Effect of Refining Processes on Magnitude and Nature of Fenitrothion and Pirimiphos-Methyl Residues in Maize Oil and Bioavailability of their Cake Residues on Rats*

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MAIZE seeds obtained from ^{14}C-fenitrothion or ^{14}C-pirimiphos-methyl treated maize plants contained about 0.12 % and 0.16 % of the originally applied radioactivity, respectively. The concentration of ^{14}C-fenitrothion residues and ^{14}C-pirimiphos-methyl residues in crude oil (hexane extract) ranged from 2.8-3.0 ppm, methanol solubles 2.3 ppm and in seed cake 1.2-1.4 ppm. Commercial processing procedures led to a gradual decrease in the total amount of ^{14}C-residues in oils with aged residues. The refined oil contained only about 13-19% of the radioactivity originally present. The deodorization and alkali treatment are the most effective steps as they removed over 50 % of the residues. Chromatographic analysis of refined oil revealed the presence of fenitrothion, 3-methyl-4-nitrophenol as main products beside smaller amounts of fenitrooxon and desmethyl fenitrothion. Whereas, pirimiphos-methyl and 2-diethyl-2-amino-4-hydroxy-6-methylpyrimidine were detected as principal compounds together with minor amounts of 2-ethyl-amino-4-hydroxy-6-methylpyrimidine and 2-amino-4-hydroxy-6-methyl-pyrimidine.

On feeding rats with the extracted seeds (cake) for three days, the bound residues were found to be considerably bioavailable. A substantial amount of administrated bound ^{14}C-residues was eliminated via respiration (50%) for ^{14}C-fenitrothion and excreted urine (47%) for ^{14}C-pirimiphos-methyl. While feces contained only 10-12% and about 22-26% of the radioactive residues were distributed among various organs of experimental animals.

Keywords: ^{14}C-Fenitrothion, ^{14}C-Pirimiphos-methyl, Maize seed oil, Insecticide residues, Refining processes and Bioavailability.

Pesticides are used globally for the protection of food, fiber, human health and comfort. Food safety is an area of growing worldwide concern on account of its direct bearing on human health. The presence of harmful pesticide residues in food and seed oils has caused a great concern among the consumers[1]. Several

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investigations suggest that most organophosphorus pesticides in edible oils can be reduced considerably by chemical refining processes [2-6].

Two main organophosphorus insecticides are recommended for use on maize namely fenitrothion and pirimiphos-methyl against aphids according to the guide lines of the Egyptian Ministry of Agriculture (2007)[7].

Fenitrothion [O, O-dimethyl-O-(3-methyl-4-nitrophenyl) phosph-orothioate, I] is marketed under the different trade names Sumithion, Novathion and Metathion. It is a broad-spectrum organophosphorus insecticide, non-systemic and non-persistent [8, 9] and moderately toxic to mammals[10, 11]. It is also used in agriculture to control insects on rice, wheat, barley, soybeans, cereals, fruits, coffee[9] and as a grain protectant and it finds use in horticulture, forestry, domestic and as a residual spray in homes for malaria control and public health applications[10,12].

Pirimiphos-methyl (actellic) [O,O-dimethyl-O-(2-diethyl-amino-6-methylpy-rimidine-4-yl)] is a fast-acting broad spectrum insecticide with both contact and fumigant action and is a more effective protectant than other organophosphorus insecticides[13]. It possesses a low order mammalian toxicity[14]. It degrades slowly in stored grains under most storage condition[15] and shows activity against a wide variety of insects including ants, aphids, beetles, caterpillars, cockroaches, fleas, flies, mites, mosquitoes, moths, and thrips[16].

Maize (*Zea mays* L.) is an economic and popular plant in Egypt, which is grown in more than one season. It is a high source of carbohydrate and good quality oil for human food, livestock feed and raw material for industrial processing [17]. Recently, corn oil production increased markedly and is refined into high quality oil for the food industry such as sweetener and starch[18].

The aim of the present work was to study the effect of simulated commercial processing procedures on the elimination of the insecticide and its residues from maize seed oil obtained from plants treated with ^{14}C-fenitrothion and/or ^{14}C-pirimiphos-methyl as well as the bioavailability to rat of their cake residues.

Materials and Methods

Chemicals

   Synthesis of ^{14}C-fenitrothion [I]
^{14}C-Fenitrothion-labelled at carbon atom of methyl groups-was synthesized by a single vesseled reaction in our laboratory (Scheme 1). This method involved the condensation of 0.007 mole of ^{14}C-methanol in toluene to 0.003 mole of thiophosphorylchloride in presence of triethylamine (0.006) mole and the mixture was stirred at 5-10°C for 30 min, then filtering the product to remove the separated triethylamine hydrochloride and the filtrate was evaporated under vacuum. The obtained (O, O-dimethylthiophosphoryl chloride) in toluene was allowed to react with 0.003 mole of potassium salt of 3-methyl-4-nitrophenol while stirring for 30 min at room temperature. This mixture was then refluxed for 2-3 hr at 60-80°C

cooled followed by addition of cold water and the organic layer was separated, dried over anhydrous sodium sulphate. Under reduced pressure the solvent was evaporated to give yellow-brown liquid (b.p 140-145°C, 0.1 mm) yield 70-75% (Scheme 1). The product was purified by preparative thin layer chromatography (TLC) (Silica gel 60 F_{254} pre-coated plates 20×20 cm layer thickness 0.5 mm, Merck, Germany). The prepared ^{14}C-fenitrothion had a specific activity 2.96 MBq/mg and a radiometric purity 98 %[19] (Scheme 1).
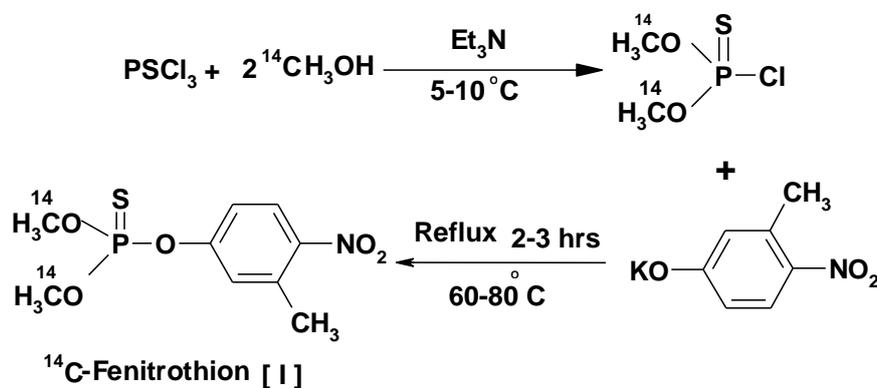
Scheme 1. Synthesis of ^{14}C-fenitrothion .

Pure fenitrothion (non-labelled) and its main degradation products were synthesized in our laboratory for comparison purposes[20-22] (Fig 1).

Fenitrothion [I]

Fenitrooxon [II]

Desmethylfenitrothion [III]
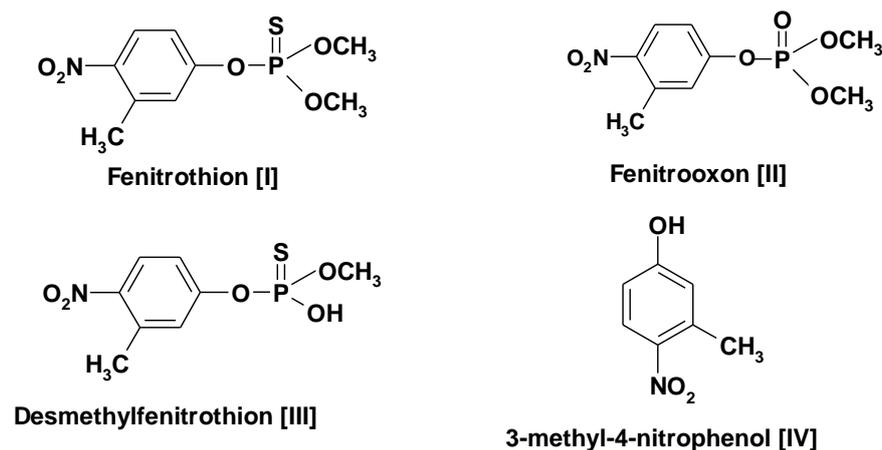
3-methyl-4-nitrophenol [IV]

Fig. 1. Fenitrothion and its main degradation products in maize seed oil .

Synthesis of non-labelled pirimiphos-methyl

A mixture of 0.05 mole of 2-diethylamino-4-hydroxy-6-mehyl pyrimidine and 0.05 mole of anhydrous potassium carbonate in dry benzene was refluxed for 3 hr and the obtained water was removed by azeotropic distillation. The resulting potassium salt was dissolved in 100 ml of methyl ethyl ketone and stirred at 60°C. O,O-Dimethyl chlorophosphorothioate (0.06 mole) was added dropwise to the pervious solution and the whole mixture was refluxed at 94-95°C for two hours to give 0.18 mole of pirimiphos-methyl as colourless liquid (b.p= 128-132 °C, 0.04 mm) yield 85-90%, purity about 95 %[23]. Its main degradation products were synthesized in our laboratory[24-26] for comparison studies as shown in Scheme 2 and Fig. 2.

[14]C-Pirimiphos-methyl labelled at position 2 of the pyrimidine ring, was purchased from Izinta Isotope Trading Inerprise of the Institute of Isotopes, Budapest, Hungary. It had a specific activity 1.98 MBq/mg. Radiometric purity was over 97%.
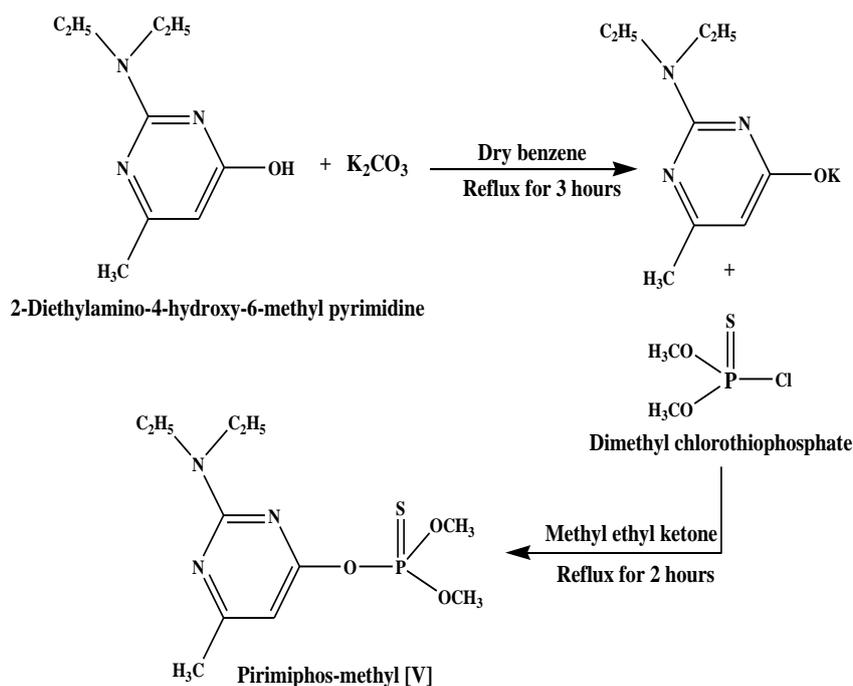
Scheme 2. Synthesis of non-labelled pirimiphos-methyl .
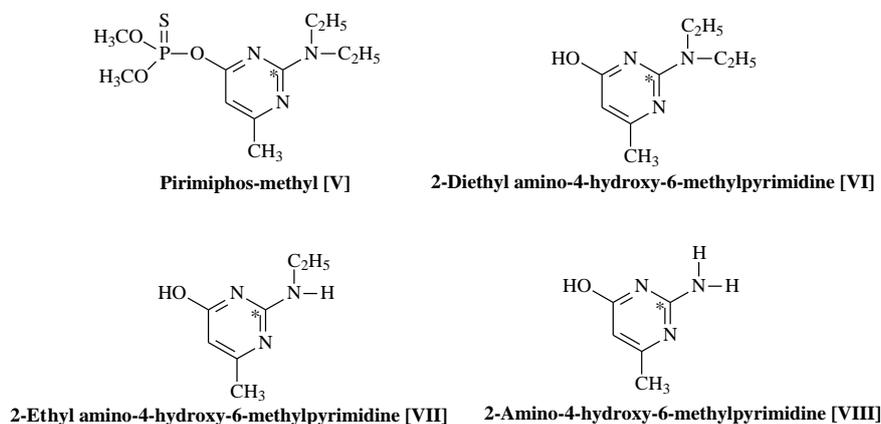
Fig. 2. ^{14}C-Pirimiphos-methyl and its main degradation products in maize seed oil.

Seeds

Pesticide-free Zea-mays seeds (Var. Giza.1 hybrid) were obtained from Agricultural Research Centre (Cairo). The seeds were cleaned from any dockage and impurities before cultivation.

Field experiment

Sound whole seeds of maize were cultivated in alluvial soil under normal field conditions in controlled and isolated field area (2 seasons). Irrigation, fertilization and soil management were carried out as practiced in the field according to the guide lines of the Egyptian Ministry of Agriculture (2007) [7]. Shortly before blooming stage, leaves of plants were treated twice, 23 days apart, with ^{14}C-fenitrothion or ^{14}C-pirimiphos-mehyl at a dose of 6 mg/plant which is equivalent to 0.8 μCi and 1.0 μCi, respectively. At maturity, seeds were collected and dried for preparation of oil and cake.

Extraction of the oil from seeds

Maize seeds (400g) were crushed and soxhlet extracted with n-hexane for 6hr and the maize oil obtained after evaporation of hexane under reduced pressure. The remaining residues were further extracted with methanol for 6 hr to isolate the polar ^{14}C-residues [27]. Both extracts and cake were used for determination of their radioactivity.

Simulated commercial processing procedures

Removal of pesticide residues from oils was carried out through several refining processes .

Alkali refining (Neutralization)

The crude oil sample (20 g) was stirred vigorously with 2N sodium hydroxide solution for about 30 min at 27 ˚C. The mixture was then centrifuged to remove the soap and excess alkali, washed several times with warm water until pH 7.0 and centrifuged at 3000 rpm for 10 min to obtain the clear oil. This process removed free fatty acids and other acidic materials from oil.

Bleaching

The alkali refined oil (Neutralized oil) was treated with 0.5% (by weight) of a factory grade Fuller's earth (Tonsil), the mixture was stirred vigorously at 80-100˚C on an oil bath for 20 min and then centrifuged. The bleached oil was filtered.

Winterization

The clear dry oil was winterized at 5 ˚C for three days and the separated saturated glycerides were removed by centrifugation.

Deodorization

Deodorization is the last major processing step in the refining of edible oils and is responsible for removing undesirable ingredients. Superheated steam was passed into the heated winterized oil at about 200-220˚C under reduced pressure for three hours to remove steam distillable materials including odours.

Aliquots of oil, after each refining process, were analyzed for their radioactivity.

Bioavailability of bound residues to rats

Two adult male albino rats weighting 180-210 g were obtained from the Animal House at National Research Centre, Dokki, Cairo. The rats were placed individually in glass metabolism cages which allowed separation and collection of feces, urine and expired air. The rats were conditioned for 2 days to a daily standard diet, and then were left for 24 hr without feed, after which they were fed a known amount of extracted maize seeds (21µg pirimiphos-methyl/rat; 84µg fenitrothion/rat) for 3 days. To make the feed palatable, the extracted maize seeds were mixed thoroughly with an equal amount of white cheese and the paste was left to dry. The expired $^{14}CO_2$ was trapped in 10% NaOH solution. Urine, feces and $^{14}CO_2$ were collected separately for 3 days, and assayed for their radioactivity. After 72 hr, each rat was lightly anaesthetized with ether and blood removed from the pumping heart. The rat was then sacrificed by decapitation and samples of selected organs such as liver, kidney, fat, blood and heart were analyzed for total radiocarbon by combustion in Harvey Biological Oxidizer (Model OX-600) and followed by Liquid Scintillation Counting (LSC).

Isolation and characterization of radioactive residues

Samples of oil, after each refining process, were partitioned between acetonitrile and hexane to remove the oil. The radioactive residues were almost completely retained in the acetonitrile layer. Analysis of crude and refined oil extracts were achieved by thin layer chromatography (TLC). Urine was extracted with chloroform (chloroform I) and the aqueous layer was then acidified with 2N HCl and heated for 2 hr on a water bath and reextracted with chloroform (chloroform II) to obtain the conjugated metabolites, Aliquots from both chloroform I, II were analyzed by TLC on silica gel plates using suitable systems.

Thin Layer Chromatographic (TLC) analysis

Residues were characterized by TLC on silica gel plates (20×20 cm; 0.25 mm thickness) with fluorescent indicator (Kiesel gel 60 F_{254}, Merck, Germany) using suitable systems for fenitrothion and pirimiphos-methyl. Authentic samples were run alongside as references for identification and the plates were seen under UV-light at 254 nm and spots were made visible by spraying the plates with Vanillin/Sulphuric acid (2.5%).

High Performance Liquid Chromatography analysis (HPLC)

Analysis by HPLC was afforded on a Helwet Packard (1100 U.S.A) equipped with gradient system pumping, solvent programmer, the stainless steel column (125×4 mm I.D) was packed with 5 μm ODS-Hypersil, variable-wavelength UV detector operated at 254 nm. The mobile phase consisted of a mixture of acetonitrile: water (1:1 by volume).

Radiometric measurements

Radioactivity in oil (acetonitrile extracts), methanol extracts, urine and other liquid samples were measured directly by LSC (Packard Model RI-CARB 2300 TR) in vials using a dioxane-based scintillation cocktail[28].

Solid samples, ground maize seeds, cake, feces and animal tissues were assayed for radioactivity by combusting a definite weight (100 mg) in a Harvey Biological Oxidizer (Model OX-600), followed by LSC. The internal standard ([14]C n-hexadecane 0.2 μCi/ml=74 KBq/ml) technique was used for quench correction. Thin layer plates were divided in 1 cm increments, scraped into vials, eluted with methanol, covered with scintillator and counted.

Results and Discussion

Field studies with aged residues

The persistent insecticides are beneficial for controlling pests for extended periods however their residues in consumable parts of the crops may be harmful to the consumers [29].

[14]C-Residues in seeds, oil and cake

Under local agricultural conditions, the aged insecticide residues were detected at harvest in mature seeds after foliar application of the used organophosphorus insecticides. Maize seeds obtained from ^{14}C-fenitrothion or ^{14}C-pirimiphos-methyl treated maize plants contained about 0.12 % and 0.16 % of the originally applied radioactivity, respectively. Zayed *et al.* reported that, 0.37 % and 0.11 % of ^{14}C-residues were detected in soybeans and cotton seeds

which were obtained after spraying soybean plant with pirimiphos-methyl [30] and cotton plant with malathion [27] under similar conditions.

Table 1 shows that ^{14}C-residues associated with crude oil (hexane extract) are 10-12% and those in methanolic extract 5-7% for pirimiphos-methyl and fenitrothion, respectively. While the ^{14}C-residues in cake accounted for 63 % of total residues of the two insecticides inside the seeds.

TABLE 1. Distribution of ^{14}C-fenitrothion and ^{14}C-pirimiphos-methyl residues in 100g of maize seeds.

Fractions	^{14}C-fenitrothion			^{14}C-pirimiphos-methyl		
	Weight (g)	Residues		Weight (g)	Residues	
		(µg)	%		(µg)	%
Maize seeds	100	160 ±1.3	100	100	180 ± 1.7	100
Hexane extract (oil)	7	19.5 ± 0.2	12	6	18 ± 0.6	10
Methanol extract	5	11.5 ± 0.2	7	4	9.0 ± 0.4	5
Cake	82	101 ±1.5	63	85	117 ± 1.0	65
Recovery	94	132	82	95	144	80

Data are means of three replicates.
Total ^{14}C-in seeds = 100 %.

Effect of refining processes

The effect of individual simulated commercial processing procedures on ^{14}C-residues is shown in Table 2. These processes led to a gradual decrease in the total amount of ^{14}C-residues in oils with aged residues which are attributed to alkali hydrolysis, effect of adsorption, effect of heat and/or evolution of $^{14}CO_2$ gas. The deodorization process proved to be the most effective step in reducing of ^{14}C-fenitrothion residues as it eliminated 31 % of the residues originally present in crude oil, while the alkali treatment (neutralization), bleaching and winterization processes removed about 27 %, 13 % and 10 % of these residues, respectively.

TABLE 2. Effect of commercial processing procedures on magnitude of ^{14}C-insecticide residues in maize seeds oil.

Treatment	^{14}C-fenitrothion		^{14}C-pirimiphos-methyl	
	^{14}C-Residues in oil with aged residues		^{14}C-Residues in oil with aged residues	
	ppm	% Reduction	ppm	% Reduction
Crude oil	2.80 ± 0.15	-------	3.00 ± 0.11	-------
Alkali refining	0.75 ± 0.17	27	0.78 ± 0.10	26
Bleaching	0.36 ± 0.10	13	0.63 ± 0.15	21
Winterization	0.28 ± 0.20	10	0.39 ± 0.01	13
Deodorization	0.87± 0.10	31	0.81 ± 0.02	27
Total Reduction	--------	81	--------	87

ppm = µg insecticide/gm oil.
Data are means of three replicates.

Both deodorization and neutralization processes seem to cause maximum reduction in the amount of ^{14}C-pirimiphos-methyl residues, since they eliminated about 53%, whereas the bleaching and winterization processes reduced 34 % of the residues originally present in crude oil (Table 2). It is worthy to mention that, on refining soybeans oil which is contaminated with some organophosphorus insecticides such as malathion, dichlorvos and chlorpyrifos residues, deodorizing process is the most effective step in removing pesticide residues due to combination of decomposition and volatilization [31]. Recently, many authors also reported that, simazie residues in olive oil and prothiofos residues in canola oil are highly eliminated during deodorizing step [32, 33].

The total refining processes reduced about 81-87% of the total ^{14}C-residues as shown in Table 2. Zayed et al., [2, 30] showed that the amount of both ^{14}C-carbofuran and ^{14}C-pirimiphos-methyl residues in soybeans oil decreased to 16 % and 25 %, respectively through the refining processes. It is also found that the complete refined sunflower oil obtained from treated sunflower plants lost about 64 % of the total ^{14}C-zineb residues in crude oil [34] and 69 % of ^{14}C-prothiofos residues in canola oil [33] **.**

Identification and characterization of ^{14}C-residues in crude and refined oil
Chromatographic analysis (TLC and HPLC) of the crude oil in case of ^{14}C-fenitrothion aged residues, shows mainly the presence of the parent compound fenitrothion [I] and three minor degradation products were identified as fenitrooxon [II], desmethyl fenitrothion [III] and 3-methyl-4-nitrophenol [IV]. R_f and R_t values of these metabolites are shown in Table 3. The amount of each minor compound did not exceed than 10 % of the total residues. Generally, the residues in all steps of refined oil were identified as I and IV and traces amounts of II and III which were almost completely eliminated through oil processing (Table 3 & Fig 1). Compound IV can be detected only by coloring reagent. The methanol extractable residues consisted of the hydrophilic compounds as free metabolites dimethyl phosphorothioate and dimethyl phosphate in addition to conjugated metabolites which upon hydrolysis with 2N HCl at 100 °C and then extraction with chloroform gave compounds III, IV.

M. Farghaly *et al*.

TABLE 3. R_f, R_t values and percent of fenitrothion and its degradation products in maize oil after subjecting to different commercial processing procedures.

Compound	R_f values			R_t values	Relative percentage %				
	Sys. 1	Sys. 2	Sys. 3		A	B	C	D	E
Fenitrothion [I]	0.90	0.75	0.90	9.32	70	55	45	35	15
Fenitrooxon [II]	0.82	0.44	0.74	2.47	10	5	3	Trace	-------
Desmethyl fenitrothion[III]	0.72	0.35	0.78	0.96	5	3	2	--------	--------
3-methyl-4-nitrophenol [IV]	0.80	0.38	0.76	2.24	Detected by color[a]				

A= Crude oil, B=Alkali refined oil, C=Bleached oil, D= Winterized oil, E= Deodorized oil

System 1= Toluene: Ethyl acetate: Acetic acid (5:7:1)

System 2= Toluene: Acetic acid (7:1)

System 3= Acetonitrile: Water: Ammonia (85:14:1)

a = Coloring reagent: Vanillin/sulphuric acid

The chromatographic analysis (TLC and HPLC) of the crude oil and refined oil are shown in Table 4. They contained pirimiphos-methyl [V] and 2-diethyl-2-amino-4-hydroxy-6-methylpyrimidine [VI] as major degradation products, in addition to smaller amounts of 2-ethyl-2-amino-4-hydroxy-6-methylpyrimidine [VII] and 2-amino-4-hydroxy-6-methylpyrimidine [VIII]. The main residues in the totally refined oil were only identified as V and VI compounds (Table 4). While the chromatographic analysis of methanol revealed the presence of V and VI as free metabolites beside the hydrophilic compounds in addition to conjugated metabolites which upon acid hydrolysis were identified as VI and VII, VIII compounds (Fig. 2). The formation of these products indicates that fenitrothion is degraded via oxidation of P=S to P=O to yield oxon analogue [II], cleavage of the P-O-alkyl to yield desmethyl compound [III] and hydrolysis to yield its phenol [IV].While, pirimiphos-methyl is degraded via hydrolysis to give [VI] in addition to cleavage of the N-alkyl groups to give VII and VIII.

TABLE 4. R_f, R_t values and percent of pirimiphos-methyl and its degradation products in maize oil after subjecting to different commercial processing procedures.

Compound	R_f values			R_t values	Relative percentage %				
	Sys. 1	Sys. 2	Sys. 3		A	B	C	D	E
Pirimiphos-methyl [V]	0.95	0.96	0.90	6.96	65	45	34	28	10
2-Diethylamino-4-hydroxy-6-methylpyrimidine [VI]	0.80	0.72	0.12	6.58	20	35	40	45	54
2-Ethylamino-4-hydroxy-6-methylpyrimidine [VII]	0.61	0.45	0.0		5	4	2	Trace	-------
2-Amino-4-hydroxy-6-methylpyrimidine [VIII]	0.30	0.21	0.0		5	3	2	Trace	-------

A= Crude oil, B=Alkali refined oil, C=Bleached oil, D= Winterized oil, E= Deodorized oil
System 1= Chloroform: Methanol (17:3)
System 2= Diethyl ether: Hexane: Methanol: Ammonia (16:4:4:1)
System 3= Cyclohexane: Ethylacetate (1:1)

It is worthy to mention that, the removal efficiency during the refining processes seems to depend upon the nature of residues present. Data obtained emphasize the importance of studying the effect of oil refinement on reduction and/or elimination of aged pesticide residues in edible oils.

Bioavailability to rats
Elimination and distribution of ^{14}C-fenitrothion and/or ^{14}C-pirimiphos-methyl, after feeding rats with the extracted maize seeds (cake) for three days are shown in Table 5. In case of ^{14}C-fenitrothion, the major portion of radioactivity was eliminated via expired air (50 %), feces (10%) and urine (5%). Appreciable amount of ^{14}C-residues were also detected in liver, kidney, blood, fat and heart (22 %).

TABLE 5. Elimination and distribution of bound ^{14}C-fenitrothion and ^{14}C-pirimiphos-methyl residues on extracted maize seeds (cake) fed to rats for three days.

Sample	^{14}C-fenitrothion		^{14}C-pirimiphos-methyl	
	Insecticide equivalent[(1)] (µg)	Percentage of administrated dose (%)	Insecticide equivalent[(2)] (µg)	Percentage of administrated dose (%)
Carbon dioxide	42.0 ± 1.30	50	1.50 ± 0.26	7
Urine	4.5 ± 0.26	5	9.90 ± 0.04	47
Feces	8.5 ± 0.20	10	2.50 ± 0.30	12
Blood	6.7 ± 0.20	8	2.00 ± 0.20	9.5
Liver	5.0 ± 0.36	6	1.25 ± 0.03	6
Fat	4.0 ± 0.26	5	0.55 ± 0.15	2.5
Kidney	1.7 ± 0.15	2	0.95 ± 0.02	4.5
Heart	0.9 ± 0.05	1	0.85 ± 0.04	4
Recovery	73.3	87	19.5	92.5

(1) Administrated dose = 100% (84 µg equivalent/rat).
(2) Administrated dose = 100% (21 µg equivalent/rat).
    Data are means of three replicates.

In case of ¹⁴C-pirimiphos-methyl, the total excretion represented about 66% of the originally consumed dose. The main excretion route was the urine (47%) while the radioactivity in expired air and feces represented 7 % and 12 %, respectively. About 26% of the administrated radioactivity was detected among various organs of the rat. This indicates that the main routes of elimination are via respiration and urine for fenitrothion and pirimiphos-methyl, respectively.

The highest percent was observed in blood 8-9.5%, for both insecticides as shown in Table 5. The total recovery of ¹⁴C-activity amounted to 87 % and 92.5 % of the administrated dose for fenitrothion and pirimiphos-methyl, respectively. This indicates a high percentage of release of bound residues followed by distribution of these residues or their degradation products among tissues of experimental animals.

In case of fenitrothion, chromatographic analysis of urine showed the presence of the hydrophilic compounds only in free and conjugated form. This result agrees with some previous reports[35-37] which noted that, in adult rats, the insecticide is metabolized to phosphorothioate and dimethylphosphate compounds. While compound VI represented the main substance in collected urine in free and conjugated form besidetraces of compounds VII and VIII in case of pirimiphos-methyl. These findings are in accordance with other recent studies[38].

Bound ¹⁴C-fenitrothion and ¹⁴C-pirimiphos-methyl in maize seeds proved to be highly bioavailable to rats. The data obtained are in line with many recent studies which indicate a moderate to high bioavailability of soybean bound residues of pirimiphos–methyl[38], chlorpyrifos[39], dichlorovos[40], fenitrothion[41] and canola bound residues of prothiofos[33].

Conclusion

Refining processes led to progressive degradation of fenitrothion and pirimiphos-methyl insecticides. Their residues in maize seed oil included the parent compound beside both free and conjugated metabolites. About 81-87% of the ¹⁴C-activity in the crude maize seed oil could be eliminated by stimulated commercial processes locally used for oil refining. The deodorization was found to be an efficient step for reduction of the insecticide residues. Neutralization and bleaching processes led to significant losses of the pesticide residues in oil as well. The removal efficiency during the refining processes seems to depend upon the nature of the residues present. The data obtained emphasize the importance of studying the effect of oil refinement on the reduction and/or elimination of aged pesticides residues in edible oils.

The present results indicate that maize-bound fenitrothion or pirimiphos-methyl residues are highly bioavailable to rats fed with the extracted seeds. The total recovery of ¹⁴C-activity of insecticides amounted to 87-93% of the administrated dose. The main excretion routes are via expired air and urine and

small amounts of the radioactivity were distributed among various organs of experimental animals. Therefore, it should be stressed that the presence of bound pesticide residues can no longer be ignored in the evaluation of toxicological hazards.

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تأثير عمليات التنقية على كمية و طبيعة متبقيات الفينتروثيون والبريمفوس- ميثيل فى زيت الذرة والإتاحة البيولوجية للمتبقيات الصلبة (الكسب) على فئران التجارب

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تمت الدراسة على بذور الذرة الناتجة من النباتات المزروعة والمعاملة بالمبيدات الفوسفورية العضوية الفينتروثيون أو البريمفوس-ميثيل المرقمين بالكربون-١٤. وقد تبين إحتوائهما على ٠,١٢ ٪ و ٠,١٦ ٪ على التوالى من التركيز المستخدم. وقد اتضح ان تركيز متبقيات كل من مبيد الفينتروثيون والبريمفوس-ميثيل تتراوح ما بين ما بين ٢,٨-٣ (جزء فى المليون) فى زيت الذرة الخام و ٢,٣ (جزء فى المليون) فى مستخلص الميثانول فى حين كانت ١,٢-١,٤ (جزء فى المليون) فى الكسب (المتبقيات المتحدة بالحبوب) على التوالى.

وقد وجد أن عمليات التنقية تؤدى الى تناقص تدريجى لمتبقيات المبيدين فى الزيت الخام، وأن عمليتى إزالة الرائحة والمعاملة القلوية هما الأكثر تأثيراً فى عمليات التنقية لأنهما يعملان على إزالة نسبة تتراوح ما بين ٥٣-٥٨٪ من المتبقيات الموجودة فى الزيت الخام، وأن الزيت النهائى المنقى يحتوى على حوالى ١٣-١٩٪ من المتبقيات الأصلية.وقد دلت التحاليل الكروماتوجرافية للزيت الخام على وجود المبيد الأصلى مع ثلاثة مواد أخرى من نواتج التكسير فى كلا المبيدين.

وعند دراسة التمثيل البيولوجى للمتبقيات المتحدة الصلبة (الكسب) وذلك بتغذية الفئران الكبيرة لمدة ثلاثة أيام فى جهاز التمثيل البيولوجى، وجد أن أكبر نسبة من المتبقيات يتم إخراجها عن طريق التنفس (هواء الزفير) ٥٠٪ فى حالة مبيد الفينتروثيون وعن طريق البول (الكُلى) فى حالة البريمفوس-ميثيل (٤٧٪) فى حين كانت نسبة المتبقيات بالنسبة لكلا المبيدين فى البراز تتراوح ما بين ١٠-١٢٪ بالإضافة الى حوالى ٢٢-٢٦٪ من المتبقيات موزعة فى أعضاء الجسم المختلفة لفئران التجارب.