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Fate of ^{14}C-Ethion Insecticide in the Presence of Deltamethrin and Dimilin Pesticides in Cotton Seeds and Oils, Removal of Ethion Residues in Oils, and Bioavailability of Its Bound Residues to Experimental Animals

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ABSTRACT: Ethyl-1-^{14}C-ethion and some of its degradation products have been prepared for comparison purposes. Cotton plants were treated with ^{14}C-ethion alone and in the presence of deltamethrin and dimilin pesticides under conditions simulating local agricultural practice. ^{14}C-Residues in seeds were determined at harvest time; about 47.5% of ^{14}C-activity was associated with oil. After further extraction of seeds with ethanol, the ethanol-soluble ^{14}C-residues accounted for 10.6% of the total seed residues, whereas the cake contained about 37.3% of the total residues as bound residues in the case of ethion only. The bound residues decreased in the presence of deltamethrin and dimilin pesticides and amounted to 8.1 and 10.4% of the total residues, respectively. About 95% of the ^{14}C-activity in the crude oil could be eliminated by simulated commercial processes locally used for oil refining. Chromatographic analysis of crude cotton oil revealed the presence of ethion monooxon, *O*,*O*-diethyl phosphorothioate, and *O*,*O*-diethyl phosphoric acid in addition to one unknown compound in the case of ethion alone or ethion and dimilin. The same degradation products are found in the case of ethion and deltamethrin in addition to ethion dioxon, whereas ethanol extract revealed the presence of ethion dioxon and *O*,*O*-diethyl phosphoric acid as free metabolites. Acid hydrolysis of the conjugated metabolites in the ethanol extract yielded *O*,*O*-diethyl *S*-hydroxymethyl phosphorodithioate. The bound residues were quite readily bioavailable to the rats. After feeding rats with the cake containing ethion-bound residues, a substantial amount (60%) of ^{14}C-residues was eliminated in the urine, whereas the ^{14}C-residues excreted in expired air and feces were 10 and 9%, respectively. About 11% of the radioactive residues were distributed among various organs.

KEYWORDS: *^{14}C-ethion, residues, cottonseed oil, refining processes, bioavailability*

INTRODUCTION

Insecticide mixtures are usually used in the field to enhance the spectrum of control when multiple pests are attacking simultaneously.[1]

Cotton (*Gossypium hirsutum* L.) is an important economic crop in Egypt. The seed of the cotton plant can provide cottonseed oil, which is an important type of edible oil. The extracted meal can be used as animal feed because the protein content is 18−30% on undecorticated meal, whereas it is 35−45% on decorticated meal.[2]

Several investigators have suggested that most organophosphorus pesticides in edible oils can be reduced considerably by chemical refining processes.[3−7]

Ethion insecticide *O*,*O*,*O*′,*O*′-tetraethyl *S*,*S*′-methylene bis(phosphorodithioate) is an organothiophosphate member of the organophosphate pesticide family.[8] It was first developed as a nonsystemic insecticide and acaricide for use on fruit trees, including citrus fruits, nut trees, cotton, and seed and forage crops as well as a wide variety of fruits and vegetables.[9] It is also used for controlling aphids, spider mites, scale insects, thrips, lepidopterous larvae, leafhoppers, maggots, suckers, and soil-dwelling insects on a wide variety of food, fiber, and ornamental crops, including grapes, fruits, vegetables, and nuts.[10]

Some crops, such as cotton, need heavy repeated applications of different pesticides, including organophosphates, carbamates, pyrethroids, and organochlorines. The purpose of the present work was (i) to determine and identify ethion residues in cotton seeds and oils alone and in the presence of deltamethrin and dimilin pesticides, (ii) to determine the effect of different simulated commercial processing procedures on the elimination of the insecticide residues from cottonseed oil obtained from plants treated with ^{14}C-ethion, and (iii) to study the bioavailability of ethion-bound residues to experimental animals.

MATERIALS AND METHODS

Radiochemical. ^{14}C-Ethion labeled at the carbon atoms of ethyl groups (I) was prepared in a single-vessel reaction using ^{14}C-ethanol (specific activity = 37 MBq, Amersham, UK) according to the method of Abdel-Gawad et al.[11] The prepared ^{14}C-ethion had a specific activity of 7.4 MBq/g, and the radiometric purity was 98% (Figure 1).

For identification purposes, nonlabeled ethion and its metabolites such as ethion monooxon (**II**), ethion dioxon (**III**), *O*,*O*-diethyl phosphorothioate (**IV**), *O*,*O*-diethyl phosphoric acid (**V**) and *O*,*O*-diethyl *S*-hydroxymethyl phosphorodithioate (**VI**) have been synthesized according to known procedures.[11] The structures of these compounds are shown in Figure 2.

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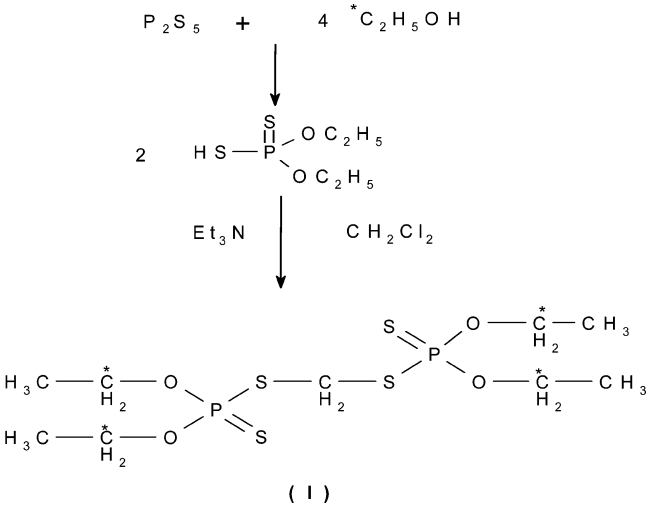

Figure 1. Pathway of ^{14}C-ethion synthesis.

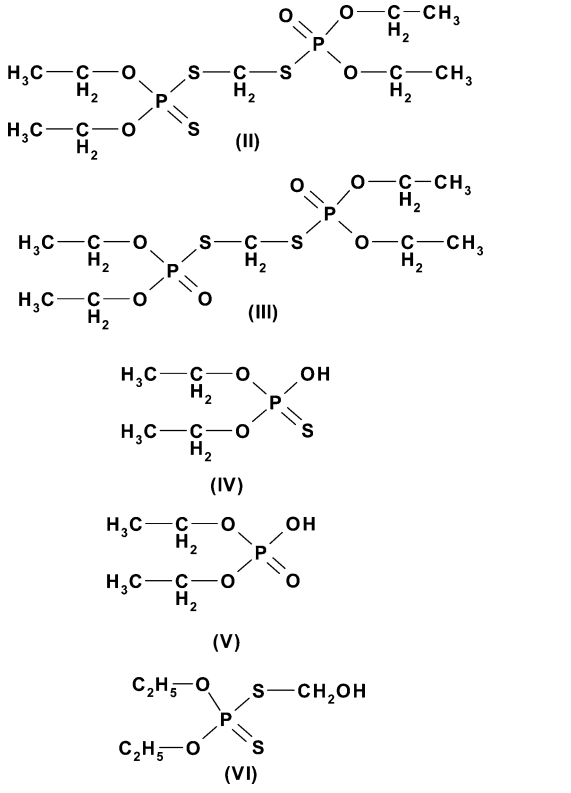

Figure 2. Main degradation products of ethion insecticide.

Field Experiments. *Treatment of Plants with Pesticides.* Pesticide-free *G. hirsutum* L. seeds (var. Giza 86) were obtained from the Agricultural Research Centre, Cairo. The seeds were cleaned from any dockage and impurities before cultivation. Sound whole seeds of cotton were cultivated under normal field conditions in a controlled, isolated field area. Irrigation, fertilization, and hand weeding were performed, and soil management was conducted as usually practiced in the field. A control field plot of similar soil characteristics to the experimental plot was cultivated as in the experimental plot and kept untreated with pesticides. Shortly at flowering stage, leaves of plants were treated twice, 21 days apart, with ^{14}C-ethion alone (8 mg/plant) and in the presence of deltamethrin (3.6 mg/plant) and dimilin (1.6 mg/plant) pesticides. Samples of cotton plant were collected manually at harvest time. The cotton seed samples were air-dried and delinted to get cotton seed for preparation of oil and cake and determination of radioactivity.

Extraction of Cotton Seeds. The analysis procedures for ^{14}C-residues in cotton seeds are shown in Figure 3. Dry cotton seeds were crushed and extracted with *n*-hexane for 12 h using a Soxhlet apparatus. After evaporation of hexane under reduced pressure, radioactivity in the cotton oil was measured. The residue remaining after extraction (cake A) was further extracted with ethanol (Figure 3). Aliquots of the hexane extract (oil) and the ethanol extract were determined for radioactivity.

The remaining cake (cake B) was air-dried and digested by adding 1 mL of Solusol (tissue and gel solubilizer); samples were digested at 40−50 °C until the tissue dissolved and then decolorized with 30% H_2O_2 (1 mL). Seventy microliters of glacial acetic was added to eliminate chemiluminescence.[12] The radioactivity was determined using a liquid scintillation counter.

Removal of Pesticide Residues from Oils. Oil processing was conducted in four steps, namely, alkali refining, bleaching, winterization, and deodorization, as previously reported.[5]

Neutralization (Alkali Refining). Samples of the crude cotton oil (17.5 g) were heated at 85 °C with continuous stirring for 30 min and then neutralized by adding 2 N NaOH ,and stirring was continued for another 30 min. The mixture was then centrifuged at 3000 rpm for 10 min. After the oil had been separated from the soap stock, i was washed with 100 mL of hot distilled water several times using a separating funnel until the washings were neutral. This process removed free fatty acids and other acidic material from the oil. The ^{14}C-activity of the oil was determined.

Bleaching. Bleaching clay performs not only color removal but also the removal of trace metals and the adsorption of phospholipids, soap, and decomposition of oxidation products such as peroxides. Neutralized oil was heated to 60 °C and fuller earth (Tonsil, 0.15 g) was added. The mixture was then heated to 100−110 °C on an oil bath for 30 min with continuous stirring. The oil was centrifuged at 3000 rpm for 10 min, and ^{14}C-residue levels were determined.

Winterization. The clear dry oil was winterized (cooled) at 5 °C for 3 days, and the high saturated glycosides that separated were removed by filtration or centrifuged at 3000 rpm for 10 min; ^{14}C-residue levels were determined.

Deodorization. Deodorization is the last major processing step in the refining of edible oil and is responsible for removing undesirable ingredients. The winterized oil was heated at 200−220 °C on an oil bath while superheated steam passed. The deodorization was continued for 3 h. The oil was then separated, and ^{14}C-residue levels were determined.

Bioavailability of Bound Residues in Rats. Three-month-old, sexually mature white male rats (derived from Sprague−Dawlay strain) weighing 150 ± 10 g were purchased from animal house colony, Dokki, Giza Egypt. The animals were individually housed in glass metabolism cages that allowed separate collection of feces, urine, and expired air. The rats were conditioned for 2 days to a daily diet consisting of standard feed mixed with extracted untreated cotton seeds for acclimatization under the laboratory conditions (29 ± 3 °C). The animals were kept without food for 24 h and then fed the diet containing bound ^{14}C-ethion residues for 3 days. To make the feed palatable, the extracted seeds were mixed thoroughly with an equal amount of white cheese, and the paste was left to dry.

The respiratory carbon dioxide was trapped in a 10% NaOH solution. Urine, feces, and $^{14}CO_2$ were collected separately for 3 days and assayed for radioactivity. After the end of 3 days, each rat was lightly anaesthetized with ether and blood removed from the pumping heart. The animal was then killed with an overdose of ether, and samples from liver, kidney, fat, blood, lung, and brain were collected and kept frozen until analysis of radioactivity by digestion and LSC.

Isolation and Characterization of ^{14}C-Residues. Analysis of radioactive compounds was achieved by thin layer chromatography (TLC). Samples of crude oil at harvest time and after each refining process were partitioned between acetonitrile and hexane to remove the oil. The radioactive residues were almost completely retained in

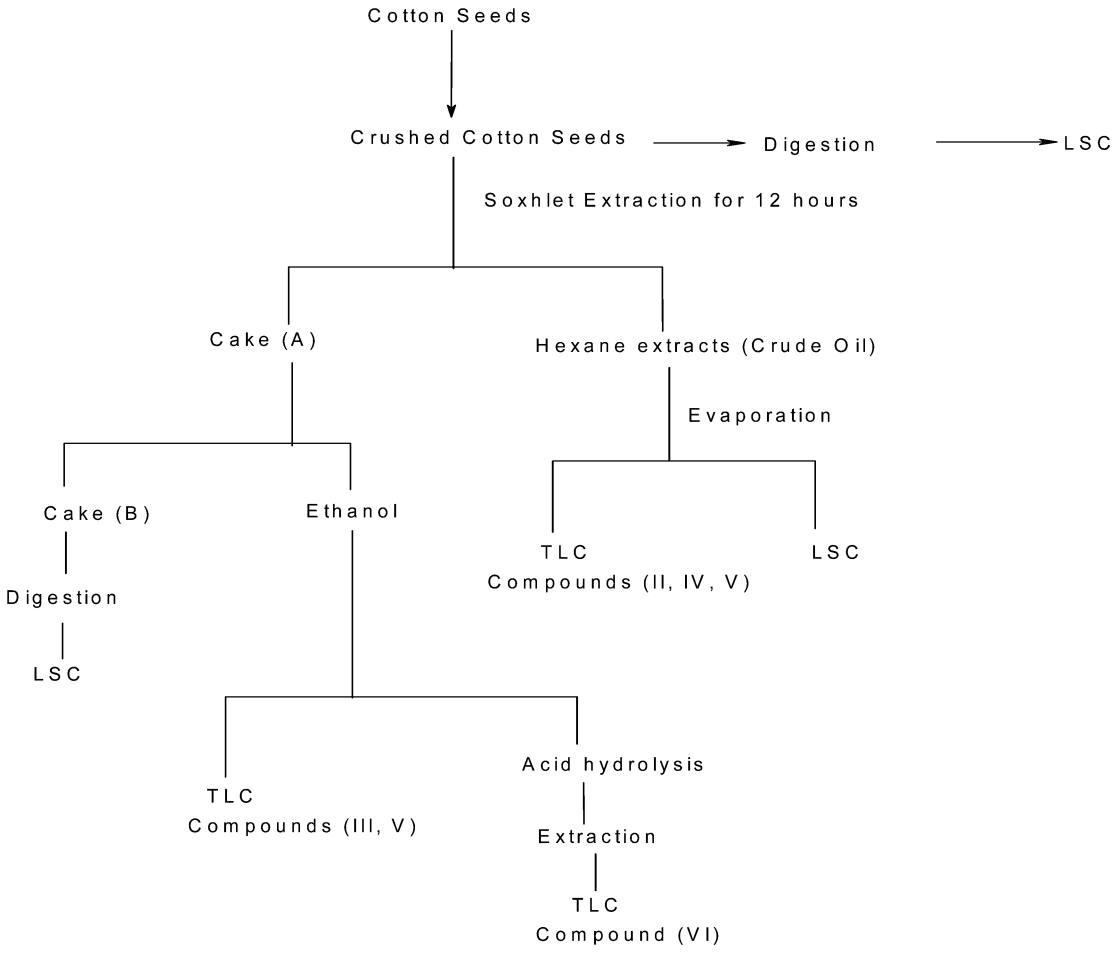

Figure 3. Analysis procedures for ^{14}C-residues in cotton seeds and oils.

the acetonitrile layer. Analysis of crude and refined oil extracts were achieved by TLC.

Urine was extracted with chloroform (chloroform 1), and the aqueous layer was then acidified with 2 N HCl and heated for 2 h at 100 °C on a water bath and reextracted with chloroform three times. The combined chloroform was dried over anhydrous sodium sulfate, filtered off, and evaporated under vacuum (chloroform 2) to obtain the conjugated metabolites. Aliquots from both chloroform 1 and 2 and ethanol layers (after hexane extraction) were analyzed by TLC on silica gel plates using suitable solvents.

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, Darmstadt, Germany) using these solvent systems: system 1, toluene/xylene 50:50 (v/v); system 2, dioxane/xylene/petrolum ether 20:40:40 (v/v/v); system 3, *n*-hexane/ethyl acetate 98:2 (v/v).

Authentic samples were run alongside as references, and spots were viewed under UV light at 254 nm and by spraying the plates with a freshly prepared Hans–Isherwood reagent,[13] or after preliminary spraying with $PdCl_2$ solution, the plates were subjected to I_2 vapor to detect the compound by color.[14,15]

Radioactivity Measurements. Radioactivity in oil (acetonitrile extracts), ethanol extracts, urine, and other liquid samples was measured directly by LSC using a Packard Tri-Carb liquid scintillation spectrometer (model 2300, PerkinElmer Life Science, Boston, MA, USA) in vials using a dioxane-based scintillation cocktail composed of dioxane (1 L), naphthalene (100 g), 2,5-diphenyloxazole (PPO; 10 g), and 1,4-di[2-(5-phenyloxazolyl)]benzene (POPOP; 0.25 g).

Cake, feces, ground cotton, and animal tissues (100 mg) were assayed for radioactivity by digestion using [1 mL of Solusol (tissue and gel solubilizer), 1 mL of 30% H_2O_2, and 70 μL of glacial acetic at 40–50 °C]. The radioactivity was determined using a LSC. The internal standard 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

^{14}C-Residues in Seeds, Oil, and Cake. The ^{14}C-residues in dry cotton seeds obtained from ^{14}C-ethion-treated plants amounted to 0.11% of the applied dose at harvest time. The percentage of residues decreases to 0.08 and 0.05% in the presence of deltamethrin and dimilin pesticides, respectively (Table 1). The insecticide was absorbed and translocated very slowly from the treated leaves to the seeds of investigated plant due to enzyme metabolic activity.[5,16] Following foliar application of soybean plants with ^{14}C-pirimiphos-methyl,[17] and cotton plants with ^{14}C-malathion,[18] the aged insecticide

Table 1. ^{14}C-Residues in Seeds after Treatment of Cotton Plants with ^{14}C-Ethion in the Presence of Deltamethrin and Dimilin Pesticides at Harvest Time

sample[a]	weight (g)	average ^{14}C-residues[b] (ppm)	%[c]
ethion	88.7	0.59 ± 0.051	0.11
ethion and deltamethrin	81.8	0.47 ± 0.024	0.08
ethion and dimilin	93.0	0.27 ± 0.011	0.05

[a]At harvest time. [b]Results are expressed as the mean ± SD for three determinations of ethion residue level for each sample. [c]Percent of applied dose.

Table 2. ^{14}C-Residues in Extracted Seeds after Treatment of Cotton Plants with ^{14}C-Ethion in the Presence of Deltamethrin and Dimilin Pesticides at Harvest Time

sample[a]	oil hexane[b] (ppm)	ethanol[b] (ppm)	cake[b] (ppm)	% recovery
ethion	0.280 ± 0.025	0.063 ± 0.005	0.220 ± 0.015	95.4
ethion and deltamethrin	0.280 ± 0.018	0.098 ± 0.003	0.038 ± 0.002	88.5
ethion and dimilin	0.140 ± 0.004	0.083 ± 0.002	0.028 ± 0.001	92.9

[a]After the second application. [b]Results are expressed as the mean ± SD for three determinations of ethion residue level for each sample.

residues in seeds amounted to 0.37 and 0.11% of the applied dose, respectively. The data obtained are in agreement with those reported by Singh et al.[19] in their studies on the persistence of ethion residues on cucumber. Although persistent insecticides are beneficial for controlling pests for extended periods, their residues in consumable parts of the crops may be harmful to the consumers when they exceed the maximum residue limit (MRL) values.[20]

About 47.5% of ^{14}C-activity in dry seeds was found to be associated with oil (hexane extract). The ethanol-soluble ^{14}C-residues accounted for 10.6% of the total seed residues, whereas the cake contained about 37.3% of the total residues as nonextractable or bound residues in the case of ethion only. The percentage of bound residues decreases when deltamethrin or dimilin pesticide was used in combination with ethion and amounted to 8.1 and 10.4%, respectively. The recovery percent of the applied radioactivity ranged from 88.5 to 95.4% as shown in Table 2. When two compounds are mixed, there are basically four types of interactions that change the efficacy of pesticide combinations; they can either be additive effects or synergistic responses (potentiating) or antagonism or enhancement in an insect species. If a mixture is potentiating, it is a useful tool in enhancing control efficacy and combating insecticide resistance. If a mixture is antagonistic, it should not be used, because it will reduce the efficiency of pest control and aggravate the resistance problem.[1,21] It is worth mentioning that Zayed et al.[22] found that repeated applications of pesticides might enhance the release of ^{14}C-bound residues in their study on the impact of repeated pesticide applications on the binding and release of methyl ^{14}C-monocrotophos and U-ring-labeled ^{14}C-carbaryl to soil matrices under field conditions.

Effect of Refining Processes. Simulated commercial processing procedures resulted in a gradual decrease in the total amount of ^{14}C-residues in oils with aged residues (Table 3). This decrease could be attributed to alkali hydrolysis, effect of adsorption, effect of heat, or evolution of $^{14}CO_2$ gas. The deodorization processes proved to be the most effective step in reducing ^{14}C-ethion residues, as they eliminated 95% of the residues originally present in crude oil. On the other hand, the alkali treatment, bleaching, and winterization eliminated 30, 75, and 82% of the originally present radioactivity in oil, respectively. These results agree with Miyahara and Saito,[23] who found that in some organophosphorus insecticides, such as malathion, dichlorvos, and chlorpyrifos, the deodorizing process was effective in removing pesticide residues due to decomposition and volatilization. This also agreed with the findings of Ruiz Méndez et al.,[24] who reported that pesticide residues (simazine) were highly eliminated in olive oil during the deodorizing step. Zayed et al.[17,25] showed that the amounts of both ^{14}C-carbofuran and ^{14}C-pirimiphos-methyl residues in soybean oil decreased to 16 and 25%, respectively, through the refining processes. Hegazi et al.[26] found that the sunflower oil obtained from treated sunflower plants lost about 64% of the total ^{14}C-zineb residues in crude oil through refining processes. Abdel-Gawad and Hegazi[5] noted that the refined oil from canola seeds treated with prothiofos lost about 69% of the total ^{14}C-residues in crude oil through the processing procedures. Also, Abdel-Gawad et al.[6,7] reported that the refined oil from maize and soybean seeds treated with ethion lost about 70 and 62.5% of the total ^{14}C-residues in crude oil, respectively, through refining processes.

Table 3. Effect of Refining Processes on ^{14}C-Ethion Residues in Cottonseed Oil

sample	^{14}C-residues[a] (ppm)	% reduction[b]
crude oil	0.280 ± 0.025	0.0
neutralized oil	0.196 ± 0.017	30
bleached oil	0.070 ± 0.004	75
winterized oil	0.050 ± 0.003	82
deodorized oil	0.014 ± 0.001	95

[a]Results are expressed as the mean ± SD for three determinations of ethion residue level for each sample. [b]Percent reduction = [(crude oil − each step)/crude oil] × 100.

Identification and Characterization of Radioactive Degradation Products in Oil. The R_f values and the average concentration of ^{14}C-ethion and its metabolites in cotton oil extract at harvest time are shown in Table 4. Chromatographic analysis of cotton oil extracts (acetonitrile layer) revealed the presence of ethion monooxon (**II**), *O*,*O*-diethyl phosphorothioate (**IV**), and *O*,*O*-diethyl phosphoric acid (**V**) in addition to one unknown compound in the case of ethion alone and ethion and dimilin. The same degradation products are found in the case of ethion and deltamethrin besides ethion dioxon (**III**).

The ethanol-extractable residues contain the hydrophilic compounds ethion dioxon (**III**) and *O*,*O*-diethyl phosphoric acid (**V**) as free metabolites, in addition to an unknown compound. Acid hydrolysis of the ethanol extract with 2 N HCl at 100 °C followed by extraction with chloroform yielded *O*,*O*-diethyl *S*-hydroxymethyl phosphorodithioate (**VI**).

The residues in refined cottonseed oil were identified as ethion monooxon (**II**), *O*,*O*-diethyl phosphorothioate (**IV**), and *O*,*O*-diethyl phosphoric acid (**V**) in addition to one unknown compound. Their amounts decreased during the refining steps (Table 5). Abdel-Gawad et al.[11] studied the distribution and elimination of ^{14}C-ethion in chamomile flowers and found the presence of the parent compound together with four metabolites, which were identified as ethion monooxon, ethion dioxon, *O*,*O*-diethyl phosphorothioate, and *O*-ethyl phosphorothioate.

The suggested metabolic fate of ethion labeled with ^{14}C-ethyl groups in cotton seeds and oil after double application to leaves in the presence of deltamethrin and dimilin pesticides at harvest time is shown in Figure 4. Three pathways are suggested for the degradation of ^{14}C-ethion in cotton seeds. The first is oxidation of P═S to P═O, yielding ethion monooxon (**II**) and ethion

Table 4. ^{14}C-Ethion and Its Degradation Products in the Presence of Deltamethrin and Dimilin Pesticides in Extracts of Cottonseed Oil (Crude Oil) at Harvest Time

substance	R_f in system[a]			metabolites concentration (ppm)		
	1	2	3	ethion	ethion and deltamethrin	ethion and dimilin
ethion (**I**)	0.75	0.80	0.75	0.000	0.000	0.000
ethion monooxon (**II**)	0.88	0.92	0.65	0.060	0.035	0.027
ethion dioxon (**III**)	0.69	0.56	0.55	0.000	0.037	0.000
O,*O*-diethyl phosphorothioate (**IV**)	0.33	0.23	0.42	0.036	0.053	0.024
O,*O*-diethyl phosphoric acid (**V**)	0.61	0.71	0.82	0.071	0.031	0.022
unknown	0.10	0.15	0.00	0.095	0.119	0.056

[a]System 1, toluene/xylene 50:50 (v/v); system 2, dioxane/xylene/petrolum ether 20:40:40 (v/v/v); system 3, *n*-hexane/ethyl acetate 98:2 (v/v).

Table 5. R_f Values and Concentrations of ^{14}C-Ethion and Its Degradation Products in Cottonseed Oil after Subjection to Commercial Processing Procedures

substance	R_f in system[a]			metabolites concentration[b] (ppm)		
	1	2	3	crude oil	A	D
ethion (**I**)	0.75	0.80	0.75	0.000	0.000	0.000
ethion monooxon (**II**)	0.88	0.92	0.65	0.060	0.010	0.001
O,*O*-diethyl phosphorothioate (**IV**)	0.33	0.23	0.42	0.036	0.020	0.006
O,*O*-diethyl phosphoric acid (**V**)	0.61	0.71	0.82	0.071	0.060	0.003
unknown	0.10	0.15	0.00	0.095	0.091	0.003

[a]System 1, toluene/xylene 50:50 (v/v); system 2, dioxane/xylene/petrolum ether 20:40:40 (v/v/v); system 3, *n*-hexane/ethyl acetate 98:2 (v/v). [b]A, neutralization; D, deodorization.

dioxon (**III**). The second is cleavage of the P−S−alkyl to yield *O*,*O*-diethyl phosphorothioate (**IV**) and *O*,*O*-diethyl phosphoric acid (**V**). The third is hydrolysis to yield *O*,*O*-diethyl *S*-hydroxymethyl phosphorodithioate (**VI**). Katagi and Mikami[27] noted that the metabolism of organophosphorus pesticides in plants has revealed cleavage of the P−O−aryl linkage and *O*-dealkylation to be among the most predominant metabolic pathways. Ethion is converted via desulfuration in the liver by cytochrome P-450 enzymes to its active oxygen analogue, ethion monooxon.[28]

Bioavailability in Rats. Elimination and distribution of ^{14}C-ethion residues following the feeding of rats with the extracted cotton seeds (cake containing 13.2 μg equiv per rat) are shown in Table 6. Most of the radioactivity was eliminated via urine (60%), whereas residues in feces and expired air were 9 and 10% of the ingested ^{14}C-activity, respectively. Appreciable amounts of ^{14}C-residues (11%) were also detected in the liver, kidney, blood, lung, brain, and fat. Bound ^{14}C-ethion in cotton

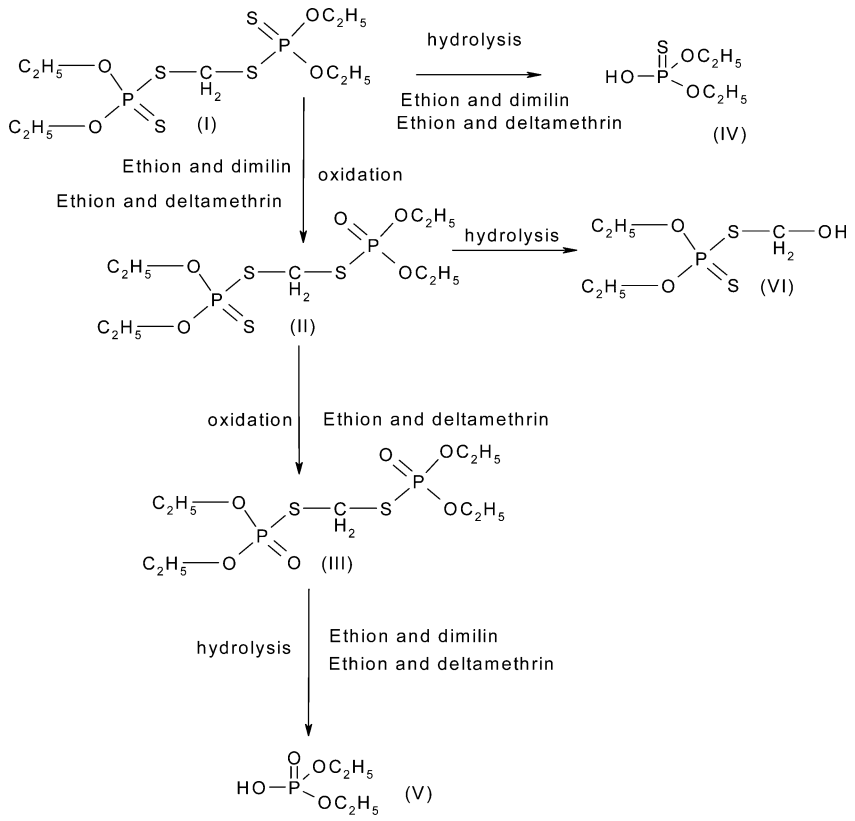

Figure 4. Metabolic pathway of ^{14}C-ethion in cottonseed oil and in the presence of deltamethrin and dimilin.

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Table 6. Excretion and Distribution of ^{14}C-Bound Ethion Residues in Cotton Seed after Feeding to Rats for 72 h

sample	insecticide[a] equivalent (μg)	% of administered dose
carbon dioxide	1.30 ± 0.11	10
urine	7.90 ± 0.680	60
faces	1.20 ± 0.070	9
liver	0.50 ± 0.020	4
kidney	0.10 ± 0.008	1
fat	0.20 ± 0.009	1.5
blood	0.40 ± 0.010	3
lung	0.10 ± 0.007	1
brain	0.07 ± 0.003	0.5
total recovery	11.77	90

[a]Administered dose = (13.2 μg equivalent per rat) 100%. Results are expressed as the mean ± SD for three determinations of ethion residue level for rats.

seeds proved to be highly bioavailable to rats. Similar observations were reported from studies on the bioavailability of soybean-bound residues of dichlorovos[29] and fenitrothion.[30] Also, the data obtained are in line with many other studies that indicate a moderate to high bioavailability of grain-bound ^{14}C-pesticide residues in experimental animals.[5,31,32]

Chromatographic analysis of urine extracts (chloroform layer) revealed the presence of three metabolites, which were identified as ethion dioxon (**III**), *O*,*O*-diethyl phosphorothioate (**IV**), and *O*,*O*-diethyl phosphoric acid (**V**) as free metabolites in addition to one uknown substance. Compound *O*,*O*-diethyl *S*-hydroxymethyl phosphorodithioate (**VI**) was detected as a conjugated metabolite, and its concentration was 0.004 ppm (Table 7). This result agrees with some previous studies,[5,33,34]

Table 7. ^{14}C-Ethion and Its Degradation Products in Urine of Male Rats Fed with Cake Containing Ethion-Bound Residues after 72 h

	R_f in system[a]			
metabolite	1	2	3	urine extract chloroform layer[b] (ppm)
III	0.69	0.56	0.55	0.042 ± 0.0040
IV	0.33	0.23	0.42	0.015 ± 0.0011
V	0.24	0.50	0.71	0.033 ± 0.0028
unknown	0.10	0.15	0.00	0.038 ± 0.0031

[a]System 1, toluene/xylene 50:50 (v/v); system 2, dioxane/xylene/petrolum ether 20:40:40 (v/v/v); system 3, *n*-hexane/ethyl acetate 98:2 (v/v). [b]Results are expressed as the mean ± SD for three determinations of ethion residue level for rats.

which noted that in adult rats, prothiofos gave prothiofos oxon, *O*-ethyl phosphoric acid, and 2,4-dichlorophenole. Fenitrothion insecticide was converted to phosphorothioate and dimethyl-phosphate. Malathion insecticide was converted to malathion monocarboxylic acid and malathion dicarboxylic acid. Carbofuran, on the other hand, gave carbofuran phenol and 3-hydroxycarbofuran. Ethion is converted via desulfuration in the liver by cytochrome P-450 enzymes to its active oxygen analogue, ethion monooxon.[28]

Conclusion. The joint action of deltamethrin and dimilin in combination with the organophosphate ethion was studied on cotton plants by using radiolabeled insecticide. Ethion produced a good potentiation with deltamethrin and dimilin, respectively. The residues of ethion in cotton oil included both free and conjugated metabolites. The ^{14}C-ethion residues decrease in the presence of deltamethrin and dimilin pesticides. The number of degradation products increases in the presence of deltamethrin more than with dimilin. The bound residues decrease in the presence of deltamethrin and dimilin pesticides as compared with ethion alone. The major part of the insecticide residues (about 95%) could be eliminated during processing of the oil. Refining processes led to progressive degradation of the parent insecticide. The deodorization was found to be an efficient step for reduction of the insecticide residues. Alkali treatment, bleaching, and winterization 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 residue present. The data obtained emphasize the importance of studying the effect of oil refinement on reduction or elimination of aged pesticide residues in edible oils. The present results obtained indicate that cotton-bound ^{14}C-ethion residues are highly bioavailable to rats. 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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Notes

The authors declare no competing financial interest.

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