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Organochlorine Pesticide Levels in Female Adipose Tissue from Puebla, Mexico

Stefan M. Waliszewski · K. Sanchez · M. Caba · H. Saldariaga-Noreña · E. Meza · R. Zepeda · R. Valencia Quintana · R. Infanzon

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Abstract The objective of this study was to determine the levels of organochlorine pesticides HCB, α-β-γ-HCH, *pp*′DDE, *op*′DDT and *pp*′DDT in adipose tissue of females living in Puebla, Mexico. Organochlorine pesticides were analyzed in 75 abdominal adipose tissue samples taken during 2010 by autopsy at the Forensic Services of Puebla. The results were expressed as mg/kg on fat basis. In analyzed samples the following pesticides were detected: *p,p'*-DDE in 100% of samples at mean 1.464 mg/kg; *p,p'*-DDT in 96.0.% of samples at mean 0.105 mg/kg; *op*′DDT in 89.3% of monitored samples at mean 0.025 mg/kg and *β*-HCH in 94.7% of the samples at mean 0.108 mg/kg. To show if organochlorine pesticide levels in monitored female's adipose tissues are age dependant, the group was divided in three ages ranges (13–26, 26–57 and 57–96 years). The mean and median levels of all organochlorine pesticides increase significantly ($p < 0.05$) from the first to second and from the first to third group. At the same time, the increase of mean and medians levels from the second to third group were not statistically significant ($p > 0.05$). The present results compared to previous ones from 2008 indicates an increase in the concentrations during the 2010 study, but only the differences for *pp*'DDE and *op*'DDT were statistically significant. The 2010 group of females was older compared to the 2008 group. The presence of organochlorine pesticide residues is still observed, indicating uniform and permanent exposure to the pesticides by Puebla inhabitants.

Keywords Organochlorine pesticides · Adipose tissue · Female

S. M. Waliszewski (✉) · R. Infanzon
Institute of Forensic Medicine, University of Veracruz,
SS Juan Pablo II s/n, 91980 Boca del Río, Mexico
e-mail: swal@uv.mx

K. Sanchez
Forensic Medicine Services, Puebla, Mexico

M. Caba · E. Meza · R. Zepeda
Biomedical Research Center, University of Veracruz,
Xalapa, Mexico

H. Saldariaga-Noreña
Faculty of Chemical Sciences,
University Autonomous of Coahuila, Saltillo, Mexico

R. Valencia Quintana
University Autonomous of Tlaxcala, Tlaxcala, Mexico

The exposure to organochlorine pesticide residues and the consequential health risks remain an important concern for public health. Due to their chemical stability, lipophilic nature, and tendency to bioconcentrate, the organochlorine pesticides remain ubiquitous contaminants in food, air and the human body. They have a lipid affinity that causes them to enter the circulatory system where they are transported via the lipid component in plasma and deposited in lipids of adipose tissue. The adipose tissue organochlorine pesticide levels are commonly used as biomarkers of exposure in epidemiologic studies comparing the levels with health status and pathological effects (Eskenazi et al. 2009; Ferre-Huguet et al. 2009).

The uses of DDT and HCH throughout the decades provided great benefits, especially in tropical countries combating the spread of disease-transmitting organisms. It has also lead to investigations of the magnitude of their residues in all elements of the human environment. These pesticides with low or intermediate volatility, when they were sprayed, were deposited upon superficial soils, from which they would volatilize and tend to distribute uniformly throughout the environment (Alegria et al. 2008;

Wong et al. 2008; Santiago and Cayetano 2011; Ritter et al. 2011).

Humans are part of the environment and are contaminated with the same chemicals which are present in surrounding air, water, food, consumer articles, etc. The lipid-rich tissues of the human body accumulate lipophilic environmental pollutants by virtue of physiological cellular component interactions, identifying them as bioindicators of body burden. Thus, the determination of levels in human tissues, such as adipose or blood serum, can reflect the magnitude of local environment pollution (Waliszewski et al. 2010).

Environmental exposure to persistent organochlorines can provide an internal dose to females, as these compounds have been found in human adipose tissue, maternal blood serum and umbilical cord serum. This is evidence of exposure to critical tissues during important windows of development (Meeker et al. 2009; Herrero-Mercado et al. 2011). Conventional wisdom regarding environmental exposures to persistent pesticides presumes declines in concentrations over time and that the origin of those chemicals lies in a person's cumulative exposure over a lifetime.

Many persistent compounds can still be detected in samples of the human body decades after they are banned. For example, DDT was banned in Mexico in 1999 but its breakdown product *pp*′DDE and insecticide *pp*′DDT are still detected in human samples (Waliszewski et al. 2011). This continuing occurrence may result at least in part from inhaled vapors, originated from superficial soils (Martínez-Salinas et al. 2011; Ritter et al. 2011) and consumption of foods that contain organochlorine residues (Borchers et al. 2010). With the new data, researchers can better understand how and why organochlorine pesticide levels vary within certain populations. For example, lower total levels exist among Mexicans living in different geographic places in Mexico (Waliszewski et al. 2010) and when compared within sexes, levels were generally higher in females than in males.

Our previous study (Waliszewski et al. 2010), that monitored human populations indicated higher mean β-HCH, *pp*′DDE, *op*′DDT and total DDT levels in females compared to males from Puebla and levels equal to those in the Turkey population (Daglioglu et al. 2010). This data prompts us to monitor female Puebla inhabitants to determine the organochlorine pesticide levels tendency among the population and to compare the levels between the studies of 2010 and 2008.

Materials and Methods

Human adipose samples (approximately 5 g) from abdominal cavities of 75 randomly selected females were taken during 2010 by autopsy at the Forensic Medicine Services of Puebla. The donors originated from Puebla City and from the state of Puebla. The samples were stored in glass jars which were previously washed with sulfuric acid, immediately frozen and kept at −25°C until analyzed. The dated samples were labeled with donor data.

The analyses of organochlorine pesticide residues in human adipose tissue were performed by gas chromatography with ECD according to a method previously described (Waliszewski et al. 2004c; 2010). All of the samples were analyzed for: HCB, α, β, γ-HCH, *pp*′DDT, *op*′DDT, and *pp*′DDE. The minimum detection limits for the residues analyzed were as follows: 0.001 mg/kg for HCB, 0.002 mg/kg for the HCH isomers and *pp*′DDE, and 0.003 mg/kg for *pp*′DDT and *op*′DDT. To determine the quality of the method, a recovery study was performed on 10 spiked replicates of blank cow fat samples, which presented contamination levels below the detection limits. The fortification study, done at 0.01–0.03 mg/kg levels, depending on the pesticide, showed mean values from 85% to 92% recovery. The standard deviation and coefficient of variation were below 10, indicating excellent repeatability of the method.

Statistical calculations were conducted using the statistical software Minitab version 12. Concentrations of organochlorine pesticide (mg/kg on fat base) were expressed as frequencies, arithmetic means, medians and geometric means (GM). The significance of categorical factors on pesticide levels was determined by using 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

The mean age of the monitored females from the Puebla population was 54.4 years (range 13–96), with a median of 57 years.

During the monitoring study, only the presence of β-HCH, *pp*′DDE, *op*′DDT and *pp*′DDT were detected, thus only these compounds are discussed. Table 1 summarizes results of organochlorine pesticides from 75 adipose tissue samples expressed as frequencies, ranges, mean ± standard deviation of mean (SD), median and geometric mean (GM) levels. All concentrations are expressed on lipid base (mg/kg). *pp*′DDE was found in 100% of the samples, whereas *pp*′DDT, β-HCH and *op*′DDT were presented in 96.0%, 94.6% and 90.6% of the samples, respectively.

pp′DDE was found at the highest mean concentration of 1.464 mg/kg on lipid base. This metabolite of insecticide *pp*′DDT is followed by the isomer of β-HCH of

Table 1 Organochlorine pesticide levels (mg/kg on fat basis) in female adipose tissue from Puebla inhabitants (n = 75)

Pesticide	Frequency	Ranges	Mean ± SD	Median	GM
β-HCH	71/75	0.002–0.797	0.108 ± 0.171	0.041	0.041
pp'DDE	75/75	0.023–4.619	1.464 ± 1.345	1.111	0.704
op'DDT	67/75	0.002–0.076	0.025 ± 0.017	0.019	0.019
pp'DDT	72/75	0.016–1.130	0.105 ± 0.136	0.080	0.075
Σ-DDT		0.023–4.941	1.590 ± 1.411	1.189	0.861
pp'DDE/pp'DDT		0.59–59.10	16.70 ± 14.27	14.50	10.46
Age		13–96	54.5 ± 24.0	57.0	48.1

lindane (γ-HCH) insecticide that had a mean concentration of 0.108 mg/kg and by the insecticide pp'DDT at 0.105 mg/kg. The results from op'DDT indicated a lower mean concentration of 0.025 mg/kg. A comparison of mean, median and geometric mean concentrations of organochlorine pesticides detected in analyzed samples, shows a decrease of values from mean to median and to geometric mean, which points out a prevalence of lower concentrations among the total samples and the existence of occasional cases of extreme exposure which are indicated by the range values. The calculated pp'DDE/pp'DDT ratios, that evaluate the age of exposure, evidenced a mean value of 16.70 that decreases when expressed as a median and GM. The ratio indicates antique exposures and the predominance of pp'DDE, a metabolite of insecticide pp'DDT, as the principal contaminant of Puebla females. The data presented in Table 1 explain that in Puebla there still exist contamination sources with DDT, promoting accumulation of the compounds in the human body by inhalation of contaminated air and by consumption of contaminated foods containing the residues. Among the females 13.3% (10/75) contained higher than 0.2 mg/kg β-HCH levels, which ranged 0.207–0.797 mg/kg, 32% (24/75) of pp'DDE levels were higher than 2.0 mg/kg, which ranged 2.001–4.619 mg/kg and 6.6% (5/75) of pp'DDT levels were higher than 0.2 mg/kg, which ranged 0.202–1.130 mg/kg.

Table 2 compares the present monitoring results with the previous ones from 2008 (Waliszewski et al. 2010) and the statistical comparisons to observe possible differences among means and medians to evaluate the possible trend in their levels. The β-HCH increases in the concentrations during the 2010 study, but the differences between means and medians are not statistically significant ($p > 0.05$). pp'DDE concentration levels in the 2010 female population increase significantly, with differences that are statistically significant between means ($p = 0.049$) and medians ($p = 0.037$). op'DDT reveals the same tendency, an increasing level, and the populations are shown to be different. The concentration of insecticide pp'DDT also increases from 2008 to 2010, but differences between means and medians are not statistically significant ($p > 0.05$). Total DDT increases, caused by an increase in pp'DDE, revealed statistically significant differences between means and medians ($p < 0.05$). Calculated pp'DDE/pp'DDT ratios, which indicate the age of exposure, show a decrease in mean value from 30.9 to 16.7 and increase of median from 7.6 to 14.5 indicating that both populations are not homogenous. Evaluating ages of females studied revealed that these are statistically different for means and medians ($p < 0.05$), showing the 2010 female group as older compared to 2008.

To observe if age of females can have an influence on organochlorine pesticide levels, the 2010 population was

Table 2 Comparison of the organochlorine pesticide levels (mg/kg) in female adipose tissue between 2008 and 2010

Pesticide	Mean ± SD		Median	
	2008	2010	2008	2010
β-HCH	0.054 ± 0.098	0.108 ± 0.171	0.015	0.041
pp'DDE	0.865 ± 1.082*	1.464 ± 1.345*	0.205*	1.111*
op'DDT	0.013 ± 0.006*	0.025 ± 0.017*	0.015*	0.019*
pp'DDT	0.090 ± 0.147	0.105 ± 0.136	0.053	0.080
Σ-DDT	0.978 ± 1.091*	1.590 ± 1.411*	0.377*	1.189*
pp'DDE/pp'DDT	30.9 ± 87.3	16.70 ± 14.27	7.6	14.50
Age	35.6 ± 19.2*	54.5 ± 24.0*	30*	57.0*

* statistically significant differences ($p < 0.05$)

grouped ascendant according to age. The first half of the group was considered to make a statistical comparison with females from the 2008 study (Table 3). Applied *t* test and Mann–Whitney tests responded that no statistical differences ($p > 0.05$) exist between 2008 females and first half of the 2010 group. The results indicate that age is a principal factor that has an influence on organochlorine pesticide levels among populations which live in environmentally contaminated areas.

To further examine further whether or not organochlorine pesticide levels in monitored females adipose tissues can depend on age, the 2010 group was divided according to age into an ordered ascendant distribution of three parts, each containing a third of the population, and the frequencies, mean and median tertiles of pesticide levels were calculated (Table 4).

In general, all values of pesticide concentrations increase from first to second and to third tertile. This is in accordance with data reported for the Turkey (Daglioglu et al. 2010) and Veracruz (Waliszewski et al. 2011) populations. The mean and median levels of all organochlorine pesticides increase significantly ($p < 0.05$) from the first to second tertile and from the first to third tertile. Meanwhile, from the second to third tertile the increases in mean and median levels were not statistically significant ($p > 0.05$). The same tendency in increases is presented by the *pp*′DDE/*pp*′DDT ratio showing that the younger tertile is exposed to *pp*′DDT at comparable magnitudes to older females living in Puebla. The older tertile accumulates more organochlorine pesticide during the lifetime, especially *pp*′DDE, which increases the *pp*′DD/*pp*′DDT ratio. Evaluating differences among three tertiles of ages revealed that these were statistically different ($p < 0.05$) in means and medians.

The results which compare organochlorine pesticide levels between human adipose tissue of females from Puebla and Veracruz (Waliszewski et al. 2011) analyzed during 2010 year are presented in Table 5. Statistical tests applied to determine possible differences between mean and median concentrations of organochlorine pesticides from both populations showed that only medians of *op*′DDT and means of *pp*′DDT levels were statistically different ($p < 0.05$), while other organochlorine pesticide levels, including the *pp*′DDE/*pp*′DDT ratio, were not

Table 3 Comparison of organochlorine pesticide levels (mg/kg on fat basis) in female adipose tissue from Puebla inhabitants in 2008 to the first half of population from 2010 grouped ascendant according to age

Pesticide	Mean ± SD		Median	
	2008	2010	2008	2010
β-HCH	0.054 ± 0.099	0.047 ± 0.076	0.014	0.016
pp′DDE	0.865 ± 1.082	0.823 ± 1.140	0.205	0.317
op′DDT	0.013 ± 0.006	0.026 ± 0.019	0.015	0.020
pp′DDT	0.090 ± 0.147	0.075 ± 0.057	0.053	0.062
Σ-DDT	0.978 ± 1.091	0.914 ± 1.191	0.377	0.372
pp′DDE/*pp*′DDT	30.9 ± 87.3	10.15 ± 10.04	7.6	7.17
Age	35.6 ± 19.2	33.1 ± 12.6	30	33.0

* statistically significant differences ($p < 0.05$)

Table 4 Organochlorine pesticide levels (mg/kg on fat basis) in tertiles according to age for Puebla female inhabitants

Females from 2010									
Pesticide	First			Second			Third		
	Freq	X ± SD	Median	Freq	X ± SD	Median	Freq	X ± SD	Median
β-HCH	22/25	0.023 ± 0.047	0.011	24/25	0.099 ± 0.108	0.065	25/25	0.192 ± 0.241	0.104
pp′DDE	25/25	0.373 ± 0.569	0.194	25/25	1.838 ± 1.237	1.814	25/25	2.183 ± 1.342	2.059
op′DDT	20/25	0.027 ± 0.022	0.017	23/25	0.023 ± 0.013	0.020	24/25	0.024 ± 0.016	0.019
pp′DDT	23/25	0.055 ± 0.036	0.042	24/25	0.096 ± 0.057	0.089	25/25	0.158 ± 0.212	0.117
Σ-DDT		0.445 ± 0.602	0.283		1.956 ± 1.275	1.878		2.369 ± 1.419	2.210
DDE/*pp*′DDT		7.05 ± 6.94	4.13		22.88 ± 15.04	18.98		19.65 ± 14.33	16.31
Age		26.1 ± 7.9	26.0		56.4 ± 10.11	57.0		80.9 ± 6.2	79.0

Table 5 Comparison of the organochlorine pesticide levels (mg/kg) in female adipose tissue between Puebla 2010 and Veracruz 2010

Pesticide	Mean ± SD		Median	
	Puebla	Veracruz	Puebla	Veracruz
β-HCH	0.108 ± 0.171	0.070 ± 0.083	0.041	0.039
pp′DDE	1.464 ± 1.345	1.497 ± 1.489	1.111	1.105
op′DDT	0.025 ± 0.017	0.021 ± 0.022	0.019*	0.014*
pp′DDT	0.105 ± 0.136*	0.207 ± 0.419*	0.080	0.076
Σ-DDT	1.590 ± 1.411	1.723 ± 1.745	1.189	1.175
pp′DDE/pp′DDT	16.70 ± 14.27	13.51 ± 9.82	14.50	12.52
Age	54.5 ± 24.0*	44.6 ± 12.4*	57.0*	42.0*

* statistically significant differences ($p < 0.05$)

different. The ages of monitored females were statistically different ($p < 0.05$) for means and medians indicating Puebla females as older.

In conclusion, during the monitoring study of female adipose tissue from Puebla inhabitants, the presence of organochlorine pesticide residues are still observed. Differences in pesticide concentrations comparing 2008 and 2010 year noted that for *pp′*DDE and *op′*DDT significant increases in concentrations exist, whereas for insecticide *pp′*DDT the increase was not significant, indicating uniform and permanent exposure to the pesticide for Puebla's inhabitants. The higher concentrations of organochlorine pesticides correspond to older persons. These persons are environmentally and alimentary exposed to organochlorine pesticides that they accumulate in adipose tissue during a lifetime. Comparing Puebla to Veracruz females, only *pp′*DDT levels were significantly higher in Veracruz and no significant differences among other pesticides were determined. The comparison indicates a tendency of decreasing residue levels for these pesticides in Mexico. Comparing areas which include Veracruz, which is well-known for its higher past exposure caused by the intensive use of DDT in sanitary actions for vector combat, there is a gradual disappearance of these compounds from the environment since they were banned in 1999 in Mexico.

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