# Determination of Chlorinated Pesticides in Breast Milk of Saudi Lactating Mothers

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ABSTRACT. Environmental pollutants which may occur in breast milk and in various food products and drinking water, and which are also transferred to the fetus, constitute a severe threat to the health of infants and children. Among such compounds, various organochlorines, such as pesticides p, p' DDE, o, p DDT, p, p' DDT, methoxycholr and kepone for the control of parasites. The aim of this study was to evaluate the level of organochlorine pesticides in breast milk of Saudi lactating mothers (Jeddah). Breast milk samples, randomly collected from 13 Saudi donors were analyzed for chlorinated pesticides, using SFE and gas chromatography connected to electron capture detector (ECD). DDE was found in four samples only and ranged from 0.01-0.02 ppm, and DDT was found in one sample and contains 0.043 ppm.

Residue level of DDE and DDT found in the milk samples were generally low compared to the levels reported from Riyadh and Eastern regions in the Kingdom and also in other Middle Eastern Countries.

KEYWORDS: Breast milk, organochlorine, DDE, DDT.

#### Introduction

Breast milk contains all of the nutrients needed by the newborn baby during the first weeks of life. These include the metabolic fuels (fat, protein, and carbohydrate), water, and the raw materials for tissue growth and development, such as fatty acids, amino acids, minerals, vitamins, and trance elements (Jensen, 1989).

This dynamic fluid provides a diverse array of bioactive substances to the developing infant during critical periods of brain, immune and gut development. It is essential that the clinician should be familiar with how human milk is manufactured by the mammary gland, and the properties of human milk that render is nourishing and protective of the nursing infant (Carol, 2001).

Human milk composition has a dynamic nature and varies with time, such as at postpartum, during a nursing, and with the mother's diet and certain diseases. The changes of human milk composition with time of lactation seem to match the changing needs of the growing infant over time (Kunz *et al.*, 1999).

Breast-feeding is common in developing countries, but exclusive breast-feeding is rare, and complimentary foods are introduced at an early age. Thus, poorer nutritional status was significantly associated with earlier complementary feeding (Adetugbo and Adetugbo, 1997).

Several studies performed in high babies have demonstrated a significant reduction in the prevalence and severity of topic diseases with dietary and environmental manipulations. It has been demonstrated that prolonged breastfeeding and the avoidance of cow's milk, eggs, and fish during the first three months of lactation significantly decrease both the prevalence and the severity of a topic disease prior to the age of 5 years (Businco *et al.*, 1993).

Environmental pollutants which may occur in breast milk and in various food products and drinking water, and which are also transferred to the fetus, constitute a severe threat to the health of infants and children. Among such compounds, various organochlorine, such as pesticides for the control of parasites (DDTs, HCHs) (Zetterstrom, 1999).

Among these pesticides is chlorine containing compounds including DDT, aldrin, dieldrin and lindane. The organochlorine acts through disruption of neurotransmission. PCB's, which are not used as pesticides, are also organochlorine with similar human action, and thus have the potential for an additive effect. The greatest concern with the organochlorine is the long-term effects. The U.S. EPA has concluded that DDT, DDE, and DDD are probable human carcinogens. On this basis, both Canada and the U.S. banned the organochlorine however; they continue to be very prevalent posing long-term health risks (Davies, 1988).

The organochlorine is still widely used in developing countries including Central and South America, India, China and many other countries. Products imported from these countries are obvious sources of DDT and other organochlorine. They are also transported in air, oceans and bioaccumulation in organisms (Davies, 1988). In fish and wildlife, there is evidence of reproductive and developmental effects as a consequence of chronic exposure. There is increasing concern that exposure of humans to these chemicals may be causing adverse effects on reproductive function. A number of chemicals in the environment possess estrogenic activity and these compounds include pesticides as well as plasticizers, estrogenic agents administered to livestock and a variety of other chemicals (Health and the Environmental, 1995).

The aim of this study was to evaluate the level of chlorinated pesticides in human breast milk among Saudi lactating mothers in order to take precautionary actions if the level was high.

# **Materials and Methods**

## Milk Collections

13 samples of human milk were obtained from healthy mothers at 2 to 4 months of lactation. At this stage of lactation, the milk is mature and the composition and concentration of constituents have been shown to be relatively stable (Parr *et al.*, 1991). All mothers ranged in age from 17 to 42 years. The mother was requested to express the milk totally from one (right) breast with manual pump in the morning, taking care the milk only made contact with glass surface to prevent contamination. In order to prevent the possible contamination, all glass parts, and samples vials were rinsed in 10% (v/v) pure nitric acid and deionized water before use. Samples were collected in sterilized plastic containers and transported to the laboratory on ice and frozen at  $-20^{\circ}$ C immediately on arrival until analyzed.

## Sample Preparation

A relatively new technique in analytical laboratories is the use of supercritical fluid extraction (SFE) as part of the complete sample preparation method to which a sample is subjected before analysis. The purpose of sample preparation is to isolate chlorinated pesticides from the rest of the sample (Lehotay, 1997). Sample preparation, extraction and clean up were carried out according to the procedure of US FDA (PAM-1). HP 7680 Supercritical Fluid Extractor was used to extract chlorinated pesticides from breast milk.

### Gas Chromatographic Determination

Chlorinated pesticide residues in the last fraction from SFE were determined by gas chromatography using Agilent, 6890 Series, GC system with electron capture detector. The conditions used for the analysis were same as recommended by PAM 1 (Pestdata) to facilitate identification by comparison of the relative retention times. HB5 capillary column coated by H.P. was connected to an electron capture detector at 300°C. Injection port temperature was maintained at 210°C while the column initial temperature was 50°C, this was held for 1 min and then programmed to 190°C at 30°C/min. Finally, 210°C at 3.6°C/min the final temperature was held for 3 min. Helium was used as carrier gas (2 ml/min) and 1 ul sample was injected splitless.

Identification of the peaks in the sample was carried out by comparing their retention times with those in the standard mixture and also their relative retention time (relative to chloropyrifos) to the Pestdata in PAM-1.

#### Results

In the present study, a total of 13 random samples were collected. All of these donors were Saudis. In most of the cases, only one sample was collected. Table 1 lists the levels of pesticides found in milk samples. It can be seen from this table that only 4 samples contained DDE which ranged from 0.01-0.02 ppm. In addition to DDE, related residues were also present though in smaller amount. DDT was detected in one sample only and it was 0.043 ppm.

No.	Nationality	α-HCH	Lindane	B-HCH	Нер-ерх	Aldrin	Dieldrin	Endrin	DDE	DDD	DDT
1	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
2	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
3	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
4	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.02	0.0	0.0
5	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
6	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.01	0.0	0.0
7	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
8	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
9	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.01	0.0	0.0
10	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
11	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
12	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
13	Saudi	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.01	0.0	0.043

TABLE 1. Level of organochlorines pesticide residues in breast milk from Saudi lactation mothers  $(ng/\mu l \text{ solvent} = ppm)$ .

## Discussion

When reviewing literature on occurrence and toxicity of the commonly reported industrial environmental pollutant chemicals in human milk, it is lot; chemicals that are not normal constituents of human milk should be considered undesirable contaminant. The following substances were detected in human milk, which are: persistent organochlorine pesticides, polybrominated compounds, polycyclic aromatic hydrocarbons, trace elements, mycotoxins, nitrate, nitrite, nitrosoamines, nicotine, caffeine, ethanol and drugs (Somogyi and Beck, 1993).

Food is the primary route of exposure. Foods, which may contain DDT, include: meat, fish and poultry, diary products and root and leafy vegetables (Davies, 1988).

The exposure of the nursing infant to pesticides that contaminate breast milk, which are the halogenated ring structures like dichllrodiphenyl trichloroethane (DDT) and polychlorinated biphenyls (PCBs) was well known (EPA, 1975). These pesticides appear widely in milk without specific or even known exposure on the part of the mother. They are not well excreted, and are in milk at concentrations higher than serum such chemicals are generally stable ring structures with variable halogenations, usually chlorination (EPA, 1975). The compounds are lipophilic and so apparently concentrate in milk, which is about 3% fat while serum is usually less than 1% fat (EPA, 1975).

Fat soluble materials such as the organochlorine, DDT and chlordane, can be stored for long periods of time in maternal body fat. Body fat mobilization and turnover are increased during lactation and fat-soluble substances may also be mobilized. Fat-soluble substances may be released from fat during weight loss, which typically occurs during lactation (Hendershot, 1984).

The actual amount of pesticides that the breast-feeding infant receives is related to the intake during pregnancy via the placenta and the concentration of pesticides in the breast milk, both of which are related to the maternal body burden, as well as the volume and duration of breast-feeding (Heifetz, *et al.*, 1989).

Saudi Arabia does not have a long history of pesticide use in agriculture or public health. However, imported foods, which form the bulk of the total supply, has not been strictly controlled from the pesticide residues point of view. This may explain the presence of pesticides in breast milk of lactating mothers who might consume such food. In addition, large part of the population spends their vacation abroad and their they might expose to such contamination.

Comparison of the finding of results with the similar study conducted in the Eastern province of the Kingdom specifically in Al-Ehssa region by Al-Saleh *et* 

*al.* (2003) revealed that the average ranks of p, p' DDE, p, p' DDT and sigma p, p' DDT in lactating mother were significantly higher than those living in Riyadh region and obviously than in Jeddah region. The authors attributed that to the implication of the spraying activities to control vector borne disease in Al-Ehssa region. Also comparing our finding of results with study conducted in Kuwait by Saeed *et al.* (2000) indicated that the average of DDE residue in Kuwaiti donors is 833 microgram/kg and DDT averaged 12.4 microgram/kg fat. They concluded that the levels of DDT pesticides in Kuwaiti milk samples were lower than levels reported from other Middle Eastern countries, although methodologies may not be directly comparable.

The conclusion is that comparing the average levels of organochlorine pesticides in the breast milk of lactating mothers in Saudi Arabia with those reported from other countries of the middle east, such as Kuwait, Iran, Jordan, Egypt, and Iraq, respectively (Saeed *et al.*, 2000; Cok *et al.*, 1999; Alawi *et al.*, 1992; Dogheim *et al.*, 1991 and Al-Omar *et al.*, 1985) it appears that the levels found in the present study was relatively low.

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تقدير المبيدات الكلورية في حليب الأمهات المرضعات السعوديات

*المستخلص.* يمكن أن يظهر التلوث البيئي في حليب الأمهات ، كما هو الحال في مختلف المنتجات الغذائية الأخرى ، وكذلك مياه الشرب ، والتي بدورها يمكن أن تنتقل إلى الأجنة مسببة تهديدات خطيرة لصحة الأطفال الرضع والأطفال الأكبر عمراً، من بين هذه المركبات، الأنواع المختلفة من الكلورينات العضوية مثل المبيدات الحشرية وأيضاً نوع من السموم الفطرية ، وهو ما يسمى بالأفلاتوكسينات.

الهـدف من هذه الدراسة هو تقدير مستوى الكلورينات العضوية الموجودة في المبيدات الحشرية في حليب الأمهات المرضعات السعوديات في مدينة جدة.

تم تجميع ١٣ عينة من حليب الأمهات السعوديات عشوائيًا حيث تم تقدير كمية المبيدات الحشرية الكلورونية ، وذلك عن طريق استخلاص المبيد الحشري باستخدام جهاز SFE ، ثم حقن المستخلص في جهاز الكروماتوجراف الغازي.

من خـلال النتـائج وجـد في أربعـة عينات فقط مـادة DDE وكـانت تتراوح تراكيزها ما بين (٠١, • إلى ٠٢, • من الجزء من المليون).

كما وجدت مادة DDT في عينة واحدة فقط وكان تركيزها (٣٤ • ، • من الجزء من المليون).

ووجد أن تركيز كل من DDE & DDT في عينات الحليب منخفضة دائمًا مقارنة بالدراسات الأخرى في دول الشرق الأوسط.