Egyptian Journal of Chemistry

http://ejchem.journals.ekb.eg/

Impact of the insecticide ^{14}C-chlorfenvinphos in a terrestrial ecosystem

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Abstract

Pesticides are an essential component of agricultural production. The increased use of pesticides greatly aided agricultural production, reduced stored grain losses, and improved human well-being in general. However, the use of pesticides may lead to unwanted residues such as traces of pollutants from food, the environment and living tissues. The environmental fate of ^{14}C- chlorfenvinphos was studied using an agricultural ecosystem that included soil, plants, beetles, earthworms and one type of common bird in Egypt (Asfour Baladi). This study was conducted on a restricted field area that was cultivated with maize plants as target crop and soybean plants as an alternate crop. The residue level in soybean seeds was almost 6 times higher than in dry maize seeds. The high concentration of 14C- residues in beetles (1.28µg/g) was observed on day 26 after spraying ^{14}C- chlorfenvinphos and decreased thereafter. The earthworms, on the other hand, showed a progressive increase with time and its maximum residue was detected after 75 days (3.25µg/g) .The dead birds showed the highest concentration of residues in the brain, liver and heart as compared with alive birds in the ecosystem. The chromatographic analysis indicated the presence of the parent compound in addition to three degradation products especially in dry seeds and soil.

Key words, ^{14}C-chlorfenvinphos, terrestrial ecosystem, soil, plants, insects, birds.

1. Introduction

Ecosystems are integrated and stable systems; they include humans, all other species on this planet, and basic biotic and a biotic processes. Prediction and assessment of ecological impacts caused by pesticides alert humans to the dangers of some alterations to the environment. Components of ecosystems cannot be changed or destroyed without directly or indirectly impacting the human condition. It is to humans' benefit to consider all uses and risks of pesticides to ensure preservation of critical systems in the environment. Large amounts of pesticides cause a lot of ecological and environmental problems, among which, the impact of pesticides on biodiversity is the most important. Pesticides represent a relevant stressor for many terrestrial species [1]. At the same time, we have realized over the past decades that agricultural chemical residues have spread into the environment, causing major pollution of terrestrial ecosystems and poisoning human foods [2-6].

They have been shown potentially affect to all groups of organisms in ecosystems e.g. microorganisms [7], invertebrates [8], plants [9] and fish [10]. The Intensive use of agrochemicals and contamination of the natural environments by organic pollutants, such as pesticides is a matter of great concern worldwide and have stimulated scientists to develop reliable methods to assess the potential of the transformations of these chemicals. [11, 12]. In the last few decades, pesticides have been used on an increasingly wider scale throughout the world, although lately there is a tendency to slow down, or at least to use less harmful molecules [13]. Organophosphate pesticides (OP) belong to the most toxic environmental pollutants and they cause numerous intoxications and fatalities all over the world [14, 15]. These compounds are known to damage the different organs and systems at both acute and chronic exposure [16-18]; however, the main unfavorable effect of their action in mammals is the impact on the nervous system. Pesticides can con-

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Receive Date: 20 October 2020, Revise Date: 03 December 2020, Accept Date: 12 July 2021
DOI: 10.21608/EJCHEM.2021.46847.2956

taminate soil, water, turf, and other vegetation. In addition to killing insects or weeds, pesticides can be toxic to a host of other organisms including birds, fish, beneficial insects, and non-target plants[19] .

Chlorfenvinphos; 2-chloro-l-(2',4'-dichlorophenyl) vinyl diethylphosphate is one of the most important members of the vinyl phosphate insecticides family. It is sold under the common trade names Birlane, Dermaton and Supona.It has a broad-spectrum as contact and non-systemic insecticide with low mammalian toxicity. It is used against insects, worms, parasites in veterinary medicine and in soil. Recently, it has been reported that chlorfenvinphos in combination with cypermethrin (pyrothroid), gives excellent control of all known resistant tick strains. This combination of chemicals also controls three host ticks and buffalo fly and may be used on beef and dairy cattle, horses, deer, sheep and goats by means of plunge dips, spray races or hand sprays [20].

Aim of the present work

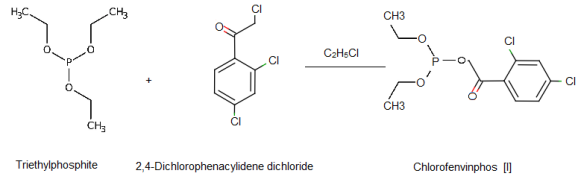
Most studies were usually performed on one organism at a time. So they do not give an insight on the overall behavior of the insecticide in an ecosystem. The present work is an attempt to study the fate of the vinyl phosphate insecticide 14C- chlorfenvinphos in an agricultural ecosystem. The ecosystem included soil, plants (maize and soybean) earthworms, beetles and birds. For these 14C–chlorfenvinphos (2-chloro-1- (2,4-dichlorophenyl) vinyl diethyl phosphate was used for studying pesticide residues. Nuclear techniques provide a versatile and valuable tool.

MATERIALS AND METHODS

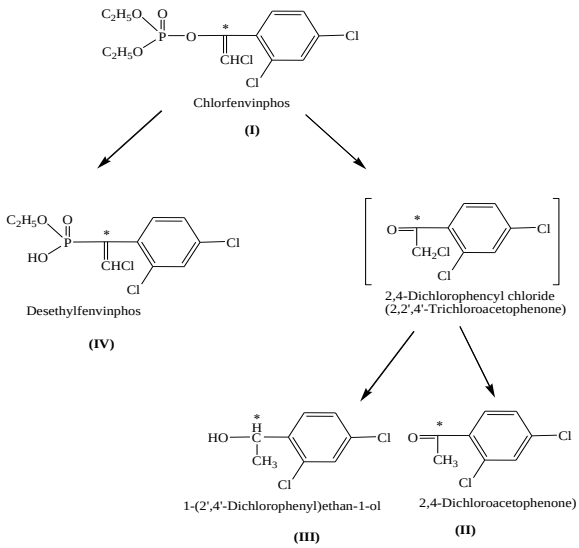
All solvents and chemicals used for extraction, plates of thin layer chromatography (TLC) and solid phase extraction (SPE) columns were of analytical grade.

Chemicals

14C- Chlorfenvinphos (2-chloro-1-(2,4-dichlorophenyl) vinyl diethyl phosphate labelled at carbon atoms of vinyl groups was obtained from Izinta Isotope Trading Enterprise of the Institute of Radioisotopes ,Budapest, Hungary . It had a specific activity of 1 MBq/mg and radiometric purity of 98%. Pure non-labelled chlorfenvinphos was synthesized by the reaction of triethyl phosphite with 2, 4-dichlorophenacylidene dichloride according to a known method [21] as shown in Scheme 1. For identification purposes, substances of chlorfenvinphos metabolites have been synthesized according to procedures reported by Hutson [22]. as shown in Scheme 2



Scheme 1: Synthesis of chlorfenvinphos insecticide



Scheme 2: Chlorfenvinphos insecticide and its main degradation products

Description of Terrestrial Ecosystem:

A restricted field area of 25 m2 (5mx5m) was used in this experiment .The plot was furnished with fresh fertile clay- loamy soil. It was separated from the surrounding bigger plot by a double walled plastic network from all sides. This proved to be efficient in preventing soil insects and birds from escaping.

Soil

Clay loamy soil from a cultural area in north delta of Egypt with the characterteristics: 49% clay, 29% silt, 22% sand, 1% organic carbon, pH 7.6 and 22% moisture content .

Seeds

Pesticides- free Zeamays seeds (Var. Giza. 2 hybrid) and soybean seeds (Var. Clark) were obtained from Agriculture Research Centre (Cairo).The seeds were cleaned from any dockage and impurities before cultivation.

Earthworms and Beetles

A total number of 300 earthworms (Sp.Allolobophora) and 400 beetles (Calosoma chlorostictum) were added to the soil of the ecosystem before spraying the plants. These insects were purchased from local source and identified in Department of Entomology at Faculty of Science, Cairo University.

Birds

House sparrows (Passer dometicusniloticus; total number 45) were introduced into the ecosystem shortly after spraying the plants with the insecticide14C- chlorfenvinphos. Birds were obtained from local sources and identified at Zoology Department, Faculty of Science, Cairo University.

Application of the Insecticide

The ecosystem plot was cultivated with the target crop (maize) for two successive years (April. 2012 and 2014), and with soybean plants as an alternate crop .The total number of plants in the plot was 80 (50 maize plants and 30 soybean plants) . At blooming (after two months following the cultivation), plants were sprayed twice, 23 days apart, with 14C- chlorfenvinphos using a standard laboratory sprayer. Each plant was treated with a solution of labelled insecticide which was diluted with the non –labelled pure insecticide in water (total spray: 5 mg /2.5 µCi /plant; 3 days: after 2 days of the first spray; 26 days: after 2 days of the second spray.

Sampling

At different time intervals 3, 24, 26, 60 and75 days following foliar application, samples of soil, plants, green seeds, beetles, earthworms and birds were taken for analysis .For roasting soft green maize ears were taken at 70 days. Mature dry seeds were collected at the end of the experiment. For soil sampling, hard polyvinyl chloride cylinders (HPVC) (50cm length × 5cm.i.d.) open at both ends, were used. Columns were inserted in the field soil. Columns in duplicates were removed carefully for analysis at various intervals (over 75 days). The soil columns were sliced into 0-5, 5-10 and 10-15 cm sections for analysis and carefully removed from the cylinder sections. The soil was air dried, mixed thoroughly and three sub-samples (from each depth) were sieved and analyzed from each column. Soil was stored at –200C till analysis.

Extraction and Clean-up
Soil

Three samples (30g) from each zone were extracted in a Soxhlet apparatus with methanol for 6 hours. This proved to be sufficient for removal of all extractable. Further extraction of extracted soil with the solvent gave no further extractable residues of the pesticides. Extractable residues were determined in a Liquid Scintillation Counter (LSC) [23]. Bound 14C-residues were determined by combustion in a Harvey Biological Oxidizer (OX-600). For each column, the overall residue load, and hence persistence, was calculated by summing increment loads of the column. The extractable residues in plant tissues, beetles, earthworms and organs of the birds were isolated through homogenization with excess methanol in ultraturax for 30 minutes, followed by centrifugation.

Seeds

Samples of mature maize and soybean seeds were crushed in a mortar, and extracted with methanol for 4-6 hours in a soxhlet apparatus to obtain the free residues.

Feces

Feces of house sparrows in the ecosystem plot were collected and extracted with methanol before the second spray and at the end of experiments.

The pretreatment of methanolic extracts of seeds and soil included clean-up processes using specific column as Solid Phase Extraction (SPE) 500 mg C18 Sep-Pak cartridge, (Water Association, Millipore, USA.) .Cartridge was conditioned with ethyl acetate (10 mL) followed by methanol (5 mL) and distilled, deionized (DD) water (10 mL) without allowing the cartridge to dry out. The sample extract (1 ml) was diluted with water: methanol (1: 1) loaded on and passed through the C18 Sep-Pak cartridge at an approximated speed of one drop per second. The cartridge was dried by blowing nitrogen over it for 20 minutes. The adsorbed pesticide and its residues were eluted by ethyl acetate (3 ml). The organic extract was evaporated to dryness on the rotary evaporator. The residue was dissolved in acetonitrle (0.5 ml) and then analyzed by Thin layer chromatography) TLC).

Chromatographic Analysis

The extractable residues from different samples were analyzed using authentic samples of chlorfenvinphos and its degradable on precoated silica gel chromatoplates 20x20 cm; 0.25mm thickness with fluorescent indicator (kieselgel 60 F254, Merck, Germany) using the three solvent systems..

System 1. Ethanol: Benzene (1:9) System 2. Acetone: Hexane (2:8)

System 3. Dichloromethane: Acetonitrile: n-Hexane (3:3:8 v/v/v)

Authentic samples were run alongside as reference materials for location of spots. Plates were seen under UV – light at 254 nm and spots were made visible by spraying the plates with Hans Isherwood reagent [24].

Radioactivity Measurements

Radioactivity of extractable residues was directly determined by counting in a Packard 3320 Liquid Scintillation Spectrometer. The remaining non – extractable residues as well as the total 14C-activity in soil, plant ,seeds, insects and organs of birds were determined by combusting a definite weight of the sample (100-250 mg) in a Harvey Biological Oxidizer (Model OX- 600),followed by counting the liberated 14CO2 in a liquid Scintillation Counter. The internal standard technique was used for quench correction.

RESULTS

Plants

Table 1 shows the level of extractable and non-extractable (bound) 14C-chlorefenvinphos residues in maize and soybean plants of the ecosystem at different intervals. After the first spray, the total 14 C-residues in the whole plant amounted to about 15.5 and 40.5 µg/g for maize and soybean plants, respectively. After the second spray, this value increased to 20.4 µg/g in maize and 48.5 µg/g in soybean plant. The total recovery of extractable and non-extractable represented about 81-96 % of the total residues of the whole plant. Near maturity, the 14C-residues in maize and soybean plants showed a drastic reduction (1.6 and 1.0 µg/g) representing about 8--2% of the total residues on the whole plant after the second spray, respectively(Table1).The amount of bound residues (non-extractable) showed a significant increase with time. After 60 days from the first spray, the leaves of maize which are usually used as fodder for animals have about 0.70 µg/g (3.4%) of the total residues after the second spray.

Seeds

Soft green seeds of maize ears had a radioactivity corresponding to 0.74 µg/g. Roasting of these seeds led to a slight decrease in the chlorfenvinphos 14C-residues (0.17 µg/g , 23 % decrease) .The residues in mature dry soybean seeds (2.70 µg/g) were almost 6 times higher than those in the target crop maize (0.44 µg/g) (Table2) . The binding capacity of 14C-chlorfenvinphos in maize seeds represented 50% whereas in case of soybean it was about 20% of the total residues in seeds.

TABLE. 1. 14C- Chlorfenvinphos residues in maize and soybean plants.

Sampling days	Sample	^{14}C-Residues in plants			
		Total µg/g	Extractable µg/g	Bound µg/g	Recovery %
3	A	15.5±0.40	12.50±0.02	0.12±0.02	81.40
	B	40.5±0.50	37.00±0.35	1.10±0.02	94.07
24	A	8.10±0.91	6.40±0.80	0.70±0.09	87.65
	B	17.2±0.36	13.50±0.80	3.00±0.09	95.93
26	A	20.40±0.30	15.60±0.50	0.85±0.40	80.60
	B	48.50±0.70	41.70±0.40	3.90±0.20	94.02
60	C	0.70±0.03	0.54±0.02	0.10±0.01	91.40
	B	7.43±1.00	5.35±0.90	0.65±0.40	80.75
75	A	1.60±0.04	1.00±0.02	0.35±0.03	84.37
	B	1.00±0.03	0.70±0.22	0.20±0.01	90.00

TABLE 2. ^{14}C- Chlorfenvinphos residues in dry Seeds

Seeds	^{14}C-Residues			
	Total µg/g	Extractable µg/g	Bound µg/g	Recovery %
Maize	0.44 ±0.17	0.20 ±0.01	0.22± 0.03	95.45
Soybean	2.70 ± 0.24	2.05 ± 0.10	0.50±0.05	94.44

Data mean of 6 samples.

Soil

Samples of soil analyzed after the first and second

sprays showed that the major portion of 14C-activity was concentrated in the upper zone layer (0-5 Cm) (Table 3). The extractable residues were found to decrease with time whereas the extent of binding increased. A trace of 14C-residues was detected in the zone (5-10cm) after 24 days of the first spray which increased by time and reached its maximum value 0.09 µg/g after 75 days.

Beetles

Samples of carabidae beetles collected and analyzed prior 24 days following the first spray had comparatively low chlorfenvinphos residues, whereas after the second spray the highest residue was determined (1.28 µg insecticide/g beetle). These results were obtained from alive beetles (Table 4). It is worthy to mention that almost all beetles were alive. About 30% of 14C –residues decreased in beetles after 75 days of the first spray of the insecticide.

Earthworm

An increased accumulation of chlorfenvinphos residues with time was detected in earthworm during experimental period (Table 4) .The highest level residue (3.25µg/g earthworm) was detected 75 days following the first spray of the insecticide. No mortality was detected among earthworm during the period of the experiment.

Birds:

The number of birds was monitored during the period of the experiment. The birds were dissected four hours after death and their organs were analyzed for 14C –residues by combustion. The distribution of 14C-residues among the different organs such as liver, kidney, brain, heart, stomach and intestine of dead and alive birds is shown in Table 5. The determined radioactivity in these organs increased progressively after spraying and decreased thereafter within a few days. Some birds died during the days following the first and the second spray. The highest level of chlorfenvinphos residues in these dead birds was determined in liver and brain which amounted to 1.33--2.24 and 0.91--1.12 µg chlorfenvinphos /g organs, respectively. In surviving birds, the 14C-residues were as low as 0.1µg chlorfenvinphos /g organ after 75 days of the first spray. (Table 5). On the other hand, feces of birds contained considerable radioactivity equivalent to about 3.08 µg chlorfenvinphos/g feces after 24 days. At the end of the ecosystem experiment, the residues in feces amounted to 2.24ug/g. The major part of these residues was methanol soluble materials (67%).

TABLE 3. ^{14}C-Chlorfenvinphos Residues in Soil Column of the Ecosystem

Sampling days	^{14}C-residues (µg / g) 0 – 5cm Total µg/g	Extractable µg /g	Bound µg/g	Recovery %	5-10 cm Total µg/g	Extractable µg /g	Bound µg /g	Recovery %
3	0.91 ±0.025	0.87 ±0.015	0.036 ±0.005	99.6	N.D	N.D	N.D	N.D
24	0.56 ±0.004	0.46 ±0.002	0.056 ±0.001	92.0	0.024 ±0.005	0.02 ±0.001	0.002 ±0.001	91.7
26	0.83 ±0.010	0.66 ± 0.090	0.10 ±0.020	91.6	0.04 ±0.003	0.032 ±0.002	0.004 ±0.002	90.0
60	0.25 ±0.03	0.19 ± 0.050	0.04 ± 0.030	92.0	0.06 ±0.020	0.048 ±0.010	0.012 ±0.010	92.0
75	0.12 ±0.03	0.086 ± 0.020	0.024 ± 0.001	91.7	0.09 ±0.001	0.063 ±0.001	0.018 ±.003	90.0

TABLE 4. ^{14}C- Chlorfenvinphos Residues in Beetles and Earthworms of the Ecosystem

Insectes	Sampling (days) 3[a]	24	26[b]	60	75
Beetles	0.92±0.014	0.35±0.011	1.28±0.077	0.80±0.10	0.66±0.012
Earthworms	1.19±0.02	1.95±0.02	2.75±0.01	2.75±0.01	3.25±0.20

a. After the first spray b. After the second spray

Data Mean of 6 of the insects

TABLE 5 :^{14}C-Chlorfenvinphos residues in birds tissues of the ecosystem

Birds	^{14}C-Residues in organs(µg/g)

Sampling (days)			Liver	Kidney	Brain	Heart	Stomach	Intestine
3	2	Dead	1.33 ± 0.01	0.51 ± 0.03	.0.91 ±0.5	0.45 ± 0.02	0.55 ± 0.04	0.47 ± 0.01
	2	Alive	0.65 ± 0.03	0.063± 0.01	0.52± 0.04	0.70 ±0.06	0.35 ± 0.0 1	0.37± 0.02
24	2	Alive	0.56±0.053	0.050±0.015	0.45±0.014	0.56±0.043	0.48±0.07	0.32±0.05
26	2	Dead	2.24±024	0.72±0.01	1.12±0.015	0.58±0.01	0.71±0.01	0.62±0.04
	2	Alive	0.39±0.034	0.56±0..036	0.54±0.01	0.38±0.026	0.37±0.03	0.31±0.01
60	2	Alive	0.22±0.026	0.44±0.02	0.11±0.01	0.26±0.01	0.25±0.01	0.29±0.02
75	2	Alive	0.11±0.017	0.20±0.01	0.10±0.02	0.14±0.05	0.11±0.02	0.15±0.01

Determined by combustion: mean of 6 samples
The identification of ^{14}C-residues isolated from dry soil and mature seeds was achieved by TLC

Chromatographic analysis:

Chromatographic analysis of dry seeds and soil revealed the presence of the insecticide chlorfenvinphos [1] and three of its metabolites, 2,4-Dichloroacetophenone[11], 2,4-Dichlorophenyl)ethan-1-ol[111] and Desethyl Chlorfenvinphos [1V] as shown in table 6.

TABLE 6 . R_f – Values of chlorfenvinphos and its main metabolites in dry seeds and soil.

Compound	R_f		
	Sys.1	Sys 2	Sys.3
Chlorfenvinphos [1]	0.63	0.25	0.29
2,4-Dichloroacetophenone[11]	0.70	0.55	0.58
2,4-Dichlorophenyl)ethan-1-ol[111]	0.56	0.3	0.49
Desethyl Chlorfenvinphos [1V]	0.01	0.0	0.05

System 1 Ethanol: Benzene (1:9) System 2 Acetone: Hexane (2:8)
System 3 Dichloromethane: Acetonitrile: n-Hexane (3:3:8 v/v/v)
Visualization : Hanes – Isherwood reagent

DISCUSSION

There is a paucity of large-scale field investigations on the effects of organic toxicants on stream macro invertebrate community structure and ecosystem functions. Pesticide stress was associated with a decrease in the relative abundance and number of sensitive species in the communities. For each agrochemical a sound risk assessment should be performed. This should take in consideration not only the parent compound but also its toxicologically relevant degradation products. The present study describes a strategy for assessing and generating data upon the fate and dissipation of chlorfenvinphos in soil and plants, earthworms, beetles, and birds in a terrestrial ecosystem using radio labelled field studies.

The concentration of chlorfenvinphos14C-residues and its metabolites in maize and soybean plants was found to decrease with time. The major loss of the insecticide was caused by volatilization and reached its maximum value at the end of the experiment (about 2and7% for soybean and maize plants). These data are in agreement with other reported findings on potato, cabbage and maize plants by Beynon et al [25] and rape plants by Czaplicki et al. [26] after foliar application. The maize and soybean seeds harvested from plants grown in the ecosystem were shown to contain 14C-residues at a concentration of o.44 and 2.70 µg insecticide equivalent per gram seeds ,respectively(Table 3) .It is obvious that 14C-residues in case of soybean seeds was higher six times than in case of maize seeds . Upon foliar application of many organophosphorus insecticides such as 14C-pirimiphos—methyl[27] , 14C- chlorfenvinphos[28] and 14C-malathion [29], on soybean, maize and cotton plants, respectively, the aged insecticide residues are known to occur in seed and amounted to 2.25 µg/g,0.12 µg/g and 1.7µg/g under local agricultural practice .

Various pesticides rapidly degrade in the soil in a process called mineralization that turns pesticides into smaller compounds such as NH3, H2O, and CO2. Chemical reactions such as photolysis and hydrolysis lead to this degradation process. The main pathway for mineralization is usually the microbial metabolism and demolition. Some chemicals, for example [2, 4-dichlorophenoxyacetic acid], degrade fairly quickly in the soil, while others are degrade less easily [2, 4, 5-trichlorophenoxyacetic acid]. Other chemical compounds are very persistent and do not degrade gradually like atrazine[30,31].

Generally, Chlorfenvinphos degrades in soil, with a rate varying according to soil type, temperature, rate of application and moisture conditions. The data ob-

tained from the soil column of the ecosystem plot indicated that almost the total 14C-chlorfenvinphos was concentrated in the upper zone (0-5cm) after 3 days of the first spraying (Table4) .It was observed that the percentage of the extractable 14C-residues decreased with time while the bound 14C-residues increased. It is worthy to mention that traces of 14C-residues were detected in the second zone of soil column (5—10cm) after 24 days of the first spray .The metabolic fate of chlorfenvinphos in soils has a great similarity of that of other vinyl phosphate insecticides such as tetrachlorvinphos and dimethylvinphos in soil [32,33].

Pesticide metabolism in birds is an important process in which living organisms defend themselves from the toxic effects of foreign chemicals on their food resources. Within the organism, the chemical is converted to a less toxic form and excreted or retained in the organism. Many organs can be affected in the organism, especially the liver, depending on the chemical. Enzymes play an important role in metabolism and the presence of some enzymes, especially "mixed" oxygen in the liver, is currently used as an indication of the organism's contact with some foreign chemical compounds[34,35].

Decline in bird species has been reported over several past decades and often attributed to changes in farming practises, such as increase of agrochemical inputs, loss of mixture farming or unfarmed structures. Besides lethal and sub lethal effects of pesticides on birds, concern has recently focused on the indirect effects, such as reduction of food supplies and pesticides application. Evidences of this important indirect effect of pesticides has been reported, e.g., by Moreby and Southway[36]; Boatman et al[37]; Taylor et al[38].Inside the ecosystem, birds were found to feed on treated plants. Before maturity, the sheath of maize ears was also attacked, opened by birds and the soft grains were eaten by them. The distribution of 14C- activity among different organs of dead Egyptian house sparrows, during the period after the first and second spray (3 and 26 days) indicated that liver and brain contained the highest levels of the 14C-chlorfenvinphos residues .It is worthy to mention that the different organs in dead birds contained a higher level of the most toxic 14C-residues as compared with those still living in the ecosystem. Death of birds probably resulted from a severe inhibition of body esterase. The relatively high amounts of 14C-residues in feces of birds (up to 3.o8µg/g) indicate a high rate of excretion of the insecticide from exposed sparrow birds.

The greatest part of biomass of terrestrial invertebrates represent (60-80 %) and play an important role in soil ecosystem [39] .They are used as bio indicator of soil contamination providing an early warning of decline in soil quality. They serve as model organisms in toxicity testing.All carabida beetles collected during the ecosystem experiment were alive and had a comparatively low level of chlorfenvinphos residues. This may be attributed to the fact that beetles try to protect themselves against the insecticide by escaping to deeper levels in soil. It is obvious that the highest concentration of 14C-residues was observed in beetles after the second spray. These findings have been reported by other authors on carabid and spider by using lindane [40].

Earthworms have been widely recognized as improving soil properties including soil pH, soil structure and aeration[41], modifying the composition and activity of soil functional microorganisms [42], rending contaminants available for microbial degradation via bioturbation and burrowing[43]. These capabilities enable earthworms to effectively assist the bioremediation of soil organic pollutants [44]

The determination 14C-residues in earthworms indicate a gradual accumulation of 14C-residues with time. The maximum concentration of these residues amounted to 3.25µg insecticide/g earthworm. This level was detected after 75 days of 14C-chlorfenvinphos application on the ecosystem plants. The obtained data give an indication that earthworm represent an impotent source of contamination of terrestrial wildlife since they are characterized by high ability to cumulate a lot of pollutants such as chlorfenvinphos from soil in their tissues and thus they are used for studying the bioaccumulation potential of chemicals [45]. Similar finding was reported by Beyer et al [46, 47] in their study on long-term persistence of other organochlorine insecticides such as dieldrin and DDT in earthworm. A recent review of pesticides effects on earthworms reported on negative effects on growth and reproduction by several pesticides [48].

The mechanism for oxidative dealkylation of chlorfenvinphos proceeds initially via monooxygenation of the alpha -carbon atom of the alkoxy group to produce an unstable hemiacetal which breaks down by oxidative O –alkylation mechanisms to acetaldehyde and 2-chloro-1-(2,4-dichlorophenyl) vinylethylhydrogen phosphate. Acetophenone produced in the succeeding step is reduced to the alcohol. Some studies suggest that electrophilic metabolic intermediates or epoxides may be produced in the metabolism of

chlorfenvinphos [49, 50].

In conclusion

This study has shown that application of the insecticide 14C-chlorfenvinphos in a terrestrial ecosystem affected soil, plants, bettles, earthworms and one type of common bird in Egypt (Asfour Baladi) . In the plant, the major loss of the insecticide was caused by volatilization and reached its maximum value at the end of the experiment. 14C-residues in case of soybean seeds were higher six times than in case of maize seeds. .In birds, the distribution of 14C- activity among different organs of dead Egyptian house sparrows, during the experiment indicated that liver and brain contained the highest levels of the 14C-chlorfenvinphos residues .It is worthy to mention that the different organs in dead birds contained a higher level of the most toxic 14C-residues as compared with those still living in the ecosystem. The determination 14C-residues in earthworms indicate a gradual accumulation of 14C-residues with time. This indicated that earthworm represent an important source of contamination of terrestrial wildlife since they are characterized by high ability to cumulate a lot of pollutants such as chlorfenvinphos from soil in their tissues and thus they are used for studying the bioaccumulation potential of chemicals Carabida beetles were alive and had a comparatively low level of chlorfenvinphos residues. This may be attributed to the fact that beetles try to protect themselves against the insecticide by escaping to deeper levels in soil.

In the fact, pesticide application has become great threat to human health. Studies have shown that long - term low dose exposure to pesticides leads to the development of many diseases. Pesticide poisoning is more significant in developing countries. Pesticide residues affect environmental quality.

Conflicts of interest

The authors confirm that there is no conflict of interest.

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