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External and Occupational Environmental Exposure in Pregnant Women and Levels of Chemical Pesticides in Amniotic Fluid

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Abstract

The study continued for a full year in Tishreen University Hospital in Latakia Governorate, and 190 triplets (father, mother, and newborn) participated. This study aimed to identify the sources of exposure of the pregnant mother to chemical pesticides, such as external and occupational environmental chemicals, and to evaluate the exposure of the human fetus to diazinon and ethion residues. Then, the relationship between potential sources of exposure in the pregnant mother and the concentrations of diazinon and ethion in the amniotic fluid was studied. Consent from the father and mother was obtained to conduct the questionnaire, which included consent to take a sample of the amniotic fluid. A high-performance liquid chromatography device was used to titrate the residues of organophosphorus pesticides in the amniotic fluid, to find relationships. The SPSS Statistics 23.0 program (Statistical Package for Social Sciences) was used to determine the potential sources of exposure in pregnant mothers and the concentrations of diazinon residues in the amniotic fluid. The concentrations of diazinon residues varied between (0.02ppb) and (5.72ppb), and high values of diazinon were associated with exposure. Vocational status among parents and living in the countryside and near greenhouses and fruit tree orchards compared to other places.

Keywords: External environmental exposure; Occupational exposure; Human fetus; Amniotic fluid; Diazinon; Ethion; Syria

Abbreviations : Ppb: Parts Per Billion

Introduction

The spread of chemical pesticide residues in the rural and urban environment constitutes a very important issue due to concerns about their transfer to embryonic biological environments. The use of chemical pesticides to improve food quality in the long-term leads to soil, air, and water pollution [1-3]. Epidemiological studies in humans have revealed the spread of chemical pesticides in umbilical cord blood [4], meconium [5] and amniotic fluid [6-9] and newborn hair [10], and found that chemical pesticides or their metabolites are linked to a group of disorders that appear during the fetal stage, such as an increased rate of miscarriages in pregnant mothers [11], Poor functioning of sex hormones [12] and increased newborn weight [13], and other studies have linked fetal exposure to pesticides to autism [14] and cancers [15].

Chemical pesticides are defined as a group of chemical compounds that destroy harmful biological pests, either by repelling them, killing them, or preventing their reproduction, thus protecting agricultural crops from damage [16]. Diazinon is an organophosphorus pesticide that combats insect pests in soil, field vegetables, and fruit crops. It is used in homes and urban areas and in gardens [17], and epidemiological studies conducted in humans showed that high levels were linked to an increase in the abdominal circumference of the newborn [18], lower birth height [19], and higher risk of low birth weight [20]. Ethion is an organophosphate pesticide that fights insects on citrus trees, fruits, and some vegetables. It is not used in homes [21].

A study showed a relationship between exposure to the pesticide ethion detected in the blood of pregnant mothers and increased miscarriage rates and increased placental oxidative stress [22].

Materials and Methods

Study design

Approval was obtained from the Ethical Committee for Medical Research at the Faculty of Medicine - Tishreen University on 3/31/2022, which is consistent with the ideal Declaration of Helsinki regulating the ethics of medical research, to allow a sample of amniotic fluid to be given during a cesarean section in the operating room. Written informed consent was obtained to collect data with the consent of the pregnant mother. The informed consent document included the title of the study, a simple explanation of its subject, and the nature of the required participation. In the event of approval, the informed consent document was given for her signature. Finally, it was confirmed that there are no interactions that could affect the health of the mother or the child, and the confidentiality of the data and results of the study participants, with the study team ready to answer all questions and inquiries. After approval, the mother and her newborn are given a special serial number for entry into the study, accompanied by the date of entry. Corresponding to the day of birth.

Participants in the study

The study included 190 samples (father, mother, and newborn) of visitors to Tishreen University Hospital in Latakia Governorate.

Evaluation of sources of external environmental exposure during pregnancy

Asked about:

A. The detailed address of the family residence.

B. Residence near the main sources of environmental pollution with chemical pesticides (vegetable and wheat growing fields, fruit tree orchards, greenhouses, nurseries for growing roses and ornamental trees, and public parks).

The questions for this section suggested two answers: yes or no.

Evaluation of sources of occupational exposure during pregnancy

Each parent is asked whether he is a worker or not, and if the answer is "yes," he is asked about the type and place of work.

Collecting amniotic fluid samples during cesarean section

The amniotic fluid was extracted by a specialist doctor and 10ml of it was extracted using a syringe and a 10ml needle. The extracted amniotic fluid sample is placed in a vacuum glass tube, coded, and then transferred directly and in a special container to the fetal laboratory in the College of Science to be kept in the refrigerator at a temperature of -80 degrees Celsius.

Analysis of amniotic fluid samples

Before starting the qualitative and quantitative analyses, the amniotic fluid sample was emptied into small glass containers for the HPLC (High- Performance Liquid Chromatography) device, produced by the Japanese company Shimadzu, to be calibrated after adjusting the device, according to the analysis conditions used for the HPLC device, which included:

Oven temperature: 40°C, flow 0.8ml/min, UV/DAD detector, syringe volume: 20lμl, mobile phase acetonitrile: water ratio (V/V) 25:75.

Statistical analysis

Data on occupational and external environmental exposure and results of laboratory titrations of detected pesticides were collected in an Excel table. Data (qualitative and quantitative) were processed using the statistical program SPSS Statistics 23.0.

Results

Sample description

The total sample consists of 190 samples (mother- father). The ages of the mothers participating in the study ranged (on average, 28.3 years), and the average body mass index before pregnancy was 22.8kg/m^2. The ages of the fathers ranged (on average, 33.8 years). Most of the sample members were educated, 159 (80.7%) of the fathers and 167 (87.9%) of the mothers. The number of female births was 99 (52.1%).

Study of the concentrations of pesticides detected in the amniotic fluid

Table 1 shows the results of the descriptive statistical study of the levels of diazinon and ethion in the amniotic fluid. The values of diazinon concentrations in amniotic fluid ranged between the detection threshold (0.02ppb) and 5.72ppb. The average value (± standard deviation) of diazinon concentrations in amniotic fluid at birth was 0.23 (±0.61) ppb, and the value of the coefficient of variation was (265%).

Table 1: Results of the descriptive statistical study of ethion levels in amniotic fluid and relative classifications (ppb).

The Studied Pesticide	Min (ppb)	Max (ppb)	Mean (ppb)	SD (ppb)	CV (%)
Diazinon	0.02	5.72	0.23	0.61	265
Ethion	0.02	0.98	0.08	0.17	213

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The values of ethion concentrations in the amniotic fluid ranged between the detection threshold (0.02ppb) and 0.98ppb. The average value (± standard deviation) of ethion concentrations in the amniotic fluid at birth was 0.08 (±0.17) ppb, and the value of the coefficient of variation was (213%).

Describe the characteristics of occupational exposure to chemical pesticides among parents during pregnancy

Most of the mothers, 183 (96.3%), are non-workers (housewives) and all of them are not occupationally exposed to chemical pesticides. The number (and percentage) of fathers who are occupationally exposed to chemical pesticides was 14 (8.3%) and they work in the fields of agriculture, pesticide spraying, and vegetable selling.

Describe the characteristics of external environmental exposure to chemical pesticides in the mother during pregnancy

48 (25.2%) of the families live near a vegetable growing field, only 2 (1.1%) near a wheat growing field, and 96 (50.5%) near a public road. 68 (35.7%) are near a fruit tree orchard and 10 (5.2%) are Near green houses. Only 2(1.1%) near a public park (Table 2).

Table 2: Results of the statistical study of bilateral relationships between concentrations of pesticides detected in amniotic fluid and the level of occupational exposure of fathers.

Spearman's rho		Diazinon	Ethion
Father exposed	Correlation Coefficient	.178*	-.075-
	Sig. (2-tailed)	0.014	0.304
	N	190	190

Study the relationship between concentrations of pesticides detected in amniotic fluid and sources of occupational exposure for fathers

A weak, statistically significant positive relationship ($p \leq 0.05$) between diazinon concentrations in amniotic fluid and occupational exposure to pesticides in fathers. There is no statistically significant relationship between ethion concentrations in amniotic fluid and occupational exposure to pesticides in fathers.

Table 3: Results of the analysis of differences between the values of diazinon and ethion concentrations according to the urban level of the place of birth and residence of the parents.

Pesticide Concentration(ppb)	Mother's Place of Birth	N	Mean	Sig-F	Sig-T
Diazinon	Urban	126	0.1655	0.013	0.052
	Rural	64	0.3477		0.118
Ethion	Urban	126	0.0764	0.458	0.844
	Rural	64	0.0816		0.854
Pesticide Concentration (ppb)	**Current residence location**	**N**	**Mean**	**Sig-F**	**Sig-T**
Diazinon	Urban	101	0.0883	0	0.001
	Rural	89	0.3842		0.002
Ethion	Urban	126	0.0756	0.227	0.831
	Rural	64	0.081		0.835

Studying the bilateral relationships between the concentrations of pesticides detected in the amniotic fluid and the urban level of the place of birth of the mother and the current residence of the parents

We note that the average values of diazinon concentrations are high if you live in rural areas ($p \leq 0.01$), and there are no statistically significant differences in diazinon concentrations depending on the urban level of the mother's place of birth. Regarding ethion, there are no statistically significant differences in the concentrations of pesticides detected in the amniotic fluid depending on the urban level of the place of birth and the current residence of the mother (Table 3).

Study of bilateral relationships between concentrations of pesticides detected in amniotic fluid and the neighborhood of the parents' current residence

By studying Table 4, the following observations can be recorded:

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Table 4: Results of the statistical study of the differences between the values of pesticide concentrations measured in the amniotic fluid and the sources of external environmental exposure.

Pesticide Concentration(ppb)	The Field for Growing Vegetables	N	Mean	Sig-F	Sig-T
Diazinon	No	142	0.1864	0.054	0.017
	Yes	48	0.3465		0.019
Ethion	No	142	0.0781	0.645	0.996
	Yes	48	0.0782		0.997
Pesticide Concentration(ppb)	**A Field for Growing Wheat**	**N**	**Mean**	**Sig-F**	**Sig-T**
Diazinon	No	188	0.2291	0.462	0.031
	Yes	2	0.02		0
Ethion	No	188	0.0787	0.359	0.032
	Yes	2	0.02		0
Pesticide Concentration(ppb)	**Public Road**	**N**	**Mean**	**Sig-F**	**Sig-T**
Diazinon	No	94	0.3156	0.002	0.044
	Yes	96	0.1366		0.048
Ethion	No	94	0.0582	0.018	0.21
	Yes	96	0.971		0.21
Pesticide Concentration(ppb)	**Orchard of Fruit Trees**	**N**	**Mean**	**Sig-F**	**Sig-T**
Diazinon	No	122	0.1573	0.001	0.035
	Yes	68	0.3517		0.014
Ethion	No	122	0.0807	0.937	0.783
	Yes	68	0.0735		0.796
Pesticide Concentration(ppb)	**Green Houses**	**N**	**Mean**	**Sig-F**	**Sig-T**
Diazinon	No	180	0.215	0.688	0.023
	Yes	10	0.4401		0.032
Ethion	No	180	0.0743	0.043	0.012
	Yes	10	0.1474		0.014
Pesticide Concentration(ppb)	**Public Garden**	**N**	**Mean**	**Sig-F**	**Sig-T**
Diazinon	No	188	0.229	0.469	0
	Yes	2	0.024		0
Ethion	No	188	0.0787	0.359	0
	Yes	2	0.02		0

We note higher average values of diazinon concentrations in the amniotic fluid of mothers living near greenhouses, fruit tree orchards, or vegetable growing lands ($p \leq 0.05$) compared to those living far from them. On the other hand, we notice lower average values of diazinon concentrations in the amniotic fluid of mothers living near a public park, a field for growing wheat, a public road, or a nursery for growing roses and ornamental trees, compared to those living far from these places. We note higher average values of ethion concentrations in the amniotic fluid of mothers living near greenhouses ($p \leq 0.05$) compared to those living far from them. On the other hand, we note lower average values of ethion concentrations in the amniotic fluid of mothers living near a public park ($p \leq 0.01$) or a wheat-growing field ($p \leq 0.05$) compared to those living far from these places.

Discussion

Reviewing international studies, it was noted that the results of epidemiological studies are varied. Human exposure to chemical pesticides varies between countries due to age, sex, diet, environmental and occupational exposure, and the subjection of chemical pesticides to regulation and monitoring. However, what is common is that rural residents are exposed to chemical pesticides daily through inhalation during spraying seasons or through. Skin contact with pesticide residues is greater than that of city dwellers [23,24], as pregnant mothers in rural environments are exposed to chemical pesticides from neighboring fields [25].

Most epidemiological studies were concerned with titrating metabolites of chemical pesticides in amniotic fluid [6-8], but our

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study assayed chemical pesticide residues and aimed to clarify that the placental barrier is unable to protect the fetus from the arrival of diazinon and ethion residues, as we were able to detect diazinon in 78 (41%) and ethion. In 40 (21%) of the samples at levels higher than the detection threshold (0.02ppb), that is, the detection rate of diazinon is higher than that of ethion. In contrast, ethion was detected in meconium [26,27], while diazinon was detected in meconium [28], umbilical cord blood [29], and the urine of pregnant mothers [20].

We noted the absence of occupational exposure for mothers, but for fathers, a small sample of 14 (8.3%) of the total sample was found to be directly associated with diazinon and not with ethion levels. Occupational exposure hurts on neurodevelopment [30], and is associated with leukemia [31], but the population is exposed to chemical pesticides from the surrounding environment even if there is no professional activity in the field of pesticides [32]. It was noted in this study that current residence in rural areas and near greenhouses, fruit tree orchards, or vegetable growing lands were basic determinants of the high values of the average concentrations of diazinon in the amniotic fluid, while living near the greenhouses contributed to the high values of the average concentrations of ethion in the amniotic fluid. This reflects the widespread spread of diazinon in these places. It takes from hours to two weeks to decompose in water and soil, but successive use contributes to its accumulation in the soil [17]. As for ethion, it takes several months to decompose in the soil [21]. These results can be interpreted in light of Volatility in the air leads to the transfer of chemical pesticide residues to neighboring places [33].

Chemical pesticide residues do not contaminate the control site, but rather spread with the air [34], and thus inhaling pesticides spread in the air is an essential path to the human body unintentionally [35], where a study found that 43% of passersby are exposed to the same level of exposure as farmers [36], and several studies have shown the spread of pesticides of various types in all dust samples [37], and soil [38], and water, rainwater transports pesticides from the soil to groundwater, and therefore surface or groundwater runoff are two important paths for the movement of chemical pesticides that ultimately reach humans through skin contact with soil, water, and air [39,40].

It was observed that the average values of diazinon concentrations were lower in the amniotic fluid of pregnant mothers living near a public park, which is a small sample (1.1%) from city residents, or a field for growing wheat, and this sample is small (1.1%)2 from rural residents, and the level of concentrations is at the detection threshold. (0.02ppb). This indicates that the spread of this pesticide is low in these places or that it is rapidly metabolized.

The discrepancy in the levels of chemical pesticides detected in rural places is due to the factors of the density of agricultural land, vegetation cover, and the possibility of adhering to the instructions during the process of spraying chemical pesticides. The levels of chemical pesticides in greenhouses due to internal spraying differ from fields that are exposed to wind and volatilization with the air [41].

Conclusions and Recommendation

This study showed that diazinon and ethion residues reached the amniotic fluid, indicating direct exposure of the human fetus to these pesticides. High concentrations of diazinon levels were associated with rural areas near greenhouses and fields of fruit trees and vegetables

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