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Effect of Moisture and Organic Manure on Persistence of Flubendiamide in Soil

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Abstract Persistence of flubendiamide in soil as affected by moisture and organic manure was studied. The present study reports persistence of flubendiamide [N^2-{1,1-dimethyl-2-{methylsulfonyl) ethyl}-3-iodo-N^1-{2-methyl-4-{1,2,2,2-tetrafluoro-1 (trifluoromethyl) ethyl} phenyl}-1,2-benzene dicarboxamide] in a sandy loam soil. Dissipation of the pesticide followed mono-phasic first order kinetics. The persistence of flubendiamide was more in dry soil followed by field capacity and submerged condition with half life values of 150.5–158.4 days for submerged soil, 177.0–181.1 days for field capacity soil and 206.6–215.0 days for dry soil. It was found that there is slight effect of fortification level on dissipation of flubendiamide in soil. In all the cases i.e. dry, field capacity and submerged condition dissipation was slightly slower at 10 μg g^{-1} level. Amendment of organic manure (2.5%) to the soil enhanced the degradation of the insecticide, and the half-life values in field capacity and submerged soils were 155.1 and 130.8 days, respectively.

Keywords Flubendiamide · Persistence · Amendment · Organic manure · Dry · Field capacity · Submerged

Accurate predictions of pesticide residues in soil are critical for the development of sustainable plant protection and pest management practices. In particular, the persistence of residues in soil has both economic and environmental significance and is often used as a key indicator for the environmental impacts of pesticide use. The persistence of pesticides in soil depends on intrinsic chemical properties and extrinsic environmental factors, mostly soil properties and climatic conditions. Intrinsic pesticide properties, such as vapour pressure, water solubility, biological activity and resistance to chemical changes, indicate the tendencies of pesticide fate, while the extrinsic factors, such as soil microbial activity, soil organic carbon content, soil temperature, solar radiation and rainfall can greatly modify the pesticide fate (Rao and Davidson 1980).

Flubendiamide, N^2-[1,1-dimethyl-2-(methylsulfonyl) ethyl]-3-iodo-N^1-[2-methyl-4-[1,2,2,2-tetrafluoro-1-(trifluoromethyl) ethyl]phenyl]-1,2-benzene dicarboxamide, belongs to phthalic acid diamide group (Fig. 1). Flubendiamide is mainly effective for controlling lepidopteron pests including resistant strain in rice, cotton, corn, grapes, other fruits and vegetables (Tohnishi et al. 2005). Flubendiamide activates ryanodine sensitive intracellular calcium release channels (ryanodine receptors, RyR) in insects. It has a novel biochemical action as it affects calcium ion balance irrespective of sodium or potassium ion balance which causes contraction of insect skeletal muscle (Masaki et al. 2006).

Information of flubendiamide fate in the environment is mainly restricted to registration documents. According to USEPA, flubendiamide is stable to hydrolysis under laboratory conditions, but direct aqueous photolysis appears to be a main route of degradation. Flubendiamide degrades to des-iodo flubendiamide under field soil photolysis with a half life estimated as 11.56 days. Flubendiamide degrades to des-iodo flubendiamide under laboratory soil photolysis with a half-life estimated as 135.5 days. It also degrades very slowly in field condition (USEPA 2008). Cavoski et al. (2009) in his experiment stated that it is not only stable both under aerobic-anaerobic soil conditions and

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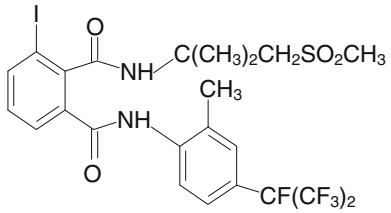

Fig. 1 Chemical structure of flubendiamide

aerobic-aquatic laboratory conditions but also it degrades in field condition very slowly. Australian Pesticides and Veterinary Medicines Authority (2009) reported that photolytic breakdown of flubendiamide on a soil surface gave calculated half-lives of 11.4 days.

Flubendiamide has been registered in India for rice and cotton cultivation; however, no information is available on flubendiamide degradation behavior under Indian subtropical conditions till date. Therefore, experiments were carried out to investigate the effect of moisture and organic manure on persistence of flubendiamide in soil.

Materials and Methods

Soil required for the study was collected from the plough layer (0–15 cm depth) of the research farm of Indian Agricultural Research Institute, New Delhi, India, with no history of pesticide application. It was air-dried in the shade, ground, sieved through a 2 mm mesh screen. The physico-chemical properties of the soil (type inceptisol) were: pH 8.18, organic carbon 0.16% measured by using the Walkley and Black (1965) method, clay 8.33%, sand 72.12%, silt 19.55% measured by employing the Bouyoucos hygrometer (1967), texture sandy loam and field capacity moisture content 20%. The organic manure was obtained locally. The physicochemical characteristics of the organic manure were a pH of 6.7 and an organic carbon content of 24.6%. The total carbon, nitrogen and hydrogen contents of manure were determined by elemental analysis and were 24.9, 10.2 and 3.1%, respectively. Analytical grade flubendiamide was obtained by extraction from the formulation (Fame 480 SC) provided by Bayer India Ltd (New Delhi, India). It was re-crystallized from methanol to obtain the analytical standard. The purity of the compound was checked by its melting point (220°C) and confirmed by TLC. The structure was characterized by NMR and confirmed by LC–MS fragmentation pattern with literature values. The stock solution of flubendiamide of 1,000 mg L^{-1} was prepared in acetonitrile and stored at 4°C. Working standards were prepared by appropriate dilutions. Organic solvents like acetone, dichloromethane and methanol were glass distilled before use. Buffer tablets were procured from M/s Qualigen India. Sodium sulphate was washed with acetone and then activated at 110°C for 4 h before use. HPLC grade solvents were procured from Merck India Ltd. These were filtered and de-gassed prior to use. The amount of water required to bring the air dry soil to field capacity moisture level was determined by placing 100 g of dry soil in a 100 ml capacity measuring cylinder. The cylinder was gently tapped to give proper packing and the bulk density was calculated. Water (10 ml) was then added carefully without disturbing the soil layer. The cylinder was capped with aluminium foil and kept undisturbed. The level of wet soil (volume) was recorded at intervals of time 15 min and the maximum value was taken for calculation. From the bulk density and volume of wetted soil, the amount of water required to wet 100 g of dry soil i.e. the field capacity was calculated. The field capacity of the test soil was found to be 20.2%.

To investigate the effect of different moisture regimes on persistence of flubendiamide in sandy loam soil study was carried out at two concentrations, 1.0 and 10.0 μg g^{-1}, under dry, field capacity and submerged. To fortify the soil at 100 μg g^{-1} level, 200 g soil was taken in a beaker and 20 ml of standard solution of flubendiamide (1,000 μg g^{-1}) was added. A further volume of acetone was added to completely wet the soil, which was stirred with a glass rod to give uniform mixing. The contents were left undisturbed overnight to allow the solvent to evaporate. The soil was then again mixed with a glass rod. This fortified soil was serially diluted with untreated soil to give 10.0 and 1.0 μg g^{-1} levels of fortification. For the 10.0 μg g^{-1} treatment, 1,080 g of untreated soil was taken in a polythene bag and 120 g of fortified soil (100 μg g^{-1}) was added and thoroughly mixed to achieve homogeneity. For the 1.0 μg g^{-1} treatment, 10.0 μg g^{-1} treated soil was mixed with untreated soil in the ratio of 1:9. The homogeneity of the treated soil was checked by drawing three samples from each treatment and analyzing for flubendiamide residues. As there was little variation in the residues among replicates, each treated soil was assumed homogeneous. The treated soil (10 g) along with untreated control samples were transferred to beakers. For treatments under field capacity moisture regime, the calculated amount of water was added to bring the soil to field capacity moisture level, while in case of submerged condition, enough water was added to raise the level of water to about 3 cm above the soil surface. No water was added in dry treatments. The beakers were closed with aluminium foil and constant weight of the beakers was maintained throughout the experiment by replenishing the lost water every alternate day. The beakers were placed at 25 (±1) °C temperature and about 90% relative humidity. Samples in triplicate, i.e. three beakers per fortification level per moisture regime, were withdrawn at 0, 3, 5, 7, 10, 15, 30, 50, 60 and 90 days

interval along with control. Triplicate samples removed for analysis by high-performance liquid chromatography (HPLC). Soil samples kept at field capacity and submerged conditions were transferred to a 150 ml stoppered conical flask and 50 ml of acetone was added. The samples were stirred with glass rod and kept for 30 min with intermittent shaking. The contents were filtered using Whatman filter paper No. 1. Soil was transferred back to beaker and re-extracted two more times using fresh 50 ml acetone each time. Acetone extracts were pooled and concentrated using rotary evaporator. The concentrated extract was diluted with saturated sodium chloride solution (100 ml, 10%), transferred to separatory funnel, exchanged into dichloromethane (3 × 30 ml). The combined dichloromethane extract was passed through anhydrous sodium sulfate. The extract was evaporated to dryness using a Kuderna-Danish rotary evaporator and the residues dissolved in HPLC grade acetonitrile for analysis. Samples kept under air dry condition was mixed with 0.2 ml of ammonia solution (25%, specific gravity 0.91). The addition of ammonia improves the extraction efficiency by desorbing the adsorbed pesticide from the soil. After about 2 h, activated Florisil (0.5 g) were added to the soil and thoroughly mixed. The mixture was dry in a glass column containing a layer (*c* 2 cm) of anhydrous sodium sulfate at the bottom. The column was eluted with about 125 ml of a mixture of acetone + hexane (5 + 95 by volume) and the extract concentrated using a Kuderna-Danish evaporator. The residues dissolved in HPLC grade acetonitrile for analysis.

To investigate the effect of organic manure on persistence of flubendiamide in sandy loam soil study was carried out at 10.0 μg g^{-1}, under two moisture regimes viz field capacity and submerged. To fortify the soil to get 10.0 μg g^{-1} fortification level above described procedure was followed. Soil samples (10 g) in field capacity and submerged condition, with 2.5% organic manure were prepared as mentioned in the previous section. Experiments were carried out in triplicate. All the beakers were placed in BOD incubator at 25 ± 1°C temperature and about 90% relative humidity was maintained. Constant weight of the beakers was maintained throughout the experiment by replenishing the lost water every alternate day. Samples were withdrawn at 0, 3, 5, 7, 10, 15, 30, 50, 60 and 90 days interval, triplicate samples removed for analysis by high-performance liquid chromatography (HPLC). Residues obtained from the various experiments were dissolved in acetonitrile for quantitative analysis. Estimation of flubendiamide was carried out by HPLC (high performance liquid chromatography) system (Merck-Hitachi)—Consisting of a L-7100 (computer operated dual pump), a L-7400 (UV detector) and a L-7200 (Auto sampler), HPLC column (30 cm)—Lichrospher, RP-18 (5 μm) was used for analysis. A mixture of acetonitrile–water (70:30, v/v) was used as the mobile phase, with a flow rate of 0.5 ml min^{-1}. The injection volume was 10 μL and the wavelength was set at 210 nm ($\lambda_{max,}$ determined by using spectrophotometer. Flubendiamide eluted at 10.1 min under these conditions. The limit of detection (LOD) was 0.01 μg g^{-1} for the analysis of these samples. A representative HPLC chromatogram showing well resolved peak of the flubendiamide is presented in Fig. 2.

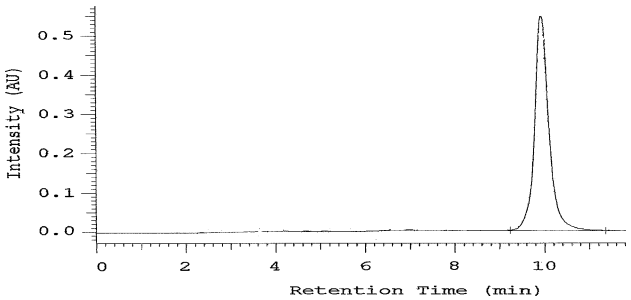

Fig. 2 HPLC chromatogram of flubendiamide

The residues were calculated on air dry soil weight basis. Residues data were subjected to regression analysis. Half life was calculated based on first-order dissipation kinetics:

$C = C_0e^{-kt}$ or $\log C/C_0 = -K_{obs}t$, Where, C = concentration after time t, C_0 = apparent initial concentration of flubendiamide (μg g^{-1}) and K_{obs} = the rate constant of the reaction. In all the cases, a first order equation provided a satisfactory fit for the data ($r > 0.9$).

Results and Discussion

Following treatment of dry soil, mean initial deposits were 0.99 and 9.92 μg g^{-1} at 1.0 and 10 μg g^{-1} fortification level. Residues declined slowly with time and at 10 μg g^{-1} level of fortification were 9.5, 8.95, 8.27 and 7.57 μg g^{-1} on 10, 30, 60 and 90 days respectively amounting to the loss of 4.5%–23.6% at 10 μg g^{-1} level (Table 1). At 1.0 μg g^{-1} fortification level residues of flubendiamide were 0.95, 0.89, 0.81 and 0.73 μg g^{-1} on 10, 30, 60 and 90 days respectively amounting to the loss of 4.0%–26.2% (Table 1). No residues/interfering peaks were observed in the soil of untreated control samples. The half-life values calculated from first order dissipation kinetics were 200.6 and 215.0 days at 1 μg g^{-1} and 10 μg g^{-1} fortification levels (Table 2).

Under field capacity mean initial deposits recorded were 0.983 and 9.91 μg g^{-1} at 1.0 and 10 μg g^{-1} fortification level. Residues declined slowly with time and were 9.38, 8.75, 7.89 and 7.01 μg g^{-1} on 10, 30, 60 and 90 days respectively at 10 μg g^{-1} level amounting to the loss of 5.3–29.2% (Table 1). At 1.0 μg g^{-1} fortification level,

Table 1 Persistence of flubendiamide in soil at 1.0 and 10.0 μg g^{-1} level

Days	Average residues (μg g^{-1}) ± SD*					
	Dry		Field capacity		Submerged	
	1.0 μg g^{-1}	10.0 μg g^{-1}	1.0 μg g^{-1}	10.0 μg g^{-1}	1.0 μg g^{-1}	10.0 μg g^{-1}
0	0.99 ± 0.01	9.92 ± 0.09	0.98 ± 0.06	9.91 ± 0.01	0.97 ± 0.05	9.9 ± 0.03
3	0.98 ± 0.07(0.5)	9.8 ± 0.02(1.2)	0.97 ± 0.03(1.3)	9.78 ± 0.08(1.3)	0.96 ± 0.06(1.8)	9.74 ± 0.06(1.6)
5	0.97 ± 0.04(2.0)	9.70 ± 0.11(2.2)	0.96 ± 0.08(2.3)	9.65 ± 0.03(2.6)	0.95 ± 0.01(3.1)	9.58 ± 0.08(2.9)
7	0.96 ± 0.02(3.0)	9.6 ± 0.87(3.2)	0.95 ± 0.01(3.3)	9.52 ± 0.06(3.9)	0.94 ± 0.03(4.1)	9.42 ± 0.09(4.5)
10	0.95 ± 0.03(4.0)	9.50 ± 0.06(4.5)	0.94 ± 0.02(4.3)	9.38 ± 0.09(5.3)	0.93 ± 0.05(4.9)	9.25 ± 0.01(6.5)
15	0.93 ± 0.06(6.0)	9.30 ± 0.07(6.2)	0.92 ± 0.06(6.4)	9.18 ± 0.08(7.3)	0.91 ± 0.08(7.2)	9.04 ± 0.07(9.4)
30	0.89 ± 0.07(10.1)	8.95 ± 0.05(9.7)	0.87 ± 0.09(11.1)	8.75 ± 0.07(11.7)	0.85 ± 0.09(12.7)	8.54 ± 0.08(14.4)
50	0.85 ± 0.01(14.1)	8.56 ± 0.11(13.7)	0.82 ± 0.07(15.2)	8.32 ± 0.03(16.0)	0.79 ± 0.01(19.4)	8.04 ± 0.03(19.4)
60	0.81 ± 0.09(18.1)	8.27 ± 0.03(16.6)	0.77 ± 0.01(21.0)	7.89 ± 0.08(20.3)	0.76 ± 0.06(25.5)	7.50 ± 0.06(24.9)
90	0.73 ± 0.05(26.2)	7.75 ± 0.07(23.6)	0.68 ± 0.03(30.8)	7.01 ± 0.08(29.2)	0.63 ± 0.02(35.5)	6.50 ± 0.12(34.3)

* Average of three replicates; figures in parenthesis shows % dissipation

Table 2 Regression equation and half-life for first order dissipation of flubendiamide

Treatment	Fortification (μg g^{-1})	Regression equation Y=	R^2 value	K values	$T_{1/2}$ (days)
Dry	1.0	−0.0015x − 0.006	0.995	0.0033	206.6
	10.0	−0.0014x + 0.981	0.914	0.0032	215.0
Field capacity	1.0	−0.0017x − 0.008	0.985	0.0039	177.0
	10.0	−0.0016x + 0.982	0.951	0.0038	181.1
Submerged	1.0	−0.0020x − 0.009	0.994	0.0046	150.5
	10.0	−0.0019x + 0.990	0.992	0.0044	158.4

0.94, 0.873, 0.776 and 0.68 μg g^{-1} residues were detected on 10, 30, 60 and 90 days respectively amounting to the loss of 4.3%–30.8% (Table 1). The half-life values calculated from first order dissipation kinetics were 177.0 and 181.1 days at 1 and 10 μg g^{-1} fortification levels (Table 2).

Under submerged condition the mean initial deposits recorded were 0.978 and 9.90 μg g^{-1} at 1.0 and 10 μg g^{-1} fortification level. Residues declined slowly with time and were 9.25, 8.54, 7.5, 6.5 μg g^{-1} on 10, 30, 60 and 90 days respectively amounting to the loss of ~6.5%–34.3% at 10 μg g^{-1} level (Table 1). At 1.0 μg g^{-1} level, 0.93, 0.856, 0.73 and 0.63 μg g^{-1} residues were detected on 10, 30, 60

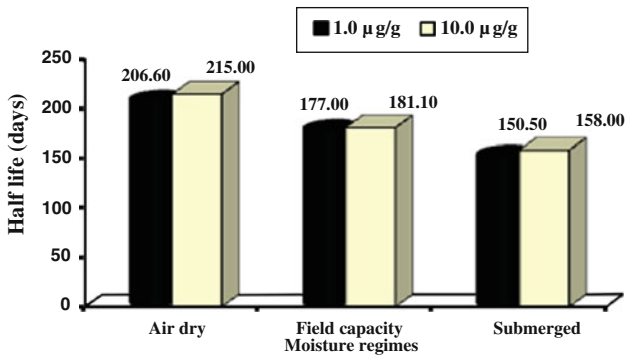

Fig. 3 Half-life of flubendiamide in sandy loam soil: effect of fortification level

Table 3 Residues of flubendiamide added to soil at 10.0 μg g^{-1} level under field capacity and submerged condition with 2.5% organic manure

Days	Average residues ± SD*	
	Field capacity with 2.5% organic manure (10 μg g^{-1})	Submerged with 2.5% organic manure (10 μg g^{-1})
0	9.90 ± 0.03	9.89 ± 0.05
3	9.89 ± 0.05 (0.1)	9.88 ± 0.01 (0.1)
5	9.8 ± 0.08 (1.0)	9.78 ± 0.02 (1.1)
7	9.71 ± 0.09 (1.9)	9.68 ± 0.07 (2.1)
10	9.58 ± 0.02 (3.2)	9.53 ± 0.06 (3.6)
15	9.36 ± 0.05 (5.4)	9.28 ± 0.02 (6.1)
30	8.70 ± 0.06 (12.1)	8.53 ± 0.03 (13.7)
50	7.82 ± 0.01 (21.0)	7.53 ± 0.05 (23.8)
60	7.38 ± 0.05 (25.4)	7.01 ± 0.07 (29.1)
90	6.03 ± 0.03 (39.0)	5.5 ± 0.02 (44.2)

* Average of three replicates; figures in parenthesis shows % dissipation

Table 4 Regression equation for first order dissipation of flubendiamide compost amended soil

Treatment	Fortification ($\mu g\ g^{-1)}$	Regression equation Y=	R^2 value	K value	$T_{1/2}$ (days)
Field capacity with 2.5% organic manure	10	−0.00194x + 1.005	0.994	0.0045	155.1
Submerged with 2.5% organic manure	10	−0.0023x + 1.006	0.992	0.0053	130.8

and 90 days respectively amounting to the loss of ~4.9%–35.5% (Table 1). The half-life values calculated from first order dissipation kinetics were 150.5 and 158.4 days at 1.0 and 10 μg g^{-1}fortification levels (Table 2).

Flubendiamide was found to persist longer under dry condition than field capacity and submerged moisture regime. The longer persistence and hence slower dissipation under dry condition could be attributed to negligible microbial activity in dry soil. Miles et al. (1984) has also reported slower dissipation of chlorfenvinphos in dry soil as compared to moist soil and attributed to lower microbial activity. The faster dissipation under submerged conditions could be attributed to partial anaerobic conditions. It seems that anaerobic microbes are more efficient in degrading flubendiamide than aerobic microbes. Smith et al. (1995) reported that cyfluthrin dissipated readily under anaerobic soil conditions. Faster dissipation under submerged condition has also been reported for pendimethalin (1992). It was found that there is slight effect of the fortification level on dissipation of flubendiamide in soil (Fig. 3). In all the cases i.e. dry, field capacity and submerged condition dissipation was slightly slower at 10 μg g^{-1} level. The half life values at 1 and 10 μg g^{-1} levels varied from 206.6–215.0, 177.0–181.1 to 150.5–158.4 days in dry, field capacity and submerged condition, respectively. The trend in dissipation at different levels of fortification could be attributed to the effect of the pesticide in soil microbes. Under field capacity and submerged moisture regimes, pesticide is lost by various processes such as biological (microbial) and chemical degradation and volatilization. Biological degradation has been found to be a major route of dissipation of pesticide, especially for synthetic pyrithroids (Chapman et al. 1981). Thus, at an increased concentration of pesticide, the microbial activity may be adversely affected, resulting in a decreased rate of dissipation.

Under 2.5% organic manure at field capacity moisture level the average initial deposits were 9.90 μg g^{-1}. Residues declined slowly with time and were 9.58, 8.7, 7.38, 6.03 μg g^{-1} on 10, 30, 60 and 90 days respectively amounting to the loss of 3.2%–39.0% at 10 μg g^{-1} level. Residue data for field capacity is presented in Table 3. The half-life values calculated from first order dissipation kinetics was 155.1 days at 10 μg g^{-1} fortification level (Table 4). Under 2.5% organic manure at submerged condition the mean initial deposit recorded was 9.89 μg g^{-1}. Residues declined slowly with time and at 10 μg g^{-1} level they were 9.53, 8.53, 7.01 and 5.51 μg g^{-1} on 10, 30, 60 and 90 days respectively amounting to the loss of 3.6%–44.2%. Residue data for field capacity is presented in Table 3. The half-life values calculated from first order dissipation kinetics was 130.8 days at 10 μg g^{-1} fortification level (Table 4).

Application of organic manure to soil enhanced flubendiamide degradation and half life values under field capacity and submerged condition were 151.1 and 130.8 days, respectively. Submerged condition causes anaerobiosis; therefore, soils become predominantly anaerobic in nature. Organic manure application to soil increases the organic carbon content of the soil, therefore, increases soil microbial activity. Increase in soil organic carbon content also causes greater sorption of flubendiamide and reduces the amount of flubendiamide in soil solution, which is available to soil microbes for degradation. However, despite greater retention of flubendiamide in organic manure amended soil, enhanced degradation of flubendiamide in soils can be attributed to increased soil microbial activity and more reduced soil conditions (higher redox potential). Probably, anaerobic microorganisms might be involved in the flubendiamide degradation.

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