# **Endosulfan and Other Organochlorine Pesticide Residues** in Maternal and Cord Blood in North Indian Population

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**Abstract** Humans are exposed to various environmental chemicals such as organochlorine pesticide residues, heavy metals, polychlorinatedbiphenyls (PCBs) etc. There is paucity of data regarding the present blood levels of organochlorine residues in North Indian population with reference to reproductive health. The present study was designed to analyze the levels of organochlorine pesticide residues in maternal and cord blood samples of normal healthy women with full term pregnancy to gain insight into the current status of pesticide burden in newborns. Hexachlorocyclohexane (HCH) contributed maximum towards the total organochlorine residues present in maternal and cord blood followed by endosulfan, pp' DDE and pp' DDT being the least. This is also the first report indicating endosulfan levels in this population. Our data indicates a transfer rate of 60-70% of these pesticides from mothers to newborns and this high rate of transfer of pesticides is of great concern as it may adversely affect the growth and development of newborn.

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Pesticides are now all pervasive and have become an almost inescapable part of our environment. According to the pesticides industry statistics, India spends \$3/ha on pesticides compared with \$24/ha spent by Philippines, \$255/ha spent by South Korea and \$633/ha by Japan. Yet, despite a comparatively low use of pesticides in India, the contamination of food products is alarming. About 20% of Indian food products contain pesticide residues above tolerance level as compared to only 2% globally. Forty percent of all pesticides used in India belong to the organochlorine (OCP) class of chemicals (Teri Vision 2000).

Organochlorine Pesticides (OCPs) like endosulfan, hexachlorocyclohexane (HCH), 1, 1, 1-trichloro2, 2-bis (4-cholorophenyl) ethane (DDT) etc. are persistent environmental organic pollutants in nature.

The storage of these toxins 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 mobilizing internal status of pollutants many years after initial exposure. During pregnancy, concern over OCP residue levels in maternal and cord becomes greater since reproductive toxicological studies have revealed that mother and fetus are more vulnerable to their toxic action. Newborns are exposed through placental transmission as well as breastfeeding (Falcon et al. 2004). Hence knowledge of biological levels of these contaminants in human tissues is necessary for the risk assessment of adverse health effects and for the identification of vulnerable groups.

In view of the restriction imposed on the use of DDT and lindane ( $\gamma$ -HCH) in recent years in India (Bhatnager

et al. 2004) and paucity of data regarding the present blood levels of OCP residues in north Indian population, the present study was designed to analyze the levels of OCP residues in maternal and cord blood samples of normal healthy women with full term pregnancy to gain insight into the current status of pesticide burden in neonates.

#### **Materials and Methods**

Maternal and cord blood samples were collected from pregnant women attending the Gynecology ward/labor room of Guru Teg Bahadur Hospital, Dilshad Garden, Delhi during the period January 2006 to September 2007. Women, having acute and chronic medical disorders (hypertension, diabetes, chronic renal disease, chronic liver disease etc.) were excluded during sample collection. An informed consent and questionnaire form having the information about age, employment, socioeconomic status, drinking water source, alcohol consumption, dietary and smoking habits, were filled prior to collection of the sample.

Blood samples (1 mL each) were collected in EDTA vacutainer tubes at the time of delivery and stored at -20°C until analysis. OCP residues extraction was done by using hexane and acetone (2:1) according the method of Bush et al (1984). Cleanup was done by USEPA method using Florisil (Sigma) by column chromatography. Quantification of organochlorine residue levels was done by Perkin Elmer Gas Chromatograph equipped with <sup>63</sup>Ni selective electron capture detector. Limits of detection were <0.05 pg perchloroethylene with nitrogen. Quantitative analysis of OCP residues in each sample was done by comparing the peak heights with those obtained from a chromatogram of a mixed organochlorines standard of known concentration. Ten samples of each maternal and cord blood in triplicate were spiked with a mixed standard of organochlorine pesticides, respectively 5 and 25 ng/mL. The average recoveries of fortified samples were exceeding 95%. Further, a quality check sample was always run with each set of samples for pesticide analysis to maintain accuracy.

## **Results and Discussion**

Humans, being at the top of the food chain are most vulnerable to health effects as ingestion of toxic chemicals is several folds higher through the process of biomagnifications. Potential hazards due to pesticide exposure are particularly higher in the developing countries because of the poor regulatory mechanisms, improper healthcare and low literacy rate. OCPs remain in the human system for many years due to their lipophilic nature and long half-life.

OCPs are supposed to be 'xenoestrogens' and can interfere with normal functioning of the endocrine system thereby increasing risk of various disorders in reproduction, cancer etc. (Torres et al. 2006).

The present study reports the result of 68 maternal and cord blood samples from mothers undergoing normal full term delivery. The mean age of the mothers was  $25.3 \pm 1.92$  years, the mean gestational age was  $37.4 \pm 0.41$  weeks and the mean weight of newborns was  $2.81 \pm 0.27$  kg (Table 1). HCH contributed maximum towards the total OCP residues present in maternal and cord blood followed by endosulfan, pp' DDE and pp' DDT contributing the least (Tables 2 and 3).

Technical-grade endosulfan is composed of two stereochemical isomers,  $\alpha$ -endosulfan and  $\beta$ -endosulfan, in concentrations of approximately 70% and 30%, respectively. US data from 1987 to 1997 indicate an average domestic use of approximately 1.38 million pounds of active ingredient per year (U.S. Environmental Protection Agency 2002). Endosulfan is being manufactured in India since 1996 and is now extensively used for gardening, wood preservation and against a variety of agricultural pests. During 1999–2000, about 81,000 metric tons of endosulfan was manufactured in India, and in terms of tonnage its production was next only to mancozeb (103,000 metric tons) and monocrotophos (95,000 metrictons)

Table 1 Demographical profile of Subjects

Characteristics	Mean $\pm$ SD
Maternal age (years)	$25.3 \pm 1.92$
Maternal weight (Kg)	$50.3 \pm 2.69$
Gestational age (weeks)	$37.4 \pm 0.41$
Newborn weight (Kg)	$2.81 \pm 0.27$
±SD is the standard deviation	
Education	
Illiterate	6
Intermediate $(10 + 2)$ Pass or less	43
Graduate	19
Drinking water supply	
Government source	49
Private source	19
Smoking habits	
Non-Smoker	65
Tobacco chewer	3
Living style	
Slum	24
Market area	27
Colony	17
Dietary habits	
Vegetarian	41
Non-vegetarian	27



Table 2 Organochlorine pesticide levels in maternal blood (ng/mL)

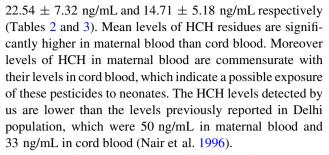
Pesticides	Minimum	Maximum	Mean ± SD
α-НСН	1.91	19.77	$7.26 \pm 4.08$
$\beta$ -HCH	1.39	25.39	$10.05 \pm 7.01$
γ-НСН	0.98	21.62	$5.23 \pm 3.11$
Total HCH	0.98	25.39	$22.54 \pm 7.32$
α-Endosulfan	0.00	6.90	$2.21 \pm 2.04$
$\beta$ -Endosulfan	0.00	6.89	$1.49 \pm 2.10$
Total Endosulfan	0.00	6.90	$3.70 \pm 4.20$
pp' DDE	0.00	6.24	$4.26 \pm 3.66$
pp' DDT	0.00	4.06	$1.46 \pm 2.99$

Table 3 Organochlorine pesticide levels in cord blood (ng/mL)

Pesticides	Minimum	Maximum	Mean ± SD
α-НСН	1.43	13.71	$4.71 \pm 3.12$
$\beta$ -HCH	2.1	22.03	$7.23 \pm 4.24$
γ-НСН	0.91	12.82	$2.77 \pm 2.41$
Total HCH	0.91	22.03	$14.71 \pm 5.18$
α-Endosulfan	0.00	5.79	$1.39 \pm 1.42$
$\beta$ -Endosulfan	0.00	3.46	$0.88 \pm 1.27$
Total endosulfan	0.00	5.79	$2.27 \pm 2.44$
pp' DDE	0.00	4.09	$3.08 \pm 2.72$
pp' DDT	0.00	2.02	$1.03 \pm 2.63$

(Habibullah et al. 2003). We have detected endosulfan in 41 samples out of 68 and the mean values were  $3.70 \pm 4.20$  ng/mL and  $2.27 \pm 2.44$  ng/mL in maternal and cord blood respectively (Tables 2 and 3). The transplacental transfer of various pesticides from mothers to infants has been reported previously, but information regarding the in-utero transfer of endosulfan from mothers to infants in Indian population is scarce. Mathur et al (2005) have reported 4.6 ng/mL of endosulfan in agricultural population in Punjab villages. The higher values found by these authors is possibly a result of occupational exposure of organochlorines among cultivators and sprayers. Presence of endosulfan in subjects of the present study is an important observation. To the best of our knowledge, no other report is available in this regard. Endosulfan levels reported from elsewhere are extremely contradictory. Torres et al (2006) have reported endosulfan II levels as high as  $76.38 \pm 52.60$  ng/mL in pregnant women giving birth via cesarean section in southern Spain. On the other hand, a study conducted in Rio de Janeiro, Brazil (Sarcinelli et al. 2003) have reported extremely low levels, ie.  $0.108 \pm 0.043$  ng/mL and  $0.214 \pm 0.101$  ng/mL endosulfan in mothers and newborns respectively.

Among the OCP residues, HCH isomers were detected in 100% of maternal and cord blood samples. The mean levels of total HCH in maternal and cord blood were



In an earlier report the level of total HCH in blood was found to be  $49.00 \pm 5.00$  ng/mL (Ramachandran et al. 1984) which were considerably higher than the present observed value. The low values obtained by us now may be a result of discontinuation of HCH use in public health programs in India since April 1997. Siddique et al (2003) have also reported lower value of total HCH (14.31  $\pm$  9.83 ng/ mL in maternal and 6.74  $\pm$  7.83 ng/mL in cord blood) in women with normal birth weight babies. Similarly low HCH levels were reported in blood samples from northern India (Mathur et al. 2005) and western India (Bhatnagar et al. 2004), however these levels were higher than those, in the present report. All the three isomers of HCH were found in the blood samples and among these  $\beta$ - HCH levels were highest. Lindane (γ-HCH) continues to be manufactured in India and is used for crop protection; possibly  $\alpha$ - and  $\beta$ -HCH are present with lindane as contaminants during the process of manufacturing. The reason for the observed levels of total HCH being comparatively lower than the earlier reports in India may be due to the restriction imposed on the use of these pesticides in agriculture. Certain studies originating from other countries, however have reported considerably lower levels of HCH (Sala et al. 2001; Sarcinelli et al. 2003; Torres et al. 2006).

We have also detected pp' DDT in 36 samples and its metabolite pp' DDE in 52 samples of both maternal and cord blood. The sum of pp' DDT and pp' DDE levels were  $5.72 \pm 4.97$  ng/mL in maternal and  $4.11 \pm 4.26$  ng/mL in cord blood (Tables 2 and 3). Blood DDT levels obtained by us were found to be slightly lower than reported earlier by other workers in India (Bhatnagar et al. 2004; Mathur et al. 2005). This is possibly due to very low use of DDT in public health programs. We have detected very low levels of aldrin and dieldrin in three samples only (data not shown). Our data indicates a transfer rate of 60-70% of these pesticides from mothers to newborns and this such a high rate of transfer of pesticides is of great concern as it may adversely affect the growth and development of newborn. Chlorinated pesticides are persistent organic pollutants, which have long half-lives and tendency to accumulate in fatty tissue. OCPs exposure is related to various adverse health effects like neurotoxicity, reproductive toxicity, immonutoxicity, carcinogenicity, oxidative stress etc. (Banerjee et al. 1996, 1997, 2001; Sarcinelli et al. 2003; Torres et al. 2006).



In conclusion, the results of the present study clearly demonstrate a prenatal uptake of organochlorine compounds and provide more information on the transplacental transfer of endosulfan in Indian population. Although, in this study we have estimated the levels in healthy women who gave birth to normal weight babies, it has been reported that the body load of these pesticides may lead to adverse health effects in the infants. Prenatal exposure to these compounds may play a more important adverse role in neurodevelopment in infants than postnatal exposure. Hence, follow up studies in babies born to mothers with high blood pesticide levels need to be undertaken in the present scenario.

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