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# Pesticide residues in human breast milk: Risk assessment for infants from Punjab, India



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#### HIGHLIGHTS

- Temporal trend for occurrence of HCH and DDT residues in human breast milk indicated significant reduction.
- Contamination of breast milk with chlorpyrifos, cypermethrin and endosulfan residues observed first time in this region.
- · Estimated daily dietary intake of DDT by few infants still exceeded FAO/WHO recommended provisional tolerable daily intakes.

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#### ABSTRACT

Punjab state in India is an agrarian society, where agriculture is the lifeline of farming community. To keep pace with increasing demands of food for growing population the indiscriminate use of pesticides has led to the contamination of environment and food commodities in this region.

Analysis of human breast milk samples (n = 53) for pesticide residues revealed the presence of  $\beta$ -,  $\gamma$ -HCH, p,p' DDD, p,p' DDD, p,p' DDT and endrin with mean concentration of 97.9, 101.7, 239.8, 1574.1, 100.3 and 90.7 ng g<sup>-1</sup> lipid wt., respectively. In addition, occurrence of  $\beta$ -endosulfan, endosulfan sulphate, cypermethrin and chlorpyrifos in this study have also been reported for the first time in human breast milk in Punjab, India. With increase in parity, HCH and DDT residue burden in donor's milk decreased. Although levels of HCH and DDT residues in breast milk samples have decreased significantly, yet estimated daily intake values for DDT are higher than the FAO/WHO permissible tolerable daily intake values for few infants.

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#### 1. Introduction

There is no doubt that organochlorine pesticides (OCPs) have played a pivotal role in increasing agricultural output and control of vector borne diseases in India (Bhatnagar, 2001). However, their widespread use in the last three decades has led to ubiquitous persistence in both the biotic and abiotic components of the environment (Gupta, 2004). The bioaccumulation and magnification potential of OCPs in food chain has become a global issue with serious outcomes on wildlife and human health (Tanabe, 2002). Prolonged exposure to OCPs can affect the liver & kidney functions (Peres et al., 2006), disruption of endocrine system (Colborn et al., 1993), mental & psychomotor development (Sagiv et al., 2008), neurological & immune system disorders (Karmaus et al., 2003), risk of breast, lung, cervix & prostate cancer (Ahmed et al., 2002) and endometriosis, hypospadias & cryptorchidias (Wolf et al.,

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2000). The pre & post-natal exposure of infants to pesticides can result in impairment of intellectual function (Eskenazi et al., 2004) and delayed effects on central nervous system functioning (Ribas-Fitó et al., 2006).

India was one of the foremost producer and consumer of OCPs particularly dichlorordiphenyl trichloroethane (DDT) and hexachlorocyclohexane (HCH), till the ban/restriction on their use in late 1990s (Kannan et al., 1997). Still, a substantial amount of these chemicals are being permitted for malaria control program and agriculture (UNIDO, 2006). In India organophosphate (OP) and synthetic pyrethroid (SP) pesticides are swiftly replacing OCPs in agriculture (Kumari et al., 2005). Although reports on contamination of food articles with OCPs, OPs and SPs are there in the region covered in present study (Bedi et al., 2004; Cheema et al., 2004; Kumari et al., 2002), but there is paucity of data in human breast milk. Additionally, earlier studies in India on pesticide residue detection in human breast milk were confined to OCPs only (Kalra et al., 1994; Subramanain et al., 2007; Mishra and Sharma, 2011). As humans are at the top of food chain, the exposure to pesticides through dietary intake and environment is a matter of great concern. Human breast milk is considered a suitable medium for investigating pesticide residues exposure to

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population and breastfed infants (WHO, 2009). Therefore, present study was contemplated to estimate the status of OCP, OP and SP residues in human breast milk in Punjab which is amongst the highest user of pesticides in India (Tiwana et al., 2009).

#### 2. Materials and methods

#### 2.1. Collection of human breast milk samples

Fifty three human breast milk samples were collected from women admitted to maternity ward in Dayanand Medical College and Hospital for child delivery in months of November & December of 2011. Detailed information from participants in relation to their residential location (Fig. 1), age, parity, dietary habits (vegetarian/ non-vegetarian), residential status (rural/urban), height, weight, number of children, history of any reproductive problem and complications during the current pregnancy was registered on the questionnaire. In addition, infant's general body conditions and illness if any were also observed and recorded. Consent was obtained on proforma duly signed by sample donors. The study protocol for the present research work was approved by the institutional medical ethics committee. Breast milk samples were collected in acetone washed glass vials in presence of pediatrician, special attention was paid to avoid cross contamination. Samples were transported to laboratory under chilled conditions (4  $^{\circ}$ C) and stored at -20  $^{\circ}$ C until analysis.

#### 2.2. Extraction of pesticide residues

Collected milk samples measuring more than 6 ml were analyzed for fat percentage by automatic milk analyzer Lactoscan LA (Milkotronic LTD. Bulgaria) for expression of pesticide residues in breast milk samples on lipid basis. Calibration of milk analyzer was done with bovine milk samples of known fat percentages. However, mean lipid content value was used for estimating pesticide residues in those milk samples which could not be collected in sufficient volume. The pesticide residues from breast milk samples were extracted by method of Battu et al. (2004) with slight modifications. Five ml of milk sample with 20 g each of activated silica gel and anhydrous sodium sulfate was thoroughly mixed and packed into an extraction glass column containing 40 ml of dichloromethane. After 90 min, solvent in column was eluted drop wise followed by further elution with 150 ml of dichloromethane:acetone (1:2 v/v) mixture. The extracted sample was concentrated using rotary evaporator at 40 °C to dryness and final reconstitution was made in n-hexane: acetone (1:1) mixture.

#### 2.3. Estimation of pesticide residues

Estimation of pesticide residues was done using Gas chromatograph (GC) equipped with electron capture detector and flame thermionic detector (Shimadzu model 2010). The GC oven temperature for electron capture detector was initially programed at a temperature of 170 °C for 13 min followed by temperature increase to

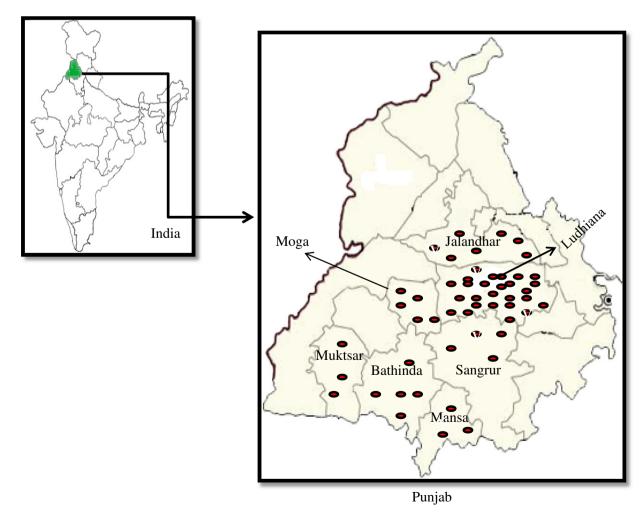


Fig. 1. Punjab map indicating the residential location of breast milk sample donors.

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