

Adipose Tissue Burden of Legacy Organochlorine Insecticides in Sudanese Women: A Pilot Study of Breast Cancer Cases and Controls

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Abstract

Cancer is a group of diseases characterized by the uncontrolled growth and spread of abnormal cells. Breast cancer (BC) is one of the most common types of cancer that occurs mostly in females. BC is a malignant tumor that starts in the cells of the breast. Persistent organic pollutants (POPs) are synthetic organic chemicals. POPs, e.g. organochlorines pesticides (OCP) and polychlorinated biphenyls (PCBs), are persistent and lipophilic. The commonly used OCP are DDT, lindane, aldrin, dieldrin, isodrin, endrin, endosulfan, heptachlor, chlordane and toxaphene. Their use was stopped in the Sudan season 1980/81. This study aims to check the presence of OCP in the breast fat/adipose-tissues collected post-operative from BC and fibroadenoma patients. A total of 20 samples were collected from Wad Medani Teaching Hospital, Gezira State (3 fibroadenoma and 17 BC). The patients were from 5 locations, viz. east, west, central south Gezira, Omdurman, Sennar and Gedarif. They were divided into 5 age-groups, viz. 20-25, 35-40, 41-45, 46-50, and 51-55 yr-old. The Folch (1957) method for extraction was adopted. Analysis was done using GC- MS technique. The results showed that in East Gezira, OCP detected were *o,p'*-DDT in age 50-yr-old (BC; 0.399 ppm), *p,p'*-DDE in 53-yr-old (5.192 ppm), dieldrin in 25-yr-old (benign; 0.492 ppm), 54-yr-old (BC; 0.513 ppm) and heptachlor in the previously mentioned 25-yr-old (1.416 ppm). In Central Gezira, the *p,p'*-DDE registered highest concentration (11.558 ppm) in 50 yr-old BC patient, followed by *o,p'*-DDE (0.861 ppm) in 45-yr-old and aldrin showed lower concentration of 0.029 and 0.049 ppm in 45 and 54 yr-old BC patients, respectively. In Western Gezira, the presence of *o,p'*-DDE, aldrin and dieldrin with concentrations of 1.238, 0.49 and 0.398 ppm in 36 and 44-yr-old, respectively. In Southern Gezira *o,p'*-DDE at 2.156 ppm and Lindane at 0.074 ppm were detected in 50-yr-old BC patient. In other areas, viz. Omdurman, lindane and endosulfan at 0.123, and 0.913 ppm, were detected in 48-yr-old BC patient, respectively. All these concentrations proved to be very much higher than those reported in the literature. For Gedarif and Sennar BC patients, no OCP was detectable. These results suggest that OCP exposure, especially for those coming from agricultural areas (Gezira), have high cancer risk potential. More detailed studies with higher number of samples from different agricultural (irrigated and rainfed) areas are needed to investigate the presence of OC, dioxins/furans, polyaromatic hydrocarbons (PAHs) and PCBs.

Keywords: Breast Cancer; Organochlorines Insecticides; DDT; Aldrin; Dieldrin; Heptachlor; Chlordane; Endosulfan; Sudan

Introduction

Cancer is a group of diseases characterized by the uncontrolled growth and spread of abnormal cells [1]. If the spread is not controlled, it can result in death. Cancer is caused by both external factors, e.g. tobacco, infectious organisms, chemicals, and radiation, and internal factors. e.g. inherited mutations, hormones, immune conditions, and mutations that occur from metabolism. These causal factors may act

together or in sequence to initiate or promote the development of cancer [2]. Certain cancers are related to infectious agents, e.g. human papillomavirus (HPV), hepatitis B virus (HBV), hepatitis C virus (HCV), human immune deficiency virus (HIV) and bacterium, *Helicobacter pylori* stomach [1,3]. Cancer is treated with surgery, radiation, chemotherapy, hormone therapy, immune therapy, and targeted therapy [4].

Breast cancer (BC) can occur in both men and women, but it's far more common in women. BC is a malignant tumor that starts in the cells of the breast. A malignant tumor is a group of cancer cells that can grow into (invade) surrounding tissues or spread (metastasis) to distant areas of the body. Two types of BC are carcinomas and sarcomas are known [5].

Persistent organic pollutants (POPs) are synthetic organic chemicals. They are all synthetic chemicals, intentionally or non-intentionally produced/released. Some are pesticides; others are industrial products or unintended by-products resulting from industrial processes or combustion [6]. POPs, such as organochlorine pesticides (OCP) and polychlorinated biphenyls (PCBs) are synthetic chemical that are highly resistant to biodegradation. Due to their persistence (stability) and lipophilicity, POPs tend to bioaccumulate and biomagnify in food-chain/web. Furthermore, POPs can be transported over long distances in atmosphere (transboundary movement) and, therefore, be distributed to other regions [6].

Public concern about contamination by POPs increased recently, because several of these compounds are identified as hormone/endocrine disrupters (EDs), which can alter normal function of endocrine and reproductive systems in humans and wildlife. There are many risks and effects of having these chemicals in our environment and none of them are a benefit to the earth. After these pollutants are put into the environment, they are able to stay (persist) in the system for decades causing problems, e.g. cancer, birth defects, learning disabilities, immunological, behavioral, neurological and reproductive discrepancies in human and other animal species. The Stockholm Convention (SC) on POPs was adopted in May 2001 with the objective of protecting human health and environment from POPs [6].

OCPs are a large class of multipurpose chlorinated hydrocarbon chemicals were used to control insect pests in field crops, horticultural crops, termites, public health and veterinary pests since their discovery before the WWII up to 1973 in some countries and 1980s in others [6]. These chemicals are difficult to degrade into less hazardous substances in the environment. OCP are lipophilic compounds that tend to bioaccumulate in fatty/adipose tissues through the food-chain. Bioaccumulation of OCP is defined as a *log* Kow value > 5 or bioaccumulation factor in aquatic species >5000. These pesticides semi volatile, enabling their entry the atmosphere transport over long distance globally, mainly by air mass movements [7]. Most of POPs are OCP, namely aldrin, endrin, isodrin, chlordane, DDT, dieldrin endosulfan and hexachlorobenzene (HCB), heptachlor, chlordane, mirex, kepone and toxaphene.

History of OCP in Sudan

Cotton cultivation in Gezira Scheme started in 1911 on a small-scale of 250 feddans (F = 4200m²). Currently, the area increased to ca. 2 million F. The annual cropping structure is composed of wheat, cotton, sorghum, groundnuts, and vegetables. Cotton, the main cash crop, and the main recipient of pesticides, the pesticide application is directed against the pest complex. However, the relative importance of each pest changed over the years. Large-scale insecticide spraying started in the scheme season 1945. OCPs were the first to be used (*viz.* DDT). DDT and dieldrin were banned (1981). In addition to the Gezira Scheme, which is the largest agricultural scheme in the world under one administration, there are several other schemes or corporations that uses pesticides. These cotton producing schemes are: The Rahad Agricultural Corporation (RAC); New Halfa Agric. The previous Nuba Mountains Corporation (rain-fed) used to apply some POPs pesticides in what is known as the Modernized Sector [6]. Currently, Sudan does not produce pesticides. In 1963, Shell Chemicals (S.I.C.C)-East Africa (EA) established a formulation plant to produce insecticides for the agricultural schemes at Marinjan, Wad Medani, Gezira State. The blending capacity was 7,000 I.G./shift (1.5 - 2.3 million Gallons within 180 days). The plant started in 1964 with production of large quantities of OCPs, i.e. endrin, DDT, dieldrin and toxaphene. These insecticides were used for cotton pests' control in the irrigated and rain-fed sectors. The products were of high quality with competitive prices and were distributed via Sudan Railways [6].

The imports of the old POPs pesticides stopped by 1980/81; however, imports for public health, veterinary use and sugarcane plantations continued up to early 1990s. However reliable data is not available whether in the schemes or the central Bank. Toxaphene was introduced as Torbidan® (methyl parathion + toxaphene + DDT) or as HelioTox (DDT + toxaphene). Endrin was withdrawn from the Sudanese market as early as mid-1960's, because of its high acute mammalian toxicity. Aldrin and dieldrin were mostly used for the preparation of seed dressing of cotton mainly at 3 g /kg seed (Aldrex-T® and Dieldrex-B®), and sometimes for rat control campaign and locust control [6]. The use of endosulfan continued up to 5 yr ago and was banned together with Lindane and became part of the POPs group. Lindane was used for cattle ectoparasite treatment, is still available in the markets, most likely smuggled from the neighboring countries [6].

Most of the published studies measured these OCP in blood. Plasma, and serum. Far fewer studies measured actual breast or body fat / adipose tissue. Since OCPs are lipophilic, adipose tissue often better reflects long-term accumulation. The study can demonstrate: a) The presence of OCP residues in adipose tissue of Sudanese women, b) which compounds are still detectable decades after many of them were banned or restricted, c) differences in residue profiles among age groups, d) geographic variation among women from different agricultural and urban settings, and e) the feasibility of adipose-tissue biomonitoring in Sudan.

Materials and Methods

Materials

- **Chemical and reagents:** Hexane, sodium anhydrous sulfate, distilled water, florisil.
- **Equipment and glassware:** Orbital Shaker, centrifuge; Oven, conical flasks (100 ml), plastic containers (5, and 50 ml), chromatographic columns, refrigerator, deep freezer, GC-MS and digital balance.

Methods

Collection of samples: The sample were collected form Wad Medani Teaching Hospital, Gezira State. Around 1 - 3g from fatty tissue/ patient were collected from the Operating Theaters, immediately coded and stored at -24 - 70°C up to the time of chemical analysis [8]. These samples were taken from BC patients and non-BC patients grouped in 5 age-categories: 20-25yr, 35-40yr, 41-45yr, 46-50yr, 51-55yr. These patients came from 5 Gezira administrative localities (Eastern Gezira, Central Gezira, Western Gezira, Southern Gezira) and other areas, viz. Omdurman, Sennar and Gedarif.

Sample extraction

Folch method: The tissue (1g) was homogenized with 30 ml hexane, agitated for 20-30 minutes in an orbital shaker at room temperature. After centrifugation at 2000 rpm, 2 phases were identified: upper phase (non-lipid) and lower phase (solvent and lipid). Clean up of the homogenate was performed using chromatographic columns packed with activated florisil (10g) (oven activated at 660°C), 1g sodium sulfate and 10 ml hexane for washing the sample. Another 10 ml hexane was used to wash the sample. The rinsate was left to evaporated and the final 3-5 ml was stored in refrigate -24-70° [9].

Analytical method: The qualitative and quantitative analysis of the samples was carried out by using GC/MS (GC/MS-QP2010-Ultra', Shimadzu Company), equipped with capillary column (Rtx-5ms-30m×0.25 mm×0.25µm). The sample was injected by using Splitless mode, instrument operating in EI mode at 70eV. Helium was the carrier gas passed with FR = 1.69 ml/min.

The temp. program: started from 50°C holding in 1 minute, with rate 20°C/min to 140°C, then the rate was changed to 10°C/min reaching 280°C as final temp. degree with minutes as hold time. The injection port temp. was 280°C, the ion source temp. was 200°C and the interface temp. was 250°C. The sample was analyzed by using scan mode in the range of m/z 50-500 charges to ratio and the total run time was 19 minutes. Identification of components for the sample was achieved by comparing their retention times (Rt; the table below) and mass fragmentation patents with those available in the library, the National Institute of Standards and Technology.

Retention times (Rt) of each tested insecticide

ID#	Name	Rt
1	Alpha-Lindane	9.325
2	Heptachlor	14.217
3	Aldrin	14.942
4	o,p'-DDE	16.275
5	Endosulfan II	16.508
6	p,p'-DDE	16.794
7	Dieldrin	17.033
8	Endrin	17.483
9	o,p'-DDT	17.817

Table

Results

According to location

East Gezira: This locality is represent by 5 villages and towns (Elfe'ig, Eltayif, Elrabkara, Abuharaz and Rufa'a town (Table 1-5). Out of all OCP, only 4 were detected in the locality patients samples. These were *p,p'*-DDT, *o,p'*-DDE, dieldrin and heptachlor. *o, p'*-DDT was detected only in one area (Rufa'a town; 0.399 ppm), whereas the metabolite *o, p'*-DDE was detected only in a patient from Elrabkara village (5.192 ppm). Dieldrin was found in patients from Elrabkara (0.0492 ppm) and Abuharaz (0.513 ppm) villages. Heptachlor was reported in a patient from Abuharaz village also (1.416 ppm).

Sample no.	Age (yr)	Home	OCP								
			<i>P,p'</i> -DDT	<i>Ö,P</i> -DDE	<i>p,p'</i> -DDE	Ald/	End.	Dield.	Lind.	Hepta	Endos.
1	52	Elkar-aiba	ND								
2	42	Elfe'ig	ND								
3	44	Elhuda	ND			0.009	ND				
4	53	Eltayif	ND								
5	42	Wad Medani	ND								
6	42	Gedarif	ND								0.913
7	45	Wad Medani	ND	0.861	ND	0.029	ND				
8	36* Fibro.	Wad Medani	ND								
9	25* Fibro.	Abu-haraz	ND				0.513	ND	1.416	ND	
10	21* Fibro.	Wad Medani	ND								

11	54	Wad Medani	ND	0.619	ND	0.046	ND		
12	48	Omdur-man	ND					0.123	ND
13	52	AmaratElbana	ND	2.156	ND			0.074	ND
14	46	Barakat	ND						
15	53	Alrab-kara	ND		5.192	ND		0.492	ND
16	50	Sennar	ND						
17	50	Rufa'a	0.399	ND					
18	45	Gedarif	ND						
19	36	Elmanagil	ND		1.239		ND	0.398	ND
20	50	Algraigrieb	ND						

Table 1: OCP detected in each sample (20 samples) for both BC and fibroadenoma patients by age and patient geographic area.

ND = Not Detected.

East Gezira	Concentration (ppm)			
	<i>p,p'</i> -DDT	<i>o,p'</i> -DDE	Dieldrin	Heptachlor
Elfe'ig	ND			
Eltayif	ND			
Elrabkara	ND	5.192	0.0492	ND
Abuharaz	ND		0.513	1.416
Rufa'a	0.399	ND		

Table 2: OCP Levels in fatty tissues collected from 5 villages of Eastern Gezira.

ND= Not Detected.

Central Gezira: The locality consists 6 administrative units. The results in table 1 and 3 revealed the presence of OCP levels found in Wad Medani, the capital of the state, and Algraigrieb village north of Wad Medani. Three compounds were detected, viz. *p, p'*-DDE, *o,p'*-DDE and aldrin. Wad Medani patients showed the presence of *o, p'*-DDE (0.861 ppm) and aldrin (0.029 ppm). Whereas the village patient adipose tissues showed the presence of high level of *p, p'*-DDE only (11.558 ppm).

Central Gezira	Concentration (ppm)		
	<i>o,p'</i> -DDE	<i>p,p'</i> -DDE	Aldrin
Wad Medani	0.861	ND	0.029
Algraigrieb	ND	11.558	ND

Table 3: OCP levels in fatty tissues collected from Central Gezira (Wad Medani town and Algraigrieb village).

ND = Not Detected.

West Gezira: This is represented by samples taken from patients came from Elmanagil town and Elhuda agricultural village (Table 1 and 4). The town patients adipose tissues showed the presence of 3 OCP, *i.e.* *o,p'*-DDE, aldrin and dieldrin with concentrations 1.238, 0.49 and 0.398 ppm, respectively. Only aldrin was detected from a patient from Elhuda (0.009 ppm).

West Gezira	Concentration (ppm)		
	<i>O,p'</i> -DDE	Dieldrin	Aldrin
Elmanagil town	1.238	0.398	0.49
Elhuda village	ND	ND	0.009

Table 4: OCP levels in fatty tissues collected from West Gezira (Elmanagil Town and Alhuda village).

ND= Not Detected.

South Gezira: The patients came from Amarat Albana village and Barakat suburb (about 5 km south of Wad Medani Town; the HQ of Gezira Scheme). Table 1 and 5 revealed that only Amarat Albana patient had residues of *o,p'*-DDE (2.156 ppm) and Lindane (0.074 ppm).

South Gezira	Concentration (ppm)	
	<i>o,p'</i> -DDE	Lindane
Amarat Elbana	2.156	0.074
Barakat	ND	

Table 5: South Gezira (Amarat Elbana village and Barakat suburb).

ND= Not Detected.

Other areas (Omdurman, Sennar and Gedarif): The patients came from Omdurman (the National Capital of the Sudan), Sennar (The capital of the Sennar State), and Gedarif Town (The Capital of Gedarif State). The results (Table 6) showed the presence of lindane and endosulfan at 0.123, and 0.913 ppm, respectively, in the tissues of Omdurman patient only None was detected in the tissues of the patients of the other 2 towns.

Locality	Concentration (ppm)	
	Lindane	Endosulfan
Omdurman	0.123	0.913
Gedarif	ND	
Sennar	ND	

Table 6: OCP residue levels in fatty tissues taken from BC patients came from Omdurman and Gedarif.

According to age- group

The age has been categorized to five groups. Group I (20-25 yr-old; Table 7) were fibroadenoma patients. The rest of the groups (11-V) were BC patients (Table 8-11).

Group I: Fibroadenoma: The patients of this group were 21, 22, and 25-yr-old (Table 7 and 8). The OCP detect were heptachlor and dieldrin only in the 25yr-old patient from Abuharaz village, with concentration 1.416, and 0.513 ppm, respectively. The other 2 younger patients, from Wad Medani Town, tissues proved to be clean of OCP residues.

Age/yr	Concentration (ppm)	
	Dieldrin	Heptachlor
21 (Wad Medani)	ND	
22 (Wad Medani)	ND	
25 (Abuharaz village)	0.049	1.416

Table 7: Group I of age (20-25yr-old): OCP levels in fatty tissues collected from three fibroadenoma patients.

Group II of age (35-40 yr-old) patients with BC: *p,p'*-DDE, aldrin, dieldrin were detected in a 36 yr-old patient (Table 8). The concentrations were 1.239, 0.049, and 0.389 ppm, following the same order. The Wad Medani 36 yr-old patient sample was clean of any tested OCP.

Age	OCP (ppm)		
	<i>p,p'</i> -DDE	Aldrin	Dieldrin
36 (Elmanagil)	1.239	0.049	0.398

Table 8: Group II of age (35-40) OCP levels in fatty tissues collected from one patients with BC.

Group III of age (41-45 yr-old) patients with BC: The patients of this group were 6 out of the 20 patients. Three of them were 42-yr-old, one 44-yr-old and 2 patients 45-yr-old (Table 9). OCP were detected in 3 patients only. The compounds were *o,p'*-DDE, endosulfan, and aldrin in one of the three 42-yr-old (endosulfan, 0.913 ppm), one of the 44-yr-old (aldrin, 0.029ppm) and the 45-yr-old (*o,p'*-DDE, 0.861 ppm and aldrin 0.009 ppm).

Age/yr	OCP (ppm)		
	<i>o,p'</i> -DDE	Endosulfan	Aldrin
42 (Gedarif)	ND	0.913	ND
42 (Wad Medani)	ND	ND	ND
42 (Elfeig)	ND	ND	ND
44 (Elhuda)	ND	ND	0.029
45 (Wad Medani)	0.861	ND	0.009
45 (Gedarif)	ND	ND	ND

Table 9: Group III of age (41 - 45) BC patients.

Group IV of age (46-50-yr-old): patients with BC: The patients of this group were 5, one of them was 46-yr-old from Barakat suburb, another was 48 yr-old from Omdurman Town and 3 were 50-yr-old from Rufaa Town, Algraigrieb village, and Sennar Town (Table 10). The later 2 locations patients sample proved to be clean of any OCP. Whereas Rufaa sample carried 0.399 ppm of *p,p'*-DDT. Moreover Omdurman 48-yr-old sample registered 0.123 ppm of lindane.

Age (yr)	OCP (ppm)			
	<i>p,p'</i> -DDT	<i>p,p'</i> -DDE	Dieldrin	Lindane
46 (Barakat)	ND	ND	0.686	ND
48 (Omdurman)	ND	ND	ND	0.123
50 (Rufaa)	0.399	ND	ND	ND
50 (Algraigrieb)	ND			
50 (Sennar)	ND	ND	ND	ND

Table 10: Group IV (46-50) collected from five BC patients.

Group V of age (51-55-yr-old): patients with BC: The group is represented by 5 patients, 2 of them of the were 52-yr-old from Amarat Albana and Alkaraiba villages, the 3rd is 54-yr-old from Alrabkara village, and 2 patients 53-yr-old from Altaif and Wad Medani (Table 11). Alkaraiba patient sample proved to be clean of any OCP. The rest of this age group with BC proved to contain in their tissue a number of these OCP, at least 2 of them. *o,p'*-DDE was detected in 52-yr-old Amarat Albana patient (2.156 ppm), also 54-yr-old Wad Medani patient (0.619 ppm). Regarding *p,p'*-DDE, it was found in 53-yr-old (Alrabkara village) at 5.192 ppm, in addition to 0.492 ppm of dieldrin. Aldrin was found in the 54-yr-old Wad Medani patient (0.046 ppm).

Age (yr)	OCP (ppm)				
	<i>o,p'</i> -DDE	<i>p,p'</i> -DDE	Aldrin	Dieldrin	Lindane
52 (Amarat Albana)	2.156	ND			0.074
52 (Alkaraiba)	ND				
53 (Alrab-kara)	ND	5.192	ND	0.492	ND
53 (Eltayif)	ND				
54 (Wad Medani)	0.619	ND	0.046	ND	ND

Table 11: Group V of age (51 - 55) five BC patients.

To summarized results, even the 3 younger group, i.e. the fibroadenoma patients, 1 of them (Abuharaz village patient, her sample showed the presence of dieldrin and heptachlor. Moreover, 10 out of the 17 BC sample contained 1 or more of the OCP. Also, 6 of the 17 BC samples were clean from these chemicals and their metabolites. Even the samples taken from the capitals of the states (Wad Medani, Omdurman, Gedarif and Sennar) showed the presence of some of these OCP. The age group 36 - 40 only *p,p'*-DDE and dieldrin were detected, whereas in the older group 41 - 45, 3 OCP were reported, viz, *o,p'*-DDE, aldrin and endosulfan. The 4th group (46 - 50) showed the presence of 4 OCP, i.e. *o,p'*-DDT, *p,p'*-DDE, dieldrin and Lindane. Finally, the > 50 yr-old group registered 5 chemicals, these were *o,p'*-DDE, *p,p'*-DDE, aldrin, dieldrin and Lindane.

Location	Concentration (ppm)							
	<i>o,p'</i> -DDT	<i>o,p'</i> -DDE	<i>p,p'</i> -DDE	Aldrin	Dield.	Hepta-chlor	Lind.	Endos.
E. Gezira	0.399	ND	5.192	0.009	0.492-0513	1.416	ND	
C. Gezira	ND	0.861-619	11.558	ND				
W. Gezira	ND	ND	1.238	0.49	0.398	ND		
S. Gezira	ND	2.156	ND		0.686	ND	0.074	ND
Others	ND						0.123	0.913

Table 12: Summary of the OCP levels in adipose tissues collected from BC came from patients of Gezira State vs. others (Omdurman and Gedarif State).

Discussion

The exposure of the general Sudanese population to these OCP is assumed to be mainly through food, since the use of this group of insecticides was associated primarily with the earlier agriculture pest control operations in the vegetables, field crops and animal production (meat, milk, poultry products) from the early 1950s to almost mid-1980s. except endosulfan and Lindane up to 2020.

The results of the present study revealed higher levels of OCP in the samples taken from older age women, attributed to longer exposure leading to pesticide bioconcentration [10]. According to location, East Gezira, represented by Rufa'a, Elrabkara village Abuharaz, *o,p'*-DDT was detected in a relatively lower concentrations. This indicates recent use of DDT in the area [11], in addition to long-term exposure before 1981 [12]. The other metabolite, *o,p'*-DDE also indicates the historical exposure since DDT use in the area was stopped when OPs, carbamate and pyrethroids were introduced [6], but due its persistence and fat-solubility, it became detectable [6,12]. Dieldrin and heptachlor in the patients with fibroadenoma were detected at relatively lower concentrations. Most of their use was mainly for seed treatment and termite's control [6,10]. Aldrin is rapidly converted to its epoxide dieldrin, and used for the same abovementioned purposes [6].

In Central Gezira samples, the OCP levels were higher. The area is represented by Algraigrieb village and Wad Medani Town. *p,p'*-DDE level was higher in Algraigrieb village, resulting from the old practices as an irrigated agricultural area. Whereas *o,p'*-DDE in Wad Medani town was detected in lower concentration. This could also be attributed to the former use of DDT in controlling mosquitoes, and the use of DDT in schools for lice control among young school children during the 1950s and 1960s. Aldrin level in Wad Medani patient was relatively lower than that of the other areas, because a families in the town used to apply it for termites control.

West Gezira, viz. Elmanagil town and Elhuda, both are agriculture areas for cotton, sorghum, groundnuts, and horticultural crops, and high animal population, in addition to twice a year mosquito control campaigns. DDT at the earlier times was extensively used. *O, P'*-DDE level was relatively high. Women who lived within 1 mile of hazardous waste sites containing OCP were found to have an increased risk for BC [13,14]. However, the same authors stated that no association was found between BC and pesticides detected in drinking water or residence on or near agricultural land [15]. Aldrin and dieldrin were extensively used for seed treatment and termites control in the area. The level was relatively low and was in line with 1996 work [16].

South Gezira (Amarat Albana village and Baraka, results revealed that *o,p'*-DDE level was relatively high. This can be justified as above.

Samples taken from patients came from Omdurman showed that lindane was detected at a relatively low level. Most of its use was for lice, mosquitoes and thrips control, especially in onions coming from the farms north of Omdurman. Endosulfan was also relatively low, since it was the last to be banned together with Lindane around 5 yr ago [6].

Therefore, it can be concluded that these 3 states have different agricultural practices, crops, and health practices, and all of them are mainly irrigated agricultural schemes, except Gedarif, which is a rainfed area. Cotton production with, a very long history goes back to early 1950s is the main crop, followed by sorghum sesame and groundnuts and others. Therefore, they have different histories, of pesticide use and patterns, in addition to urban and rural characteristics.

According to age group, group I (fibroadenoma patients), the OCP found in 25-yr-old patient were heptachlor (1.416 ppm) and dieldrin (0.049 ppm), which refers to short exposure period and might be associated with ER [11]. Group II (35-40 yr-old) patients with BC results revealed that the OCP were found in the 36-yr-old sample from Elmanagil Town (*p,p'*-DDE 1.238 ppm). Previous studies suggest that an association may be evident only among women exposed to DDT during biologically susceptible among those diagnosed with estrogen receptor/progesterone receptor-positive (ER+PR+) BC subtypes. Self-reported acute exposure to a fogger truck, which sprayed DDT prior

to 1972, was hypothesized to increase the risk of BC, particularly among women exposed at a young age or diagnosed with ER+PR+ BC [12]. Most studies suggested higher risk for exposures, during breast development, to DDT. Aldrin (0.049 ppm) and dieldrin (0.398 ppm) were also reported in this sample. Aldrin epoxide, i.e. dieldrin, reflects hazard ongoing [17].

Group III (41-45 yr-old) patients with BC results showed the *o,p'*-DDE was found at relatively low levels, which might reflect that the hazard might continue up to premenopausal, associated with ER. Aldrin and endosulfan levels were relatively lower than dieldrin.

Group IV (46-50 yr-old) patients with BC, showed detectable *o,p'*-DDT low level was relatively low the explain that might be the *o,p'*-DDT as the was only OCP. This indicates chronic exposure associated with ER [16]. Previous studies suggest that the elevation in risk for endogenous levels, women using estrogen therapies have a significantly increased risk of invasive BC [18]. Also *p,p'*-DDE level was relatively higher in older woman with BC. This mean high association with ER and ability to induce p450 Enzyme [19]. Woman of older age, at first birth, had an increased BC risk than the woman younger age at first birth [15].

Group V (51-55 yr-old) patients with BC the OCP represented *O,P'*-DDE the level was relatively higher the explain that long exposure time and relative by estrogen receptor (ER). Increase the age, increase exposure time period. Associated among postmenopausal women, there were no statistically significant differences observed in the risk factor profiles for ER+PR+ and ER-PR-BC [20]. Aldrin, dieldrin the level was relatively lower their explained that endosulfan level was relatively low the explain that acute toxic and associated with ER receptor the level was relatively lower the explain that acute toxic and used of the licein past. Evidence of an association between long-term body burden of OCPs and BC occurrence in Sudanese women. Current Scientific Consensus (2025-2026).

Based on recent systematic reviews and meta-analyses: There is biological plausibility because many OCP exhibit: estrogenic activity, endocrine-disrupting effects, oxidative stress induction, genotoxic effects.

Aldrin, dieldrin, heptachlor, chlordane and lindane and endosulfan, these compounds have received much less attention than DDT/DDE, but some of them are actually more interesting from a BC perspective, because of their endocrine-disrupting properties and persistence. Aldrin is rapidly metabolized to dieldrin in humans and the environment, so epidemiological studies often evaluate both together. Current evidence indicates no consistent association with BC and most pooled analyses did not find statistically significant increases in risk. Aldrin is generally considered a suspected endocrine disruptor, but not an established BC risk factor [23]. Dieldrin has attracted much more attention. A classic Danish prospective study reported approx. a 2x increase in BC risk among women with higher serum dieldrin concentrations. However, subsequent studies produced mixed findings. Later systematic reviews and meta-analyses found that the overall evidence was insufficient for a definitive causal conclusion. The current interpretation is dieldrin is probably the cyclodiene insecticide with the strongest historical epidemiological signal for BC, but the evidence remains inconsistent [23]. Heptachlor is particularly interesting. The most cited systematic review and meta-analysis of cyclodiene insecticides found that no statistically significant association, except for heptachlor. This does not prove causation, but among cyclodienes, heptachlor showed the strongest pooled association. The current interpretation is that the evidence stronger than aldrin [24]. Possibly stronger than dieldrin in pooled analyses. But, still insufficient for a firm causal conclusion, because the number of studies is small. Chlordane and chlordane-related compounds include: oxychlordane, trans-nonachlor, cis-nonachlor; recent endocrine-disruptor meta-analyses suggest that chlordane exposure may be associated with elevated BC risk [24]. Biologically, chlordane compounds: accumulate in adipose tissue, interfere with hormone signaling, and may affect estrogen-responsive pathways. The current interpretation is the evidence is stronger than for aldrin, but weaker than that required to establish causation.

Lindane recent meta-analyses of endocrine-disrupting chemicals reported increased BC risk associated with its exposure, especially when measured in blood biomarkers. Several studies have investigated γ -HCH (lindane), β -HCH, total HCH burden. The evidence is suggestive, but not conclusive. The current interpretation is Lindane currently has more epidemiological support than aldrin and possibly similar support to chlordane [25].

Endosulfan is somewhat different. The mechanistic evidence is strong. Laboratory studies show that endosulfan can act as an endocrine disruptor, induce oxidative stress, alter estrogen signaling, and affect mammary cell proliferation. However, compared with DDT, dieldrin, or chlordane, very few human epidemiological studies directly link endosulfan exposure with BC. There is insufficient evidence for quantitative meta-analysis. The current interpretation is endosulfan is a compound with strong biological plausibility, weak human epidemiological evidence. Thus, it is often classified as a potential mammary carcinogen requiring further study, rather than a demonstrated human BC risk factor [26].

If we were ranking them today (2026) based on the overall epidemiological literature:

- **Higher concern:** Heptachlor, dieldrin, chlordane compounds, and Lindane (γ -HCH).
- **Lower concern:** Because evidence is limited: endosulfan and aldrin.

The key point is that none of these insecticides currently has epidemiological evidence as strong as that linking tobacco smoking to lung cancer or asbestos to mesothelioma. The scientific debate today focuses on whether chronic low-level exposure during critical windows of susceptibility (fetal life, childhood, puberty, and early adulthood) may contribute to hormonally responsive breast cancers.

The present study's results on these BC risk factors according to ER are in accordance with previous epidemiological studies, which do not provide consistent evidence to conclude that development of ER positive BC is associated with exposure to estrogen-related. The present study supports the hypothesis that estrogen-related risk factors or potential estrogenic OCP (HCH) increase the risk of developing ER negative tumors, whereas DDT increases the risk of developing ER positive tumors. However, this finding should be interpreted with caution, due to the limited number of ER negative cases, so whether exposure to estrogenic OCP affect the risk and prognosis of a hormone-responsive BC needs to be clarified. It now seems likely that whatever environmental exposures contribute to the risk of BC, exposure to OCP cannot be ruled out, it should be taken into consideration in future studies that seek to understand environmental risk factors for BC with large number of samples [24].

Conclusion

OCP residues were detected in adipose tissues of Sudanese women, including BC patients and controls. The findings suggest potential differences in residue burden that warrant investigation in larger studies.

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