Early-Life Exposure to *p,p'*-DDT and *p,p'*-DDE in South African Children Participating to the VHEMBE Study: An Assessment Using Repeated Serum Measurements and Pharmacokinetic Modeling

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Highlights

• Children's *p,p′*-DDT/E levels at 12 and 24 months were higher than maternal levels.

• The pharmacokinetic model of children's lactational exposure showed good precision.

• Results suggested breastfeeding is the main contributor to children's exposure.

• Pharmacokinetic modeling can be used to estimate children's levels in sprayed areas.

ABSTRACT

Background: The World Health Organization recommends indoor residual spraying of insecticides (including dichlorodiphenyltrichloroethane [DDT]) to fight malaria vectors in endemic countries. There is limited information on children's exposure to DDT in sprayed areas, and tools to estimate early-life exposure have not been thoroughly evaluated in this context.

Objectives: To document serum *p,p'*-DDT/E levels in 47 mothers and children participating to the Venda Health Examination of Mothers, Babies and their Environment (VHEMBE) study, and to evaluate the precision and accuracy of a published pharmacokinetic model for the estimation of children's *p,p'*-DDT/E levels.

Methods: *p,p'*-DDT/E levels were measured in maternal serum at delivery, and in children's serum at 12 and 24 months of age. A pharmacokinetic model of gestational and lactational exposure was used to estimate children's *p,p'*-DDT/E levels across pregnancy and the first two years of life, and estimated levels were compared to measured levels.

Results: The geometric means of children's serum *p,p'*-DDT/E levels at 12 and 24 months were higher than maternal serum levels. Regression models of measured

children's *p,p'*-DDT/E levels vs. levels estimated with the pharmacokinetic model (which only accounted for children's exposure through placental transfer and breastfeeding) had coefficients of determination (R^2) of 0.75 and 0.82. Estimated *p,p'*-DDE levels were not significantly different from measured levels, whereas *p,p'*-DDT levels were overestimated by 36% at 12 months, and 51% at 24 months.

Conclusion: Results indicate children living in a sprayed area have serum *p,p'*-DDT/E levels exceeding their mothers' during the first two years of life. The pharmacokinetic model may be useful to estimate children's levels in the VHEMBE population.

INTRODUCTION

The World Health Organization (WHO) recommends indoor residual spraying to fight malaria vectors in endemic countries and includes dichlorodiphenylthrichloroethane (DDT) in their list of recommended insecticides (WHOPES 2015). In many countries, including South Africa, DDT has been used for decades (Mabaso et al. 2004), resulting in elevated body burdens of DDT and its breakdown product dichlorodiphenyl-dichloroethylene (DDE) in individuals living in sprayed homes (Gaspar et al. 2017). Concerns have been raised regarding the potential health effects related to exposure to these chemicals, especially in developing children (Bouwman and Kylin 2009; Eskenazi et al. 2009).

Because of their biological half-lives in the order of years (Ritter et al. 2009; Smith 1999; Wolff et al. 2000), *p,p'*-DDT/E accumulate in human tissues. *p,p'*-DDT/E have been shown to transfer to children during pregnancy (Aylward et al. 2014) and breastfeeding

(LaKind et al. 2009), resulting in exposures during critical windows of development. Multiple studies have provided evidence of the substantial impact of breastfeeding on children's *p,p'*-DDE levels. Whereas neonate's serum *p,p'*-DDT/E levels are similar or slightly lower than maternal levels at birth on a lipid basis (Aylward et al. 2014), studies with longitudinal measurements have shown that children's serum levels can increase substantially during breastfeeding (Lackmann 2006). Breastfed children have higher *p,p'*-DDE levels than children who have been fed formula, from as early as 6 weeks of age (Lackmann et al. 2004) to as late as 14 years (Gascon et al. 2015). Longer breastfeeding durations have been associated with higher children *p,p'*-DDE levels during the first years of life (Caspersen et al. 2016; Ribas-Fito et al. 2005). It has been estimated that the dose of lipophilic persistent organic pollutants like *p,p'*-DDT/E received by the child through breastfeeding can be substantially higher than the maternal daily dose (Haddad et al. 2015), primarily due to their long biological half-life and partitioning into breast milk lipids.

The scientific literature on children's exposure to *p,p'*-DDT/E in sprayed areas is scarce. In addition to placental transfer and breastfeeding, children living in houses sprayed with *p,p'*-DDT may be exposed through direct contact with contaminated dust. In a previous study of undisturbed dust from houses in the Limpopo province of South Africa, the majority of dust samples from sprayed houses (82%) had quantifiable levels of *p,p'*-DDT (Gaspar et al. 2016). To our knowledge, no study has obtained repeated *p,p'*-DDT/E levels during early childhood in the context of indoor residual spraying. While tools have been developed to estimate children's early-life exposure to *p,p'*-DDT/E (Gyalpo et al.

2012; Verner et al. 2009; Verner et al. 2013), their ability to estimate children's body burden has not been thoroughly evaluated in this context.

In this study, we aimed to i) document serum *p,p'*-DDT/E levels in a subset of mothers and children (n=47) from the Limpopo province participating to the Venda Health Examination of Mothers, Babies and their Environment (VHEMBE) study; and to ii) evaluate the precision and accuracy of a published pharmacokinetic model for the estimation of children's *p,p'*-DDT/E levels during the first two years of life.

METHODS

Study participants

The Venda Health Examination of Mothers, Babies and their Environment (VHEMBE) study population is detailed in Gaspar et al. (2017). Briefly, women who presented at Tshilidzini Hospital (Thohoyandou, Limpopo, South Africa) for delivery between August 2012 and December 2013 were eligible if they were ≥ 18 years old, spoke Tshivenda at home, lived within 20 km of the hospital, planned to remain in the area, had not been diagnosed with malaria during pregnancy, had contractions >5 minutes apart, and gave birth to a viable singleton. A total of 920 out of the 1,649 women who were approached were eligible. Out of the eligible women, 152 refused to participate, 14 did not have a baseline questionnaire, and blood volume was insufficient for *p,p'*-DDT/E analysis in 3 women, resulting in a total of 751 participants. Blood was sampled prior to delivery (n=595) or immediately after delivery (n=156) (range: four days prior to delivery to one day after delivery). Breastfeeding mothers provided breast milk samples by expressing

directly in sterile collection bottles at the one-week home visit (n=706) and at the 12-month field office visits (n=532). In addition, study nurses collected venous blood from children via venipuncture during visits to the field office at 12 months (n=642) and 24 months (n=662). Blood and breast milk samples were processed immediately after collection and stored at -80C until shipment to analytical laboratories on dry ice. Due to budgetary restrictions, children's serum p,p'-DDT/E levels could only be measured in a subset of the cohort, and breast milk p,p'-DDT/E levels could only be measured at one collection time; we selected the 12-month samples because milk is fully mature at this point (versus transition milk at one week postpartum), and it could potentially reflect maternal exposure after delivery (whereas early breast milk levels likely reflect exposure prior to delivery). Serum and breast milk samples were selected from a subset of 47 mother-child dyads based on i) availabiliby of *p,p'*-DDT or *p,p'*-DDE levels in maternal serum (previous analyses), ii) sufficient children's serum volume for *p,p'*-DDT/E analyses, and iii) availability of a breast milk sample at 12 months postpartum for *p,p'*-DDT/E analyses. All mothers or legal guardian provided informed consent prior to data and sample collection. Protocols were reviewed and approved by institutional review boards at the Université de Montréal, University of California at Berkeley, McGill University, the University of Pretoria, the Limpopo Department of Health and Social Development, and the Tshilidzini Hospital.

Maternal and child characteristics

Questionnaires were administered before hospital discharge, at the one-week home visit, and during the 12 month and 24 month visits at the field office. Mothers were weighed at study enrollment and at the 12-month and 24-month visits using a Tanita HD-351 scale (Tokyo, Japan). Birth weight was measured using a Tanita BD-815U neonatal scale (Tokyo, Japan) in the delivery suite shortly after delivery, and children were weighed at 12-month and 24-month visits using a Tanita BD-590 pediatric scale (Tokyo, Japan). Information on duration of breastfeeding was collected at 12-month and 24-months visits. The total duration of breastfeeding was approximated based on the question "How many months old was [child's name] when you stopped breastfeeding completely?" The duration of exclusive breastfeeding was approximated based on the question "How many months did you exclusively breastfeed, that is, for how many months did [child's name] only receive breast milk without any additional food or water? Data on spraying was collected through questionnaires at study enrollment, and at 12 and 24-month visits to the field office.

Measurement of serum *p,p'*-DDT/E levels

Maternal serum *p,p'*-DDT/E levels were measured at the Environmental Health Laboratory at Emory University's Rollins School of Public Health using gas chromatography-tandem mass spectrometry (GC-MS) with isotope dilution quantification (Barr et al. 2003). Limits of detection were 0.01 μg/L for *p,p'*-DDT and 0.03 μg/L for *p,p'*-DDE. Quality control samples (i.e., sealed blanks, field blanks, and spiked samples)

were also analyzed. Triglycerides and total cholesterol were measured using standard enzymatic methods (Roche Chemicals, Indianapolis, USA). Breast milk and children's serum concentrations of *p,p'*-DDT/E were measured by the Centre de Toxicologie du Québec (CTQ) of the Institut national de santé publique du Québec (INSPQ) by chromatography mass spectrometry (GC-MS) as described in Fisher et al. (2016). The limits of detection for both compounds were between 0.01 to 0.05 μg/L for child serum and 0.2 μg/L for breast milk. Lipids in children's serum samples levels were measured by enzymatic methods combined with colorimetry at the laboratory of Centre Hospitalier de l'Université Laval (Quebec, Canada). Breast milk lipid content was assessed by extraction, evaporation and gravimetric measurement. Maternal and child serum *p,p'*-DDT/E levels were expressed on a lipid basis based on triglyceride and total cholesterol levels (Phillips et al. 1989). Breast milk levels were also expressed on a lipid basis based on total lipids.

Pharmacokinetic modeling

We used a published and validated pharmacokinetic model to estimate children's *p,p'*-DDT/E levels. The model is presented in detail in Verner et al. (2013). Briefly, the model included two compartments representing the maternal and child lipids in which *p,p'*-DDT/E are assumed to distribute primarily given their high lipophilicity. The compartments were connected through placental transfer and breastfeeding. During pregnancy, maternal and fetal concentrations were assumed to be equal on a lipid basis. Transfer through breastfeeding was based on breast milk consumption, breast milk lipid content and maternal *p,p'*-DDT/E levels, assuming *p,p'*-DDT/E levels in breast milk

lipids were equal to levels in maternal lipids. Elimination in the mother and child was based on published biological half-lives. The pharmacokinetic model allows simulating maternal *p,p'*-DDT/E levels from maternal birth onwards, and child's levels during gestation, breastfeeding, and after breastfeeding has ceased.

To simulate *p,p'*-DDT/E levels for each participating mother-child dyad, the pharmacokinetic model incorporated individual-specific information on the mother (i.e., age at delivery, weight at admission for delivery, and weight at 12 months and 24 months postpartum), the child (i.e., sex, birth weight, and weight at 12 months and 24 months of age), the duration of exclusive and total breastfeeding, and the maternal serum *p,p'*-DDT/E levels. 16 out of the 47 mothers were only seen post-delivery; for these participants, body weight at delivery was estimated using a linear regression model of pre- and post-delivery body weight based on data from participants with both measurements. Prepregnancy weight and gestational weight gain were not available. We assumed a 16 kg weight gain during pregnancy for all participants based on a previous study of South African women (Wrottesley et al. 2017). During non-exclusive breastfeeding, we assumed milk intake to be half the intake during exclusive breastfeeding. For each mother-child dyad and compound, the model was run iteratively to estimate a maternal daily dose (absorbed) yielding a simulated maternal level matching the measured level. Then, the model was run using the estimated maternal daily dose to generate a time-course of maternal, child and breast milk levels from maternal birth until two years after delivery. These simulations provided complete profiles of p,p'-DDT/E for each mother-child dyad (see example in Figure 1).

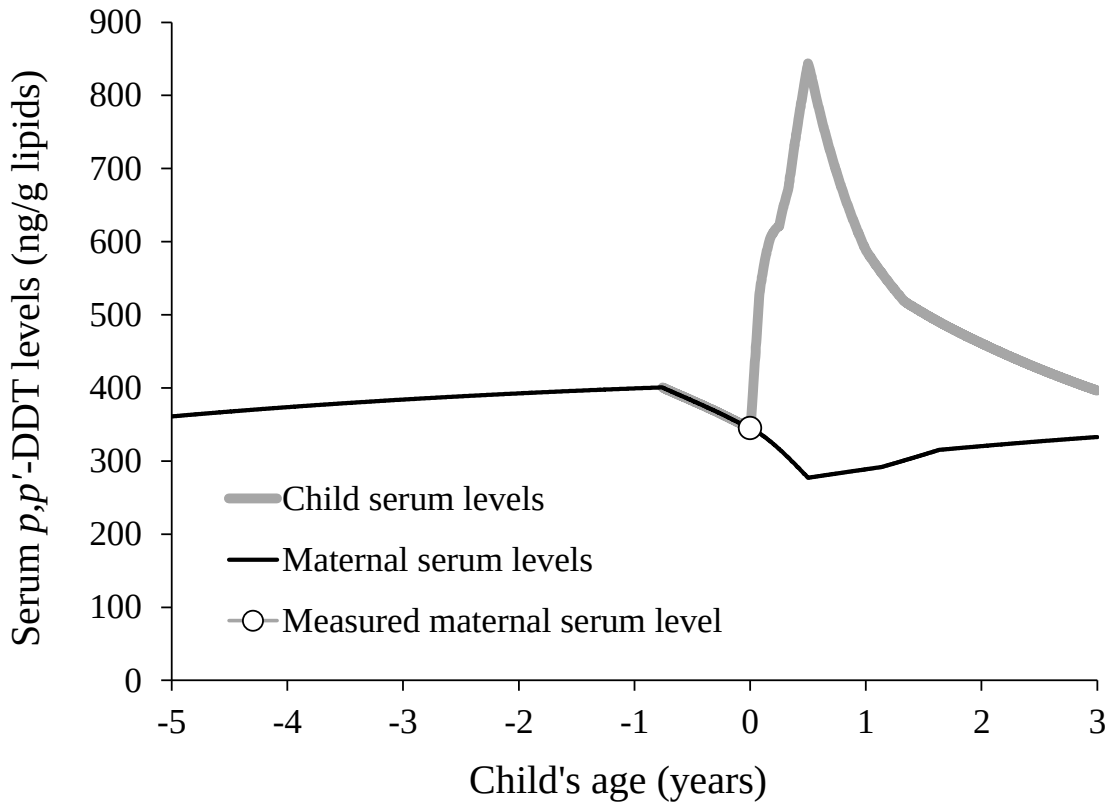

Figure 1. Example of a pharmacokinetic profile for a mother-child dyad where the mother was 25 years of age at delivery, had a serum concentration of 385 ng/g lipids at delivery and breastfed her child exclusively for 6 months.

Simulations were performed for dyads with maternal serum levels above the limit of detection for *p,p'*-DDT (n=46) or *p,p'*-DDE (n=47). We set out to perform a first set of simulations where children were exposed solely through placental transfer and breastfeeding, with the possibility to perform a second set of simulations including environmental exposure (e.g., dust ingestion) in the child if estimated levels were below measured levels in the first set of simulations.

Statistical analyses

To compare maternal serum *p,p'*-DDT/E levels at delivery to children's levels at 12 months and 24 months, we first calculated child/mother ratios for each mother-child dyad with levels above the limit of detection (one dyad with maternal serum *p,p'*-DDT level below the limit of detection was excluded from *p,p'*-DDT analyses), and subsequently calculated the geometric mean of child/mother ratios at 12 months and 24 months. We also performed Pearson correlation analyses between maternal, child and breast milk levels of *p,p'*-DDT/E (levels were $\log_{10}$-transformed prior to correlation analyses).

To evaluate pharmacokinetic model precision and accuracy, we performed linear regression analyses of measured levels vs. simulated levels (for children's serum levels and breast milk levels). Precision was assessed by calculating coefficients of determination (R^2); a precise model would yield a R^2 close to 1. Accuracy was assessed in terms of β coefficients and intercepts; an accurate model would yield a β coefficient with 95% confidence intervals including 1, and an intercept with a 95% confidence interval including 0 (no systematic bias). We also calculated mean relative errors to complement these measurements of model accuracy. Relative errors were calculated as follows (Equation 1):

$$Relative\ error_i = 100 \times \frac{Estimated\ level_i - Measured\ level_i}{Measured\ level_i}$$

RESULTS

Children participating to this study were born to mothers who were between 18 and 39 years of age at delivery (Table 1). The geometric mean total duration of breastfeeding was 18.7 months, whereas the geometric mean duration of exclusive breastfeeding was less than 2 months. One mother (2%) reported spraying with DDT during pregnancy, six mothers (13%) reported spraying with DDT during the first year after birth, and six mothers (13%) reported spraying with DDT during the second years after birth. *p,p'*-DDT/E were detected in all serum and milk samples, except for one maternal serum *p,p'*-DDT level that was below the limit of detection. The geometric means of *p,p'*-DDT/E levels were higher in children's serum than maternal serum at 12 months and 24 months (Table 1). 95% of child/mother serum *p,p'*-DDT/E level ratios were above 1, indicating that children's *p,p'*-DDT/E levels at 12 months and 24 months were mostly higher than maternal levels at delivery. The geometric means of child/mother level ratios were higher for *p,p'*-DDE (3.6 at 12 months, and 4.1 at 24 months) than for *p,p'*-DDT (2.5 at 12 months, and 2.4 at 24 months) (Table 2). The maximum child/mother level ratios were 42 for *p,p'*-DDT and 32 for *p,p'*-DDE.

Table 1. Characteristics of study participants, and DDT/E levels in serum and breast milk.

Characteristic	N (%)	GM	GSD	Range
Maternal age at delivery (years)		26	1.2	18-39
Maternal body weight at delivery (kg)		74	1.2	55-119
Maternal body weight (kg) 12 months after delivery		67	1.3	46-118
Maternal body weight (kg) 24 months after delivery		69	1.2	44-113
Sex of child				
Female	20 (43)			
Male	27 (57)			

Birth weight (kg)		3.1	1.2	1.7-4.1
Child weight at 12 months (kg)		9.1	1.2	6.6-12.1
Child weight at 24 months (kg)		11.2	1.1	9.1-13.5
Duration of exclusive breastfeeding (months)		1.7	3.3	0.1-8.0
Duration of total breastfeeding (months)		18.7	1.2	11.9-25.3
Maternal serum levels (ng/g lipids)				
p,p'-DDT		90	4.6	<LOD-1,816
p,p'-DDE		338	3.9	31-7,440
Children's serum levels at 12 months (ng/g lipids)				
p,p'-DDT		229	5.5	11-11,893
p,p'-DDE		1215	4.4	123-19,772
Children's serum levels at 24 months (ng/g lipids)				
p,p'-DDT		221	5.2	16-15,299
p,p'-DDE		1377	4.1	145-33,605
Breast milk levels at 12 months (ng/g lipids)				
p,p'-DDT		114	5.4	8-5,301
p,p'-DDE		409	4.6	28-5,984

GM: geometric mean; GSD: geometric standard deviation; LOD: limit of detection.

Table 2. Ratio between children's serum levels at 12 months and 24 months, and maternal levels at delivery.

Compound	N	Ratio child (12 mo)/mother GM (min-max)	Ratio child (24 mo)/mother GM (min-max)
p,p'-DDT	46	2.5 (0.7-42)	2.4 (0.3-34)
p,p'-DDE	47	3.6 (0.8-32)	4.1 (0.9-25)

GM: geometric mean.

Maternal serum, children's serum and breast milk levels were highly correlated, with Pearson correlation coefficients ranging from 0.862 to 0.984 (Table 3). *p,p'*-DDT and *p,p'*-DDE levels were highly correlated within each matrix, with correlation coefficients ranging from 0.895 to 0.923.

Regression models of measured children's p,p'-DDT/E levels vs. levels estimated with the pharmacokinetic model had coefficients of determination (R^2) ranging between 0.75 and 0.82 at 12 months and 24 months (Figure 2 and Table 4). Coefficients of determination (R^2) for children's levels at 24 months of age were only slightly lower than at 12 months of age. Linear regression models of measured vs. estimated children's *p,p'*-DDT/E levels had β coefficients ranging from 0.898 to 0.968, which were not statistically different from 1. The only intercept that was significantly >0 was for *p,p'*-DDE levels at 24 months (Table 4). Mean relative errors were not significantly different from 0 for *p,p'*-DDE. However, they were significantly greater than 0 for *p,p'*-DDT levels, indicating that children's *p,p'*-DDT levels were overestimated by the pharmacokinetic model. Because estimated *p,p'*-DDT/E levels were either accurate or overestimated in simulations only accounting for placental transfer and breastfeeding, we did not include additional environmental exposure (e.g., dust ingestion, dermal contact) in our model simulations. Relative errors were not significantly different in males and females (results not shown).

Regression models of measured breast milk *p,p'*-DDT and *p,p'*-DDE levels vs. levels estimated with the pharmacokinetic model had R^2s of 0.73 and 0.75 at 12 months postpartum, respectively (Figure 2 and Table 4). Although β values for regression models of measured vs. estimated breast milk levels were not significantly different from one, intercepts for both compounds were <0. Mean relative errors indicated that estimated breast milk levels were underestimated by ~30%.

Table 3. Pearson correlations between log10-transformed serum and breast milk *p,p'*-DDT/E levels.

		***p,p'*-DDT**				***p,p'*-DDE**			
		Maternal serum	Breast milk	Child serum (12 mo)	Child serum (24 mo)	Maternal serum	Breast milk	Child serum (12 mo)	Child serum (24 mo)
***p,p'*-DDT**	Maternal serum	-							
	Breast milk	0.862	-						
	Child serum (12 mo)	0.878	0.947	-					
	Child serum (24 mo)	0.866	0.928	0.984	-				
***p,p'*-DDE**	Maternal serum	0.895	0.819	0.799	0.788	-			
	Breast milk	0.759	0.912	0.810	0.850	0.868	-		
	Child serum (12 mo)	0.827	0.904	0.907	0.877	0.901	0.941	-	
	Child serum (24 mo)	0.829	0.912	0.924	0.923	0.889	0.916	0.981	-

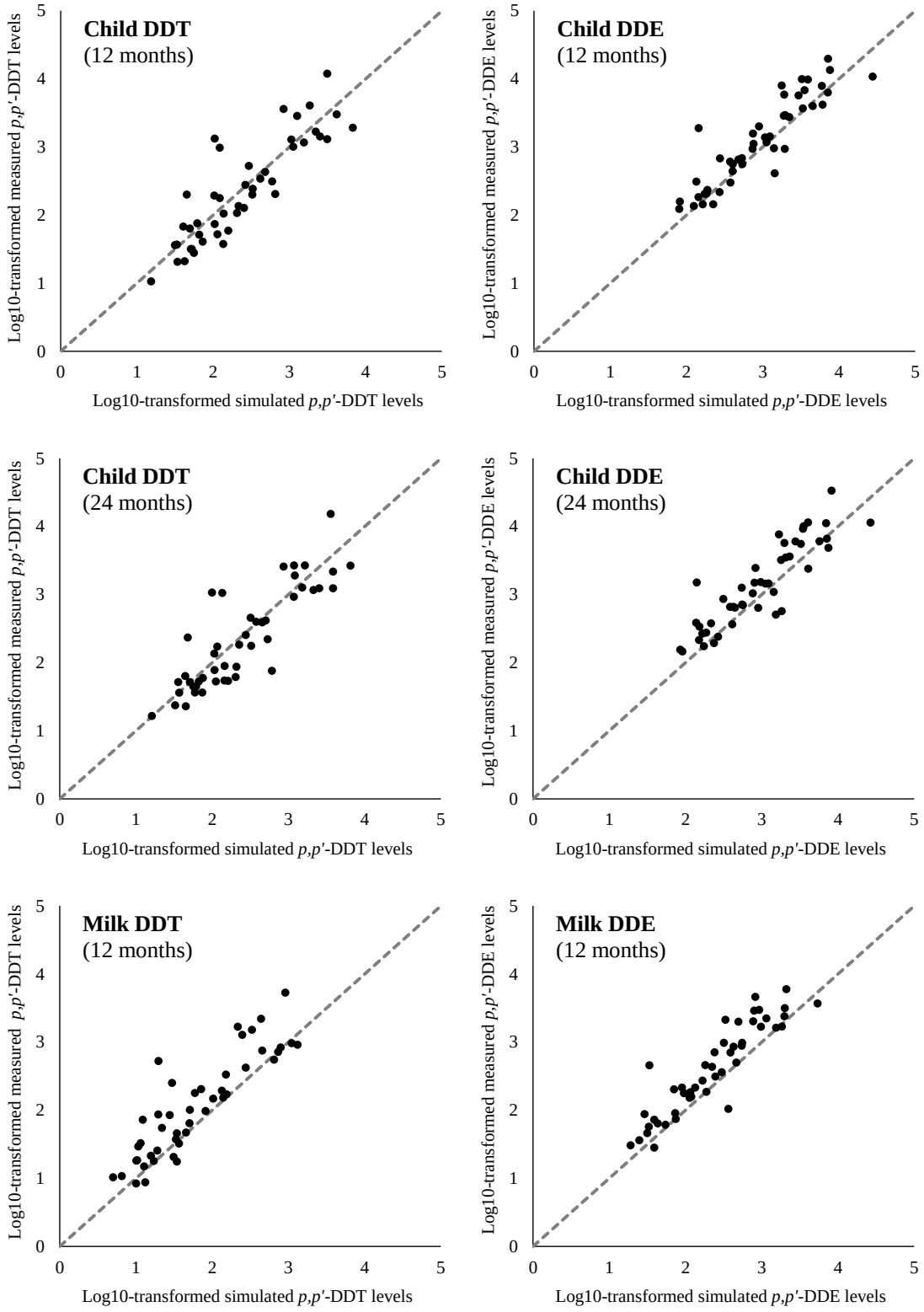

Figure 2. Measured *p,p'*-DDT/E levels vs. levels estimated using the pharmacokinetic model. Note: axes are $\log_{10}$-transformed.

Table 4. Linear regression of measured vs. estimated *p,p'*-DDT/E levels. Levels were $\log_{10}$-transformed prior to analyses.

	Intercept (95% CI)	**β (95% CI)**	**R^2**	**Mean relative error (%) (95% CI)**
***p,p'*-DDT**				
Children's serum levels (12 months)	0.051 (-0.349, 0.451)	0.968 (0.806, 1.130)	0.77	36 (11, 61)
Children's serum levels (24 months)	0.096 (-0.317, 0.510)	0.938 (0.772, 1.105)	0.75	51 (13, 89)
Breast milk levels (12 months)	-0.980 (-1.299. -0.661)	0.914 (0.747, 1.082)	0.73	-31 (-44, -18)
***p,p'*-DDE**				
Children's serum levels (12 months)	0.320 (-0.079, 0.719)	0.936 (0.804, 1.069)	0.82	-11(-29, 6)
Children's serum levels (24 months)	0.475 (0.056, 0.894)	0.898 (0.760, 1.037)	0.79	-17 (-35, 2)
Breast milk levels (12 months)	-0.970 (-1.361, -0.580)	0.925 (0.765, 1.085)	0.75	-32 (-47, -17)

The maximum children's concentrations estimated by the pharmacokinetic model had geometric means of 253 ng/g lipids (range: 16 to 6,888) for *p,p'*-DDT and 952 ng/g lipids (range: 85 to 28,217) for *p,p'*-DDE. Estimated daily lactational exposure (ng/kg/day) at ~one month postpartum (time when the daily dose on a body weight basis is the highest) was compared to estimated maternal daily dose (ng/kg/day); the geometric mean lactational/maternal dose ratio was 77 for *p,p'*-DDT and *p,p'*-DDE. In other words, nursing infants were exposed to daily doses that were 77 times greater than the maternal daily dose on a body weight basis.

DISCUSSION

In this study of South African children's exposure to *p,p'*-DDT/E, we found that children's serum levels measured at 12 months and 24 months of age were substantially higher than their mothers' at delivery. These findings are consistent with previous reports of increasing serum *p,p'*-DDE levels in breastfed infants from six weeks to six months of life (Lackmann 2006), a period during which all children included in our study were breastfed. In addition to lactational exposure, direct environmental exposure (e.g., food intake, dust/soil ingestion, dermal contact) could contribute to children's *p,p'*-DDT/E levels. However, simulations using the pharmacokinetic model, which only accounted for placental transfer and breastfeeding, could explain most if not all of the measured children's *p,p'*-DDT/E levels at 12 months and 24 months of age (mean relative errors were equal or greater than 0, see Table 4). Estimated daily doses through lactation (ng/kg/day) were >70 times higher than estimated maternal daily doses expressed in the same units. Similar conclusions with regards to the contribution of breast milk to children's body burden were drawn by Bouwman et al. (1992) based on children's blood and breast milk levels from the Kwazulu (now Kwazulu-Natal) area of South Africa.

R^2s obtained for the regression models of measured vs. estimated *p,p'*-DDT levels (0.77 at 12 months, and 0.75 at 24 months [n=46]) were higher than those obtained in a previous validation exercise in Inuit children (0.47 at 6 months [n=103]) and Slovak children (0.58 at 6 months [n=216], and 0.47 at 16 months [n=717]). R^2s obtained for *p,p'*-DDE levels (0.82 at 12 months, and 0.79 at 24 months [n=47]) were similar to that in Inuits (0.83 at 6 months [n=156]), but higher than those obtained in Slovaks (0.49 at 6 months [n=216], and 0.44 at 16 months [n=728]). Mean relative errors indicated that

children's *p,p'*-DDT levels were overestimated by 36% at 12 months, and 51% at 24 months (Table 4). Similarly, children's *p,p'*-DDT levels were overestimated in the previous validation effort in Inuits (73% at 6 months) and Slovaks (127% at 6 months, and 68% at 16 months) (data not published). This overestimation may be due to certain pharmacokinetic parameters, including the assumption that 100% of *p,p'*-DDT lactational dose is absorbed by the child. On the other hand, β coefficients for linear regressions of measured vs. simulated *p,p'*-DDT levels were not significantly different from 1, and intercepts were not significantly different from 0. Of note, confidence intervals around measures of accuracy may be inflated given the small sample size. Therefore, caution should be exerted when interpreting results based on statistical tests in this study.

Our study has a number of limitations. Although the assumption of 100% absorption in the gastro-intestinal tract is supported by studies of other lipophilic persistent organic pollutants like dioxins and polychlorinated biphenyls (McLachlan 1993), it is possible that absorption is lower for *p,p'*-DDT/E. The assumption that *p,p'*-DDT/E levels are equal in maternal and breast milk lipids may also be an oversimplification, as suggested in a study of paired serum and milk samples (LaKind et al. 2009). Many parameters including diffusion across the membranes and lipid composition in serum and breast milk could influence *p,p'*-DDT/E partitioning and explain the observed underestimation of breast milk levels. However, we decided not to calibrate the pharmacokinetic model parameters like absorption and milk:serum partition coefficients based on measured children's levels to avoid overfitting, which would have reduced model generalizability. Another limitation of our study is that we did not include individual-specific data on enzyme polymorphism and folate status, which may influence half-life (Denk and

Milutinovic 2018). Finally, it is important to note that R^2s for measured and estimated children's levels may also account for other environmental and lifestyle factors that are shared between mothers and children. For example, consumption of chicken foraging around the dwelling (Van Dyk et al. 2010) and their eggs (Bouwman et al. 2015) may be related among family members and contribute similarly to maternal and child body intake.

We believe the pharmacokinetic model used herein could be employed to estimate serum levels in children participating to the VHEMBE study whose serum was not analyzed. Not only can the pharmacokinetic model estimate levels at 12 months and 24 months, it can generate complete time courses of children's levels across the first two years of life. Multiple exposure metrics can be derived from these time courses like monthly levels, maximum levels, and area under the curve (cumulative exposure) during specific periods (see Figure 1). Such exposure metrics will allow evaluating associations between exposures during prenatal and postnatal windows of development and health outcomes, an approach that has previously been used to identify windows of vulnerability to toxic insults (Iszatt et al. 2015; Jusko et al. 2016; Rosenquist et al. 2017; Verner et al. 2010). Estimated *p,p'*-DDT levels could be mathematically corrected for the systematic overestimation (lack of model accuracy) based on calculated mean relative errors (see Table 4). For example, multiplying estimated children's *p,p'*-DDT levels by 0.69 would correct for the average 44% overestimation by the pharmacokinetic model, while maintaining model precision.

In conclusion, our study showed that children from a sprayed area in South Africa had serum *p,p'*-DDT/E at 12 months and 24 months of age exceeding their mothers' at delivery. We also demonstrated that a published pharmacokinetic model of placental and lactational transfer allows estimating children's serum *p,p'*-DDT/E levels with a precision ranging from 75 to 82%. However, indicators of accuracy suggested that estimated children's serum *p,p'*-DDT were overestimated. Because the pharmacokinetic model was developed *a priori*, and because results suggested that children's body burden during the first two years is mostly attributable to mother-child transfer, the pharmacokinetic model could be considered to be transportable to comparable populations with similar cultural practices and where indoor residual spraying of DDT is still used to fight malaria.

ACKNOWLEDGMENTS

We would like to thank study participants. This study was funded by the National Institute of Environmental Health Sciences, Award ID: 1R01ES020360-01. Marc-André Verner is the recipient of a Research Scholar J1 Award from the Fonds de recherche du Québec – Santé (FRQS).

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