentaenoic acid to gamma-linolenic acid-supplemented diets prevents serum arachidonic acid accumulation in humans. *J Nutr* 2000;130:1925–31.
- de March BGE, de Wit CA, Muir DCG. Persistent organic pollutants. In: AMAP assessment report: arctic pollution issues. Oslo: Arctic Monitoring and Assessment Programme, 1998:183–371.
- Sauer PJJ, Huisman M, Koopman-Elseboom C, et al. Effects of polychlorinated biphenyls (PCBs) and dioxins on growth and development. *Hum Exp Toxicol* 1994;13:900–6.
- Grandjean P, Weihe P, Burse VW, et al. Neurobehavioral deficits associated with PCB in 7-year-old children prenatally exposed to seafood neurotoxicants. *Neurotoxicol Teratol* 2001;23:305–17.
- Matsusue K, Ishii Y, Ariyoshi N, Oguri K. A highly toxic PCB produces unusual changes in the fatty acid composition of rat liver. *Toxicol Lett* 1997;91:99–104.
- Matsusue K, Ishii Y, Ariyoshi N, Oguri K. A highly toxic coplanar polychlorinated biphenyl compound suppresses Delta5 and Delta6 desaturase activities which play key roles in arachidonic acid synthesis in rat liver. *Chem Res Toxicol* 1999;12:1158–65.
- Koletzko B, Decsi T, Demmelmair H. Arachidonic acid supply and metabolism in human infants born at full term. *Lipids* 1996;31:79–83.
- Rodriguez A, Sarda P, Nessmann C, et al. Fatty acid desaturase activities and polyunsaturated fatty acid composition in human liver between the seventeenth and thirty-sixth gestational weeks. *Am J Obstet Gynecol* 1998;179:1063–70.
- Steuerwald U, Weihe P, Jørgensen PJ, et al. Maternal seafood diet, methylmercury exposure, and neonatal neurological function. *J Pediatr* 2000;136:599–605.
- Grandjean P, Bjerve KS, Weihe P, Steuerwald U. Birth weight in a fishing community: significance of essential fatty acids and marine food contaminants. *Int J Epidemiol* 2001;30:1272–8.
- Bjerve KS, Fischer S, Alme K. Alpha-linolenic acid deficiency in man: effect of ethyl linolenate on plasma and erythrocyte fatty acid composition and biosynthesis of prostanoids. *Am J Clin Nutr* 1987;46:570–6.
- Brock J, Burse VW, Ashley DL, et al. An improved analysis for chlorinated pesticides and polychlorinated biphenyls (PCBs) in human and bovine sera using solid-phase extraction. *J Anal Toxicol* 1996;20:528–36.
- Grandjean P, Weihe P, Needham LL, et al. Effect of a seafood diet on mercury, selenium, arsenic, and PCBs and other organochlorines in human milk. *Environ Res* 1995;71:29–38.
- Al MD, van Houwelingen AC, Kester AD, Hasaart TH, de Jong AE, Hornstra G. Maternal essential fatty acid patterns during normal pregnancy and their relationship to the neonatal essential fatty acid status. *Br J Nutr* 1995;74:55–68.
- Otto SJ, Houwelingen AC, Antal M, et al. Maternal and neonatal essential fatty acid status in phospholipids: an international comparative study. *Eur J Clin Nutr* 1997;51:232–42.
- Ghebremeskel K, Crawford MA, Lowy C, et al. Arachidonic and docosahexaenoic acids are strongly associated in maternal and neonatal blood. *Eur J Clin Nutr* 2000;54:50–6.
- Nakamura MT, Cho HP, Xu J, Tang Z, Clarke SD. Metabolism and functions of highly unsaturated fatty acids: an update. *Lipids* 2001;36:961–4.
- Sanders TA, Reddy S. The influence of a vegetarian diet on the fatty acid composition of human milk and the essential fatty acid status of the infant. *J Pediatr* 1992;120:S71–7.
- Guitart R, Guerrero X, Silvestre AM, Gutierrez JM, Mateo R. Organochlorine residues in tissues of striped dolphins affected by the 1990 Mediterranean epizootic: relationships with the fatty acid composition. *Arch Environ Contam Toxicol* 1996;30:79–83.

27. Hassoun EA, Li F, Abushaban A, Stohs SJ. Production of superoxide anion, lipid peroxidation and DNA damage in the hepatic and brain tissues of rats after subchronic exposure to mixtures of TCDD and its congeners. *J Appl Toxicol* 2001;21:211–9.
28. Kodavanti PRS, Tilson HA. Neurochemical effects of environmental chemicals. *Ann N Y Acad Sci* 2000;919:97–105.
29. Agostoni C, Trojan S, Bellu R, Riva E, Giovannini M. Neurodevelopmental quotient of healthy term infants at 4 months and feeding practice: the role of long-chain polyunsaturated fatty acids. *Pediatr Res* 1995;38:262–6.
30. Willatts P, Forsyth JS, DiModugno MK, Varma S, Colvin M. Effect of long-chain polyunsaturated fatty acids in infant formula on problem solving at 10 months of age. *Lancet* 1998;352:688–91.
31. Jacobson JL, Jacobson SW. Intellectual impairment in children exposed to polychlorinated biphenyls in utero. *N Engl J Med* 1996;335:783–9.
32. Gladen BC, Rogan WJ. Effects of perinatal polychlorinated biphenyls and dichlorodiphenyl dichloroethene on later development. *J Pediatr* 1991;119:58–63.

