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Organochlorine pesticide residues in wheat from west Azerbaijan province, Iran

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ABSTRACT

The organochlorine pesticide contamination in food supply has caused serious threat to the human progeny. The present study was therefore conducted to evaluate the organochlorine contamination in wheat from West Azerbaijan province, Iran using gas chromatograph. The contamination level has been determined according to the European Community Directives. Forty samples of wheat were obtained from local farmers and wheat factories in this province. All the examined wheat samples were found to be contaminated by organochlorine pesticide residues of cis-chlordane and methoxychlor. Chlordane isomers, methoxychlor, DDT and its metabolites, aldrin, b HCH, heptachlor and lindane have been found to be the highest organochlorine pesticide residues. In some of these samples, various organochlorine pesticide residues have been determined to be higher than European Community maximum residual limits. The residues of aldrin in two samples, cis-chlordane in three samples, trans-chlordane in two samples, p-p'-DDD in one sample, o-p'-DDE in three samples, p-p'-DDE in one sample, β -endosulfan in one sample, heptachlor in two samples, methoxychlor in two samples were found to be in excess of EC maximum residue levels (MRLs). In conclusion most of the samples have been found to be contaminated with residues; also a control of organochlorine pesticide residues in wheat is necessary.

1. Introduction

Pesticides are used at different stages of cultivation and storage of crop (1). Pesticide use in commercial agriculture has led to an increase in farm productivity. Despite the wide ranging benefits of pesticide use in agriculture, several applications can be highly improper and undesirable substances in

consumer products are the result. These include improper use of pesticides in food, over-use of pesticides and harvest crops remained washed after application (2,3). Exposure to pesticides can occur via a number of pathways such as indirect (e.g., through food, drinking water, residential and occupational exposure) and farmers (oral, inhalation and dermal). However, the major concerns are from consumption of chemical pesticide for food crops (4). Pesticides have

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been linked to a wide range of human health risks, ranging from acute impacts, for example headache and nausea, to chronic impacts, for example cancer, reproductive harm and endocrine system disruption (5). In addition, incorrect applications of pesticides may cause harm to the environment, increased resistance in the target pest organisms and deleterious effects on non-target organisms. To ensure the safety of food for consumers and to protect consumer health, the monitoring of pesticide residues in food products must be pursued. Therefore, the levels of pesticide residues allowed in foodstuffs are legislatively controlled through setting maximum residue levels (MRLs). These MRLs limit the types and amount of pesticides that can be legally present on foods, as determined by various regulatory bodies which minimize consumer exposure to harmful or unnecessary intake of pesticides worldwide.

Wheat is a staple food for a large section of the population in many countries and is one of the most important essential foods for humans worldwide (6,7). With a production of 680 million tons in 2009 wheat contributes with approximately 30% to the world's average crop consumption (8). Furthermore, wheat is grown on more cultivable land than any other commercial crops (9) with an ever increasing demand due to a continuously growing global population. Since expansions of cultivable land are limited, cropping intensity and crop yield must be increased (10). It is widely cultivated in the Southwest, West and Northwest region in Iran.

In this study, we evaluated the presence of organochlorine pesticides in one of the most consumed cereal, i.e. wheat, in Iran. There are MRLs for some pesticides in orange that regulated by Institute of Standards and Industrial Research of Iran (ISIRI) (11). Wheat is a staple food for a larger section of the West Azerbaijan population in Iran. No reports have yet been published about organochlorine pesticides in wheat from Iran. In this study, the existences of organochlorine pesticide residues, which are undesirable substances in food according to the European Community Directives, were investigated. The objective of this work is therefore to characterize organochlorine pesticide in wheat from Iran.

2. Materials and Methods

2.1. Sample collection

In this study, 40 different types of wheat

samples were examined. These samples were collected from west Azerbaijan region. All samples were obtained from local farmers and wheat factories in the region in 2013, transported to the laboratory and stored at 4°C until being analyzed.

2.2. Extraction and clean-up

Sample preparation, extraction, clean-up, fractionation and analysis were performed according to the procedure described in US FDA Pesticide Analytical Manual (12). Fat and residues were removed from wheat by dissolving in petroleum ether and ethyl ether, followed by ethyl alcohol. The organic extract was washed with large quantities of water to remove co-extractives.

Extracted fat was carefully weighed to avoid overloading the capacity of the clean-up step. Pesticide residues were isolated from fat by partition between petroleum ether and acetonitrile. Most of the fat is retained in petroleum ether while residues partition into acetonitrile in proportion to their partitioning coefficient in that system. In the subsequent step, residues in acetonitrile were partitioned back into petroleum ether when added water reduces their solubility in acetonitrile. Residues in solution were separated from sample co-extractives on a column of florisil adsorbent; eluants of increasing polarity sequentially removed residues from the column.

2.3. Gas chromatographic (GC) analysis of organochlorine residues

The pesticide analyzed on a HP (Hewlett Packard) Agilent 6890 N model gas chromatograph (GC), equipped with an electron capture detector (ECD) and fitted with a DB-5MS capillary column (30 m, 0.25 mm i.d. and 0.25 μ m). Injector and detector temperatures were 270 and 320 °C, respectively. Column temperature program was 80 °C for 1 min then increasing at 30 °C/min up to 180 °C, then increasing at 3 °C/min up to 205 °C where it was maintained for 4 min and then increased at 20 °C/min up to 290 °C where it was maintained for 2 min. Carrier gas was helium (2 ml/min). The injection was carried out split less and the injection volume was 1 μ l. Identification of pesticide was carried out by comparing sample peak relative retention times with those obtained for standards from Dr. Ehrenstorfer (Augsburg, Germany). The area of the corresponding peak in the sample

was compared and a little with that of the standard

3. Results

The levels of pesticide residues in wheat samples collected from West Azerbaijan province, Iran and compared with European Community maximum residual limits are presented in Table 1.

Table 1 presents that all of the wheat samples considered in the present study contained detectable residues of various chlorinated pesticides and some of these residue levels are higher than that of the EC MRLs (European Council Directives). The maximum and minimum amounts obtained for pesticides that had highest amount respectively trans-chlordane (0.00 - 21.1 µg/kg), methoxychlor (0.2 - 15.4 µg/kg), o, p'-DDD (0.1 - 12.2 µg/kg) and β- endosulfan (0-1.6 µg/kg) was the lowest. Chlordane isomers, methoxychlor, DDT and its metabolites, aldrin, b HCH, heptachlor and lindane were found to be the highest organochlorine

pesticide residues. The residues of aldrin in two samples, cis-chlordane in three samples, trans-Chlordane in two samples, p-p'-DDD in one sample, o-p'-DDE in three samples, p-p'-DDE in one sample, β-Endosulfan in one sample, heptachlor in two samples, methoxychlor in two samples were found to be in excess of EC MRLs.

4. Discussion

Pesticides such as insecticides have a longer and more noteworthy history, perhaps because the number of insects labeled "pests" greatly exceeds the number of all other plant and animal "pests" combined. Hence, this article focused on the use of agricultural insecticides. In studies conducted by other researchers, including Yentur et al (13) it has been traced quintozone (Pentachloronitrobenzene) and lindane in the samples of cracked wheat and p, p'-DDT, p, p'-DDE in few samples obtained from markets in Ankara, Turkey. In these samples, p, p'-DDD had not been observed. According to the

Table 1. Levels of pesticide residues in examined wheat samples and compared the pesticide residues with European Council Directives

Pesticide	Frequency of detection (%)	Samples exceeded limit with European Council Directives	Samples contaminated	Mean concentration (µg /kg)	Minimum-maximum (µg /kg)
Aldrin	77	2	31	0.09	0.0 0- 14.5
Dieldrin	70		28	0.05	0.00 - 3.2
Aldrin and dieldrin				1.6	0.10 - 12.1
cis-Chlordane	87	3	35	1.3	0.20 - 13.5
trans-Chlordane	80	2	32	1.7	0.00 - 21.1
Oxy-chlordane	75	6	30	15.4	0.10 - 16.41
∑ Chlordane				12.6	0.40 - 19.47
o, p'-DDD	80	-	32	1.1	0.10 - 12.2
p, p'-DDD	82	1	33	0.8	0.20 - 4.3
o, p'-DDE	87	3	35	1.6	0.00 - 7.8
p, p'-DDE	75	1	30	0.3	0.10 - 2.4
o, p'-DDT	67	-	27	0.7	0.00 - 3.2
p, p'-DDT	72	2	29	0.8	0.10 - 5.1
∑ DDT				5.6	0.00 - 41.2
α-Endosulfan	70	-	28	0.	0.20 - 3.4
β-Endosulfan	80	1	32	0.1	0.00 - 1.6
∑ Endosulfan				0.8	0.10 - 5.4
α-HCH	67	-	27	0.4	0.00 - 4.8
β-HCH	50	-	20	1.1	0.10 - 9.2
γ-HCH (lindane)	75	-	30	0.2	0.00 - 2.8
δ-HCH	65	-	26	0.5	0.20 - 6.9
τ-HCH	39	-	17	0.2	0.00 - 3.5
∑ HCH (τ-HCH except)				2.1	0.00 - 14.4
Heptachlor	75	2	30	0.6	0.10 - 6.5
cis-Heptachlor epoxide	62	-	25	0.3	0.00 - 4.1
trans-Heptachlor epoxide	55	-	22	0.9	0.20 - 7.3
Heptachlor and H. epoxide				2.5	0.00 - 11.5
Endrin	47	-	19	0.1	0.1 - 2.1
Hexachlorobenzene	72	-	29	1.3	0.2 - 6.8
Methoxychlor	100	2	40	1.6	0.2 - 15.4

o, p'-DDD= o, p'-Dichlorodiphenyltrichloroethane, p, p'-DDD= p, p'-Dichlorodiphenyldichloroethane, o, p'-DDE= o, p'-Dichlorodiphenyldichloroethylene, p, p'-DDE= p, p'-Dichlorodiphenyldichloroethylene, o, p'-DDT= o, p'-Dichlorodiphenyltrichloroethane, p, p'-DDT= p, p'-Dichlorodiphenyltrichloroethane, ∑ DDT=∑ Dichlorodiphenyltrichloroethane, α-HCH= Alpha-Hexachlorocyclohexane, β-HCH= Beta-Hexachlorocyclohexane, δ-HCH=Delta-Hexachlorocyclohexane, τ-HCH=Gamma-Hexachlorocyclohexane

percentages of residues observed in the cracked wheat samples it was determined that the 64% of the samples were containing pentachloronitrobenzene, 20% lindane and 4% contained DDT, while the remaining 12% did not contain any organochlorine pesticide residue. The organochlorine pesticide residue levels have not found to be above the maximum residue limits (Codex Alimentarius FAO tolerance limits) in all samples (13). Similarly, in our study, lindane p, p'-DDT, p, p'-DDE were observed in wheat samples but p, p'-DDT and p, p'-DDE were higher than the EC maximum residue limits in two and one samples.

The use of organochlorine pesticides has been restricted in world except malaria region or even banned throughout the EC. Nevertheless, they are still present in animal tissues. Use of lindane (c-HCH) and DDT had been banned in Turkey in 1985 (14). In the present study, residues of the lindane, o, p'-DDT and p, p'-DDT were observed in 30, 27 and 29 of 40 samples, respectively, but these residues were two samples of p, p'-DDT higher than the EC MRLs. In spite of the ban, the existence of lindane and DDT in the wheat samples confirms that these residues tend to accumulate in food chain. Bakore et al. (15) reported that all of the wheat samples were found to be contaminated with various organochlorine pesticide residues of DDT and its metabolites, HCH and its isomers, heptachlor and epoxide and aldrin in India. Similarly, in the present study, most of wheat samples were contaminated with these residues. Bakore et al. (15) have determined that aldrin and heptachlor have been found to be higher than that of the practical limit prescribed by WHO (16), whereas DDT were much below the tolerance limit in the summer, winter and rainy seasons. HCH during winter and rainy seasons have been found to be higher than that of the tolerance limits. Guler et al. (7) reported that most of wheat samples were contaminated with these residues of the lindane, o, p'-DDT and p, p'-DDT have been observed in 31, 35 and 30 of 40 samples but these residues were not higher than the EC MRLs. In our study, heptachlor, DDT and HCH have been found to be below the EC MRLs but aldrin and heptachlor were higher than these limits in two samples.

Toteja et al. (17) have investigated residues of DDT and different isomers of HCH in wheat grain/flour by gas-liquid chromatography

from urban and rural areas in different geographical regions of India. In their study, residues of DDT (p, p'-DDT, o, p'-DDT, p, p'-DDE) have been detected in 59.4% of 1080 samples of wheat grain, in 78.2% of 632 samples of wheat flour and different isomers of HCH were present in about 45-80% of the samples of wheat grain/flour, and 19 samples of wheat grain constituting only 1.7% of the total samples analyzed were found to be contaminated with DDT above maximum residue limit recommended by the Codex (18). In our study, HCH and DDT isomers have been observed in 39-75% and 72-87% of 40 wheat samples, respectively, but p, p'-DDD, o, p'-DDE, p, p'-DDE and p, p'-DDT were higher than the EC MRLs in one, three, one and two samples. Guler et al. (7) reported that HCH and DDT isomers have been observed in 42-91% and 83-97% of 36 wheat samples, respectively. But none of the samples exceeded the EC MRLs.

Rekha et al. (19) have stated that endosulfan residues of organochlorine in wheat have been reported in all market samples in India but pesticide residues were well below the MRLs. The results were alike in the present study, 28 and 32 of 40 samples were contaminated with alpha and beta endosulfan, respectively, and beta endosulfan was higher than these limits in one sample. Skrbic (20) has investigated organochlorine and organophosphate pesticide residues in wheat varieties from Serbia. The author has assessed levels of 20 organochlorine and 15 organophosphate pesticides in wheat. The ranges of mean values for organochlorine residues have been reported to be 0.032-0.047 $\mu\text{g g}^{-1}$ for β HCH; .0028- 0.041 $\mu\text{g g}^{-1}$ for c-HCH; <0.001-0.061 $\mu\text{g g}^{-1}$ for aldrin; 0.005-.0132 $\mu\text{g g}^{-1}$ for dieldrin; 0.015-0.111 $\mu\text{g g}^{-1}$ endrin ketone; and < 0.001-0.077 $\mu\text{g g}^{-1}$ for Endrin aldehyde. In our study, the mean values for organochlorine residues were 0.011, 0.002, 0.009, 0.005 and 0.001 $\mu\text{g kg}^{-1}$ for β HCH, γ -HCH, aldrin, dieldrin and endrin, respectively.

Saeed et al. (21) have investigated chlorinated pesticide residues in the total diet of Kuwait. They have found that chlorpyrifos-methyl presents in most of the samples containing wheat or wheat flour. The levels in wheat flour (both white and brown) ranged from 0.037 to 0.72 $\mu\text{g g}^{-1}$ but imported wheat flour has not contained any detectable residue levels. In their study, non-wheat cereals and their products have not contained any

detectable residues of chlorinated pesticides. In our study, all the wheat samples have been found to be contaminated with organochlorine pesticide residues of methoxychlor. Chlordane isomers, methoxychlor, DDT and its metabolites, aldrin, b HCH, heptachlorand lindane have been observed as the highest organochlorine pesticide residues. Organophosphorus pesticide residues have also been investigated in cereals in different countries for health risk. Maver et al. (22) have analyzed organophosphorus pesticide residues of many commodities including cereals in Slovenia. Furthermore, Bai et al. (23) have investigated the organophosphorus pesticide residues in market food including cereals in China. They have found that organophosphorus residue levels were below MRLs in the cereals.

5. Conclusion

The amount of organochlorine pesticides detected in wheat from West Azerbaijan, Iran has shown the contamination of environment due to the use of pesticides by farmers. Although most of the pesticide residues are below the EC MRLs, some residues exceed EC MRLs. Therefore, it may pose a threat to human health. Since most of the samples have been found to be contaminated with residues, a control of organochlorine pesticide residues in wheat is necessary. Further research is needed to determine the pesticide residues in various foods made from wheat for human health perspective. Based on this decline, it is concluded that tighter measures, such as education of farmers, control of the sale of pesticides and improvement of the organic agriculture and implementation of integrated pest management methods, is necessary for the reduction of pesticide residues in foods and prevention of exposure to pesticides. Contamination levels of these residues have to be considered as a serious public health problem according to European Community regulations for aldrin, trans-chlordane, oxy-chlordane and methoxychlor.

Conflict of Interests

Authors have no conflict of interest.

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