

Organochlorine Pesticide Levels in Umbilical Cord Blood of Newborn in Veracruz, Mexico

Margarita Herrero-Mercado · S. M. Waliszewski ·
M. Caba · C. Martínez-Valenzuela ·
F. Hernández-Chalate

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Abstract Organochlorine pesticides accumulate in lipid rich compartments of organisms. During pregnancy, the compounds pass through the placental barrier appear in cord blood. The aim of this study was to monitor the levels of organochlorine pesticides in 70 umbilical cord blood samples taken during deliveries in Veracruz in 2009. For organochlorine pesticides, only the presence β -HCH (4%, 3.9 µg/L median concentration on wet weight), *pp'*DDE (100%, 0.7 µg/L) and *pp'*DDT (4%, 1.4 µg/L) were detected. The total pool of samples divided according to sex of new born babies, showed no statistical differences among median concentrations. The number of deliveries considered as a determinant contamination factor affirmed there were no statistical differences among median concentrations; however *pp'*DDE levels increased from the one to two childbirth groups. Age of pregnant women as a discriminate factor manifests in a significant increase in contamination levels among first, second and third tertile.

M. Herrero-Mercado
Biomedical Sciences Doctor Study,
University of Veracruz, Xalapa, Mexico

S. M. Waliszewski (✉)
Institute of Forensic Medicine, University of Veracruz,
SS Juan Pablo II s/n, 91890 Boca del Rio, Mexico
e-mail: swal@uv.mx

M. Caba
Directory of Scientific Research,
University of Veracruz, Xalapa, Mexico

C. Martínez-Valenzuela
Department of Biological Sciences,
West University, Los Mochis, Sin, Mexico

F. Hernández-Chalate
IMSS Hospital Veracruz, Veracruz, Mexico

In general, umbilical cord blood samples in Veracruz contained organochlorine pesticides, especially *pp'*DDE, confirming the presence of these compounds in the environment and their transfer from the mother to the developing fetus.

Keywords Organochlorine pesticides · Cord blood

Organochlorine pesticides are considered to be persistent and ubiquitous environmental contaminants. Their lipophilic nature, resistance to biodegradation and slow bio-transformation rate allow their concentration in adipose tissues in animals, humans and in all elements of the food chain, which in turn elevate concern about the magnitude of potential human exposure. Persons, who live in fumigated areas, need major attention because pesticides volatilize from their point of deposition, providing potential exposures via the respiratory route (Alegria et al. 2008; Wong et al. 2008; Martínez-Valenzuela et al. 2009). Organochlorine pesticides accumulated previously in soils off-gas and migrate to the whole environment, revealing their presence in human adipose tissue, breast milk and blood serum.

The lipophilic property of organochlorine pesticides causes them to accumulate in lipid rich compartments of organisms in accordance with equilibrium patterns. In humans, this process involves the transport and equilibrium between adipose tissue and blood serum, thus yielding a distribution within body compartments and in lipid rich fractions. The liposolubility rate influences the accumulation and elimination of these pesticides from tissues and organs. Therefore, a difference in concentrations between body compartments depends principally upon the tissues' lipid content. These lipid rich compartments act as

reservoirs for lipophilic pesticides and provide data for exposure assessment and for evaluation of health risks (Waluszewski et al. 2000, 2004a, b, 2005a).

The human placental barrier is a selective filter but permits the flow of low molecular weight lipophilic or unionized molecular compounds, such as organochlorine pesticides. Their toxicity may compromise placental functions and contribute to developmental problems in offspring, placental vascular lesions, fetal growth retardation, or death (Hamel et al. 2003). Being subjected to organochlorine pesticide residues before birth is also linked with cases of developmental neurotoxicity and intellectual deficiencies such as learning and memory disabilities (Torres-Sánchez et al. 2007).

Exposure to organochlorine compounds has been a subject of interest in recent years given their potential toxicity (carcinogenicity, immunotoxicity, reproductive system illnesses, and neurotoxicity). Contrary to what happens in adults, exposure to neurotoxicants during the windows of vulnerability in critical periods of brain development can produce permanent cerebral dysfunction from infancy, or it can delay its effect until adulthood (Ortega García et al. 2006; Torres-Sánchez et al. 2007).

The storage of organochlorine pesticide residues in fat is a problem of greater concern in women because of their higher percentage of body fat. Hormonal changes that occur during pregnancy, lactation and menopause, results in the internal mobilization of pollutants many years after initial exposure. During pregnancy, concern over these pesticide levels in maternal and cord blood becomes greater since reproductive toxicological studies have revealed that fetuses, especially during organogenesis, are more vulnerable to their toxic action (Waluszewski et al. 2005b). Newborns are exposed through placental transmission as well as breastfeeding. Hence, knowledge of biological levels of these contaminants in human tissues is necessary for the risk assessment of possible adverse health effects and for the identification of vulnerable groups (Pathak et al. 2008).

The purpose of the present study was to monitor the levels of organochlorine pesticides in umbilical cord blood that occurred as a consequence of environmental maternal exposure in areas where these pesticides were sprayed to combat vector transmitted diseases.

Materials and Methods

Seventy selected volunteer mothers admitted for delivery during 2009 were asked to donate umbilical cord blood for determination of organochlorine pesticide contents. The donors of mean 27 years old originated from the state of Veracruz. The volunteers signed their consent to participate

in the study. The blood samples were taken from the umbilical cord after it was cut. The samples were then centrifuged to separate blood cells from the serum. The serum samples were stored in glass jars, immediately frozen, and kept at -25°C until analyzed.

Chemical analyses of organochlorine pesticides were performed according to a previously detailed method (Waluszewski and Szymczynski 1991; Waluszewski et al. 2004c). Gas chromatographic determinations of selected organochlorine pesticides (HCB, α -, β -, γ -HCH, *pp'*DDE, *op'*DDT and *pp'*DDT) were conducted with a Varian model 3400CX gas chromatograph equipped with a ^{63}Ni electron-capture detector. The operating conditions were as follows: separations were done on capillary column J&W 30-m \times 0.32-mm inner diameter and 0.83 μm film thickness, the temperature program was 193°C (7 min) to 250°C at $6^{\circ}\text{C}/\text{min}$, hold for 20 min; the carrier gas was nitrogen at 6.3 mL/min and a 1- μL aliquot was injected in a splitless mode.

The minimum detection limits for the residues analyzed were as follows: 0.1 $\mu\text{g/L}$ on wet weight or 0.2 mg/kg on fat basis for β -HCH, *pp'*DDE, *pp'*DDT and *op'*DDT. To determine the quality of the method, a recovery study was performed on 10 spiked replicates of blank cow blood samples, which presented contamination levels below the detection limits. The fortification study, done at 1.0 $\mu\text{g/L}$ and 2.0 mg/kg on fat basis levels, showed mean values from 83% to 91% recovery. The standard deviation and coefficient of variation were below 10, indicating excellent repeatability of the method.

Total serum lipids were determined calorimetrically with phosphovanillin according to the method recommended by Hycel de Mexico using a commercial kit for clinical laboratories.

Statistical calculations were conducted using statistical software Minitab version 12. Concentrations of organochlorine pesticide (mg/kg on fat base and $\mu\text{g/L}$ on wet weight) were expressed as frequencies, arithmetic means, medians, geometric means (GM) and 95% confidence intervals (95% CI) of geometrical means. The resulting concentrations were then used to determine the significance of categorical factors on pesticide levels by the variability among samples, pairing them to identify differences among means by applying the Student's *t* test and differences among medians by applying the Mann-Whitney test at $\alpha = 0.05$.

Results and Discussion

Lipophilic organochlorine pesticides are considered to have the ability to transfer through the placental barrier. The equilibrium models for these compounds in the human

body during pregnancy describe the partitioning rate between maternal compartments and fetus tissues, especially umbilical cord blood (Waluszewski et al. 2000). Organochlorine pesticides reside principally in lipids and their solubility does not differ significantly among lipids in the human body (Longnecker et al. 1999; Waluszewski et al. 2004a). The concentration for mother and fetus depends on the lipid content in the compartment and liposolubility of the pesticide. Pregnancy involves the transfer of nutrients and contaminants from maternal tissues through the placenta to the developing fetus. Therefore, organochlorine pesticides would normally pass through the placental barrier via passive diffusion. Posterior metabolism in the fetus of accumulated residues is low due to the poor detoxification mechanisms of the developing organs.

During the monitoring study, only the presence of β -HCH, pp' DDE and pp' DDT were detected, thus only these pesticides are discussed. The presence of pp' DDE was found in 100% of analyzed samples, whereas β -HCH and pp' DDT were present only in 4% of the samples. Summarized organochlorine pesticides levels from 70 monitored umbilical cord blood samples, expressed as ranges, mean and standard error of mean (SEM), median, geometric mean (GM) and 95% confidence intervals of geometric mean are presented in Table 1 as $\mu\text{g/L}$ on a wet basis and in Table 2 as mg/kg on a lipid basis. The highest levels are expressed by β -HCH, but its frequency is very low, corresponding only to 3 monitored samples. The same tendency can be observed for insecticide pp' DDT, which shows a presence only in 3 samples and should not influence the total panorama of exposure in Veracruz. The pesticide pp' DDE was found in 100% of the samples but the mean concentration values diminished substantively when expressed as median and geometric mean, indicating a predominance of lower levels in the monitored population.

In order to take into account the significance of new-born sex as a categorical factor on pesticide levels, the total pool sample was divided according to the sex of new-born babies (Table 3). The grouping results were compared to

Table 1 Ranges, mean and standard error of mean (SEM), median, geometric mean (GM) and 95% of confidence intervals of GM (95% IC) of organochlorine pesticide levels ($\mu\text{g/L}$ on wet weight) in umbilical cord blood serum (n = 70)

Pesticide	Ranges	Mean \pm SEM	Median	GM	95% CI
β -HCH	3.7–8.7	5.5 \pm 1.6	3.9	5.1	1.5, 6.5
pp' DDE	0.1–13.5	1.3 \pm 0.2	0.7	0.7	0.6, 0.9
pp' DDT	0.1–1.8	1.1 \pm 0.5	1.4	0.6	0.1, 2.8
Σ -DDT	0.1–14.9	1.3 \pm 0.3	0.7	0.7	0.6, 0.9

Table 2 Ranges, mean and standard error of mean (SEM), median, geometric mean (GM) and 95% of confidence intervals of GM (95% IC) of organochlorine pesticide levels (mg/kg on lipid basis) in umbilical cord blood serum (n = 70)

Pesticide	Ranges	Mean \pm SEM	Median	GM	95% CI
β -HCH	21.9–38.7	28.0 \pm 5.4	23.3	27.1	12.4, 58.6
pp' DDE	0.6–102.4	6.9 \pm 1.6	3.1	3.5	2.7, 4.5
pp' DDT	0.2–10.2	5.9 \pm 2.9	7.4	2.4	0.1, 6.4
Σ -DDT	0.6–112.6	7.2 \pm 1.8	3.1	3.5	2.0, 4.5

Table 3 Mean and standard error of mean (SEM), median, geometric mean (GM) and 95% of confidence intervals (95% CI) of organochlorine pesticide levels ($\mu\text{g/L}$ on wet weight) in umbilical cord blood serum according to sex of new born babies

Feminine (n = 32)					
Pesticide	Ranges	Mean \pm SEM	Median	GM	95% CI
β -HCH	3.9	3.9	3.9	3.9	
pp' DDE	0.1–13.5	1.4 \pm 0.4	0.8	0.8	0.2, 1.0
pp' DDT	1.4	1.3	1.3	1.4	
Σ -DDT	0.1–14.8	1.4 \pm 0.4	0.8	0.8	0.5, 0.3
Masculine (n = 38)					
Pesticide	Ranges	Mean \pm SEM	Median	GM	95% CI
β -HCH	3.7–8.7	6.3 \pm 2.5	6.3	5.7	
pp' DDE	0.1–8.9	1.2 \pm 0.7	0.6	0.7	0.5, 0.9
pp' DDT	0.1–1.8	0.9 \pm 0.8	0.9	0.4	
Σ -DDT	0.1–10.7	1.2 \pm 0.3	0.6	0.7	0.6, 0.9

determine differences in organochlorine pesticide concentrations. The applied *t* test and Mann–Whitney tests showed that there were no statistically significant differences among means and medians at $p > 0.05$, suggesting semi-homogenous exposure, and eliminating new-born gender as a discriminating factor. However, one case of a twin pregnancy resulting in babies of different sex (feminine and masculine) revealed significant differences in organochlorine pesticide concentrations: feminine: total lipid content 87 mg/L, pp' DDE 29.7 mg/kg, 2.6 $\mu\text{g/L}$; masculine: total lipid content 601 mg/L, pp' DDE 5.1 mg/kg, 3.1 $\mu\text{g/L}$ and pp' DDT 0.2 mg/kg, 0.20 $\mu\text{g/L}$. The obtained differences express that the feminine fetus presented in umbilical cord blood serum 6.9 times lower total lipid content and 5.7 times higher pp' DDE concentration expressed on fat basis and 1.2 time lower expressed on wet basis than the male fetus. Moreover, the masculine fetus had a presence of pp' DDT in the umbilical cord blood serum which was not detected in the serum of its feminine twin. These results elucidate independence in circulatory nutrition between twins and different passes of nutrients and organochlorine pesticides through the placental circulatory system during twin pregnancy.

Table 4 Median concentrations of organochlorine pesticides ($\mu\text{g/L}$ on wet weight) in cord blood serum according to parities: 1, 2, 3 and more

Pesticide	One (n = 35)	Two (n = 20)	Three and more (n = 15)
β -HCH	3.8	8.8	3.9
pp' DDE	0.6	0.9	0.7
pp' DDT	1.8	0.7	n.d.
Σ DDT	0.6	0.9	0.7

Table 5 Mean concentrations of organochlorine pesticides ($\mu\text{g/L}$ on wet weight) in cord blood serum in three tertiles according to age of mothers

Terciles (years)	β -HCH	pp' DDE	pp' DDT
First	20	3.7	0.4
Second	26	3.9	0.7
Third	29	8.7	1.3

Another determinant factor that can influence contamination level is parities. For this purpose, the total sample pool was divided according to the mother's number of deliveries, and the results are presented in Table 4 as median organochlorine pesticide concentrations. The significance of observed differences among β -HCH and pp' DDT median concentrations and three delivery groups cannot be calculated because of the small number of cases. The increase of pp' DDE median concentrations from the one to two childbirth group and decrease in the three or more childbirth group is affirmed statistically with significance at $p < 0.05$.

In order to consider the age of the mother as a discriminate contamination factor, the studied sample pool was divided according to age (years) of parturient, calculating the first (up to 20 years), second (between 20 and 26 years) and third (29 or more years) tertile (Table 5). For all organochlorine pesticides studied the mean concentrations increased significantly with age from first to second and to third tertile. Applying the *t* test for comparison of means, statistically significant ($p < 0.05$) differences for pp' DDE concentrations among three tertiles were obtained.

In conclusion, the present monitoring study indicates the passage of organochlorine pesticides through the placental barrier, confirming their presence in umbilical cord blood serum. The concentrations were not dependant on the sex of the new-born or the number of deliveries. The level of organochlorine pesticides that circulates through placental barriers depends on the age of pregnant women, with higher age favoring higher organochlorine pesticide concentrations caused by more years of exposure to the environmental contaminants.

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