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Air, Soil and Water Research

# Effect of Sunlight and Ultraviolet Light on Dissipation of Fipronil Insecticide in Two Soils and Effect of pH on its Persistence in Aqueous Medium

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**ABSTRACT:** In a laboratory investigation, the effect of natural sunlight and UV light exposure on dissipation of fipronil insecticide from two soils (clay loam and sandy clay loam) and the effect of pH on the persistence of fipronil in aqueous medium were studied. Dissipation of fipronil insecticide under sunlight followed biphasic first order kinetics in both soils. The half-life of the insecticide in sandy clay loam type soil was found to be 5.71 days for the first faster phase and 23.88 days for the second slower phase, whereas, in clay loam soil, the corresponding half-lives were 4.02 and 8.38 days, respectively. Under the UV light exposure, the dissipation of fipronil followed a single phase first order kinetics in both the soils with a half-life of 3.77 days in clay loam and 5.37 days in sandy clay loam, respectively. Residues of fipronil dissipated faster in clay loam than in sandy clay loam under both sunlight and UV lamp light. As compared to sunlight, dissipation was found to be faster under UV lamp light. Persistence of fipronil in aqueous medium under different pH conditions revealed that fipronil residues were below the limit of detection (LOD), $<0.05\ \mu g\ g^{-1}$, after 40 days of sampling at all the three pH. The dissipation of fipronil from aqueous medium increased with increasing pH from 5.0 to 9.0; the corresponding half-lives were 14.12, 9.83, and 6.76 days at pH 5.0, 7.0, and 9.0, respectively.

**KEYWORDS:** dissipation, fipronil, half-life, pH, residue, soil, sunlight, ultraviolet light

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## Introduction

Pesticides are used for the control of insect borne diseases and these are considered to be indispensable for production of an adequate food supply for the increasing world population.[1] Photodegradation is an important factor influencing the fate of pesticides in the fields.[2] Similarly, pH of the aquatic bodies influences the rate of degradation of pesticides due to hydrolysis reaction. It has been stated that pesticides persist longer under laboratory conditions as compared to field conditions[3,4] and the degradation of some highly persistent pesticides like DDT is faster in the presence of sunlight under field conditions than in the dark.[5] An understanding of the fate of a pesticide in soil is fundamental for accurate assessment of its environmental behavior and in ensuring the safe use of new and existing products.

Fipronil [5-amino-1-(2,6-dichloro-$\alpha,\alpha,\alpha$-trifluoro-*p*-tolyl)-4-trifluoromethyl-sulfinyl-pyrazole-3-carbonitrile] is a broad spectrum phenyl pyrazole class of insecticide discovered and developed by Rhoune Poulene in 1987 with principal trade names 'Regent' and 'Termidor.' It is active against a wide range of soil and foliar insects such as grasshoppers, rice skippers, vine weevil, termites, and black ants in agricultural, forestry, and pastoral land as well as in urban environment.[6–8] The insecticide is classified as a chiral pesticide and released into the environment as a racemic mixture. Fipronil interferes with the γ-aminobutyric acid (GABA)-gated channels[9]



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and disrupts normal nerve influx transmission (eg, passage of chloride ions) by targeting the GABA-gated chloride channel and at sufficient doses, causes excessive neural excitation, severe paralysis, and insect death.[10,11] Degradation of fipronil in soil results from exposure to sunlight at the surface to produce fipronil-desulfinyl, oxidation near the surface to yield fipronil-sulfone, hydrolysis throughout the upper layer to produce fipronil-amide and reductive processes below the surface that lead to production of fipronil-sulfide.[12] The effect of light and pH on dissipation of fipronil insecticide under the subhumid environment in India have not been reported so far and therefore, the present investigation was undertaken to study the degradation pattern of fipronil as influenced by light (UV and sunlight) and pH under the above conditions.

## Materials

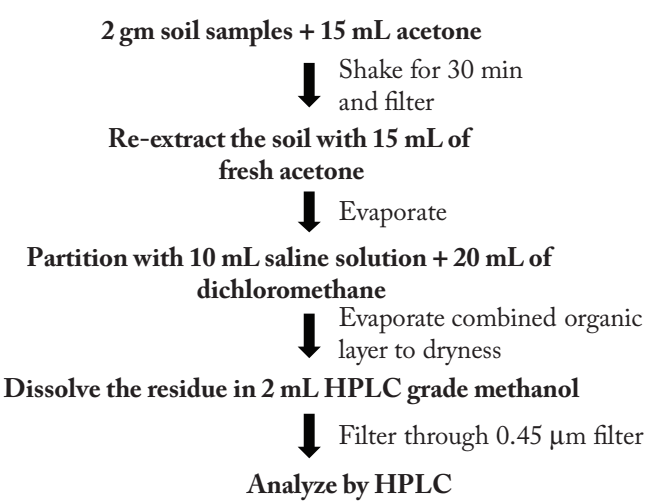
Technical grade fipronil of 98% purity was obtained from Gharda Chemicals Pvt. Ltd., Mumbai, India and its standard stock solution (1000 µg $L^{-1}$) was prepared in methanol. Chemicals and organic solvents used in the study were obtained from M/s Merck and M/s Spectrochem; organic solvents were glass distilled prior to use. Tris-buffer (tris-hydroxymethylamino methane) used for preparing buffers (pH 5, 7, and 9) was procured from M/s Finar Chemicals Ltd, India. Surface (0–15 cm) samples of clay loam and sandy clay loam were obtained from the collections of Department of Soil Science, GB Pant University of Agriculture and Technology, Pantnagar, India. Clay loam soil had 5.36 pH and 0.927 dS $m^{-1}$ electrical conductivity in 1:2 soil water suspension and 13.91 g organic C per kilogram soil while sandy clay loam soil had 7.34 pH and 0.180 dS $m^{-1}$ electrical conductivity in 1:2 soil–water suspension and 3.45 g organic C per kilogram of soil.

## Methods

The soil samples were dried in shade, crushed with a wooden roller, and passed through a sieve having an opening of 2 mm diameter and stored in polyethylene bags. For dissipation study, 10 g air dry soil of each clay loam and sandy clay loam were taken in pre-weighed beakers and 10 mL of 10 mg fipronil per liter of standard stock solution of fipronil was added to each beaker. 20 mL of acetone was added to each soil and mixed thoroughly for uniform distribution of insecticide. It was left undisturbed overnight till complete evaporation of acetone occurred, then it was mixed with 90 g of untreated soil to get a uniform fortification level of 10 µg fipronil per gram of soil. 2 µg of each fortified soil sample was transferred to pre-weighed petri-plates (5 cm internal diameter) in series and 0.36 mL of water was added to sandy clay loam and 0.5 mL of water to clay loam soil to bring the soil to field capacity moisture. One set of each fipronil fortified soil type was kept under UV light lamp (254 nm) or under sunlight for about six to seven hours daily. The moisture loss was made up every day by weighing the petri-plates. For monitoring the dissipation of fipronil, soil samples were drawn in duplicate on 0 (two hours after addition), 1, 3, 5, 7, 10, 15, 22, 30, 45, and 60 days after fortifications.

The fortified soil samples were transferred to conical flasks and 25 mL of acetone was added to dip the soil. The contents were stirred with a glass rod and kept on a shaker for 30 minutes with intermittent shaking. The contents were filtered using Whatman filter paper No. 1 and the soil was re-extracted using 20 mL of fresh acetone. Acetone extract was pooled and concentrated to 10 mL using rotary evaporator. The concentrated extract was then transferred to a separatory funnel and 10 mL of saline solution was added to it. Thereafter, 20 mL of dichloromethane was added to the combined extract and partitioning was done to collect the non-polar phase. The partitioned extract was evaporated to dryness using rotary evaporator and the residue was dissolved in 2 mL of HPLC grade methanol. Clean up of soil extract was done by filtering the extract through the column containing anhydrous sodium sulfate to remove the moisture content using methanol as an eluent. The eluted methanol was dried under the stream of nitrogen and re-dissolved in 2 mL mobile phase, which was passed through 0.45 µm millipore disk filter prior to injection in the HPLC system.

For studying the dissipation of fipronil from water at three different pH values, bulk solution of decimolar tris-hydroxymethyl aminomethane buffer was prepared in double distilled water and fortified with 10 mg fipronil per liter. The pH of 1.2 L solution was separately adjusted to 5, 7, and 9 using HCl or NaOH. The electrical conductivity of water was determined by using an EC meter and the hardness of water was determined by complexometric titration with EDTA.[13] The dissipation of fipronil was studied by drawing 25 mL portions in duplicate after thorough mixing at 0 (two hour), 3, 5, 7, 10, 15, 25, 40, 60, and 90 days after fortification. The minimum and maximum temperature during the whole experiment was 18°C and 35°C, respectively. The drawn water samples were transferred to separatory funnels and to each 30 mL of dichloromethane were added. The combined extract was diluted with 30 mL of 0.1% NaCl solution and shaken. The solution was






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**25 ml buffer solutions (pH 5, 7 and 9) in three separatory funnels**

↓ Add 30 mL of dichloromethane to each

**Dilute with 30 mL of 0.1% NaCl**

↓ Shake vigorously

**Partition with 25 mL of fresh dichloromethane**

↓ Organic layer evaporated to dryness

**Dissolve the residue in 2 mL of HPLC grade methanol**

↓ Filter through 0.45 µm filter

**Analyze by HPLC**

kept for some time to allow the layers to separate. The lower layer of dichloromethane was collected and again partitioned with 25 mL of fresh dichloromethane. The combined extract was evaporated to dryness. The residue was dissolved in 2 mL of methanol (HPLC grade) and filtered through 0.45 µm filter prior to HPLC analysis.

For quantifying fipronil in soil and water samples, Dionex (Ultimate 3000) HPLC system (model no. 8009589) of Thermofisher Scientific Pvt. Ltd., Mumbai, India, with variable wavelength UV detector was used. The operating conditions used were C-18, 5 µm, 250 × 4.6 mm column, methanol:water (85:15) mobile phase in isocratic mode at a flow rate of 1 mL/minute and UV detection at 276 nm. The retention time of fipronil under the above conditions was 4.5 minutes.

Recovery studies were also performed with soil and water samples fortified with fipronil (2 and 10 mg fipronil per kilogram of soil and 2 and 10 mg fipronil per liter of water) to establish the reliability of the analytical methods and the efficiency of extraction and clean up steps adopted in the present investigation. The limit of detection (LOD) and limit of quantification (LOQ) of fipronil were 0.05 and 0.12 mg $L^{-1}$, respectively. The recovery values ranged between 84.98 and 90.26% for clay loam and sandy clay loam soils, respectively. Recoveries of fipronil from water ranged between 86.35 and 93.76% at the two fortification levels.

## Results and Discussion

Fipronil concentration declined consistently with time in both the soils (clay loam and sandy clay loam soil) under both sunlight and UV lamp light (Table 1). Under sunlight, fipronil concentration in both the soils declined very fast during the first 10 days, and thereafter, the dissipation of fipronil occurred slowly and detectable concentration of fipronil persisted up to 30 days in clay loam and up to 60 days in sandy clay loam soil. On the other hand, under UV lamp light, the dissipation of fipronil occurred at a consistent rate and detectable concentration of fipronil could be detected up to 22 days only in both clay loam and sandy clay loam soils. The distribution of points in plots of logarithmic amount of fipronil recovered from both the soils under sunlight revealed that two straight line patterns (biphasic) were evident, while under UV radiation, only a single straight line pattern could be noted (figs. not shown). The data pertaining to the dissipation of fipronil in both soils under sunlight or UV were fitted to a first order kinetic equation:

$$C = C_0 e^{-\lambda t}$$

Where, $C$ is the amount of fipronil recovered from soil and water at time $t$, $C_0$ is the amount of fipronil recovered at $t = 0$,

**Table 1.** Persistence of fipronil in clay type and sandy clay loam soil kept in sunlight and UV lamp light.

| DAYS | SUNLIGHT | | U.V. LIGHT | |
|---|---|---|---|---|
| | CLAY LOAM (mg/kg) | SANDY CLAY LOAM (mg/kg) | CLAY LOAM (mg/kg) | SANDY CLAY LOAM (mg/kg) |
| 0 | 10.64 (0) | 6.91 (0) | 4.79 (0) | 7.62 (0) |
| 1 | 6.87 (35.44) | 5.58 (19.14) | 3.11 (35.13) | 5.59 (26.64) |
| 3 | 5.78 (45.69) | 3.49 (49.47) | 3.07 (35.85) | 5.44 (28.60) |
| 5 | 2.88 (72.88) | 2.73 (60.48) | 2.55 (46.76) | 3.65 (52.09) |
| 7 | 2.10 (80.26) | 2.44 (64.70) | 1.02 (78.78) | 2.23 (70.73) |
| 10 | 2.03 (80.88) | 2.05 (70.34) | 0.37 (92.31) | 1.47 (80.70) |
| 15 | 1.60 (84.91) | 2.00 (70.96) | 0.29 (93.98) | 0.72 (90.55) |
| 22 | 1.04 (90.27) | 1.91 (72.31) | 0.093 (98.06) | 0.52 (93.17) |
| 30 | 0.47 (95.60) | 1.78 (74.22) | N.D. | N.D. |
| 45 | N.D. | 1.18 (82.91) | N.D. | N.D. |
| 60 | N.D. | 0.54 (92.20) | N.D. | N.D. |
| 90 | N.D. | N.D. | N.D. | N.D. |

**Note:** Values in parenthesis show % dissipation, N.D. < 0.05 µg/g soil.



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**Table 2.** Calculated values for different constants involved in dissipation kinetics of fipronil under sunlight and UV light.

| PARAMETERS | SUNLIGHT SOIL TYPE | | | | U.V. LIGHT SOIL TYPE | |
|---|---|---|---|---|---|---|
| | CLAY LOAM | | SANDY CLAY LOAM | | CLAY LOAM | SANDY CLAY LOAM |
| | INITIAL RAPID PHASE | LATER SLOWER PHASE | INITIAL RAPID PHASE | LATER SLOWER PHASE | SINGLE PHASE | SINGLE PHASE |
| Rate constant (k, $day^{-1}$) | 0.172 | 0.082 | 0.121 | 0.029 | 0.183 | 0.128 |
| Half-life (days) | 4.01 | 8.38 | 5.7 | 23.88 | 3.77 | 5.37 |
| Coefficient of determination ($R^2$) | 0.901 | 0.984 | 0.907 | 0.909 | 0.950 | 0.950 |
| Regression equation | 0.0749x + 0.9439 | 0.0359x + 0.7644 | 0.0527x + 0.7721 | 0.0126x + 0.5599 | 0.0798x + 0.6367 | 0.056x + 0.8188 |

$\lambda$ is the degradation constant (in $day^{-1}$) and $t$ is time (days). The computed values of dissipation constant, half-lives, and coefficient of determination ($R^2$ values) are shown in Table 2. The computed values of coefficient of determination ($R^2$) for the dual dissipation pattern of fipronil in soils under sunlight varied from 0.901 to 0.984 for clay loam and from 0.907 to 0.909 for sandy clay loam soil; all $R^2$ values were statistically significant at $P \leq 0.01$ which demonstrated that both rapid and slow dissipation phases conformed to the first order kinetics. Under sunlight, the half-life values of fipronil for first and second phase of dissipation in clay loam soil were 4.01 and 8.38 days while for sandy clay loam the corresponding values were 5.70 and 23.88 days, respectively (Table 2). The computed values of dissipation rate constant for the first rapid phase was much higher than second slower phase in both the soils and on comparative basis, the clay loam soil having higher organic C content (13.91 g $kg^{-1}$ soil) maintained higher values of dissipation rate constants as compared to sandy clay loam soil (3.45 g $kg^{-1}$ soil). Interestingly, under UV lamp light, the values of insecticide residues in both soils when fitted to the first order kinetic revealed that dissipation of fipronil under UV light occurred through a single phase, which conformed to the first order kinetics ($R^2$ values = 0.950, significant at $P \leq 0.01$). Under UV light, the half-life values of fipronil were 3.77 days for clay loam and 5.37 days for sandy clay loam soil (Table 2). Just like under sunlight, under UV radiation too, the dissipation rate constant was higher for clay loam soil than sandy clay loam soil. The results of the present investigation clearly demonstrated that the effect of radiation source on dissipation of fipronil is partly due to the photolysis of fipronil under different irradiation[14] besides possible shifts in soil microbial communities.[15] Fitzmaurice and Mackenzie[16] reported that under natural soil conditions, three metabolites of fipronil (amide, sulfone, and sulfide forms) were detected while Raveton et al[17] noted that besides desulfinyl derivative, a wide range of photoproducts like 4-unsubstituted-, sulfide-, aniline-, and sulfone-derivatives were formed and some of these derivatives might dissipate faster to reduce the half-life of fipronil under UV light irradiation. The differences in photolytic products under natural sunlight and UV lamp light could result in differences in dissipation patterns and half-lives of fipronil in soils. Relatively faster dissipation of fipronil in clay loam soil than sandy clay loam soil especially, under sunlight during the later slower phase could be attributed to higher organic C content in the former soil (13.91 g C per kilogram of soil), which consequently enabled supporting higher microbial biomass C in clay loam as compared to sandy clay loam (3.45 g C per kilogram of soil). Fitzmaurice and Mackenzie[16] also recorded more rapid degradation of [$^{14}$C]-fipronil in a clay loam soil having 1093 µg microbial biomass C per gram of soil (41 g organic C per kilogram of soil) as compared to a sandy loam having 145 µg microbial biomass C per gram of soil (13 g organic C per kilogram of soil).

This difference in rate of dissipation of fipronil dissipation during initial faster and later slower phase under sunlight could be attributed to initial dissipation because of faster desulfuration at the 4-position of the pyrazole ring giving the desulfinyl derivative, which might accumulate due to strong bonding of metabolites in soil to slow down subsequent dissipation of parent compound during the later phase.

The persistence and percent dissipation values of fipronil in water at different pH values (5, 7, and 9) are shown in Table 3. Fipronil persisted till 40 days at all the three studied pH values

**Table 3.** Persistence of fipronil at three different pH values (5, 7, and 9).

| DAYS | PERSISTENCE (mg/L) | | |
|---|---|---|---|
| | pH 5 | pH 7 | pH 9 |
| 0 | 8.09 (0) | 9.65 (0) | 10.58 (0) |
| 3 | 6.80 (15.98) | 6.98 (27.63) | 6.24 (37.99) |
| 5 | 5.03 (37.75) | 5.05 (47.64) | 2.39 (76.22) |
| 7 | 4.45 (44.89) | 4.17 (56.80) | 1.57 (84.35) |
| 15 | 3.92 (51.47) | 2.80 (70.92) | 0.61 (93.89) |
| 25 | 3.07 (62.04) | 1.57 (83.75) | 0.46 (95.42) |
| 40 | 0.87 (89.16) | 0.48 (95.05) | 0.12 (98.83) |
| 60 | N.D. | N.D. | N.D. |
| 90 | N.D. | N.D. | N.D. |

**Note:** Values in the parenthesis show % dissipation N.D. < 0.05 mg/L.



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**Table 4.** Calculated values for different constants involved in dissipation kinetics of fipronil in water at three pH values.

| PARAMETERS | WATER pH | | |
|---|---|---|---|
| | 5.0 | 7.0 | 9.0 |
| K ($day^{-1}$) | 0.049 | 0.07 | 0.102 |
| Half-life (days) | 14.12 | 9.83 | 6.76 |
| Coefficient of determination ($R^2$) | 0.932 | 0.985 | 0.901 |
| Regression coefficient | 0.0213x + 0.7802 | 0.0306x + 0.8145 | 0.0445x + 0.6345 |

(5, 7, and 9). However, the percent dissipation of fipronil was the highest at pH 9, followed by pH 7 and pH 5. This could be ascribed to the fact that the fipronil molecule is relatively stable at acidic (5) and neutral pH (7.0) but undergoes rapid hydrolysis to form fipronil-amide[18] at alkaline pH (9.0), which has been reported to dissipate faster.[19] The data on fipronil residue in water at all three pH values also conformed to a monophasic first order kinetics ($R^2$ values = 0.901–0.985; all significant at $P \leq 0.01$, (Table 4)). The half-life values of fipronil in aqueous medium at 5, 7, and 9 pH were 14.12, 9.8, and 6.7 days, respectively. Feung and Yenne[20] also reported first order kinetics of fipronil in aerobic aquatic system with a half-life of 14.5 days.

## Conclusion

From the above results it was concluded that dissipation of fipronil under UV lamp light was higher than under sunlight and the dissipation of fipronil was faster in clay loam soil with higher organic C content as compared to sandy clay loam type soil under both sunlight and UV light. The dissipation kinetics of fipronil in soil kept under sunlight followed biphasic first order kinetics while the soil samples placed under UV lamp followed monophasic first order dissipation kinetics. The dissipation of fipronil from water occurred following a monophasic first order kinetics and the dissipation rate increased with increase in pH. After 40 days of sampling, the amount of fipronil residue was found to be non-detectable (ND), $<0.05\ \mu g\ g^{-1}$.

## Author Contributions

Conceived and designed the experiments: AS. Analyzed the data: AS and AV. Wrote the first draft of the manuscript: AV. Contributed to the writing of the manuscript: PCS. Agree with manuscript results and conclusions: AV, AS, SSC, PCS. Jointly developed the structure and arguments for the paper: AV, AS, SSC, PCS. Made critical revisions and approved final version: AS, PCS. All authors reviewed and approved of the final manuscript.

**DISCLOSURES AND ETHICS**

This paper was subject to independent, expert peer review by a minimum of two blind peer reviewers. All editorial decisions were made by the independent academic editor. All authors have provided signed confirmation of their compliance with ethical and legal obligations including (but not limited to) use of any copyrighted material, compliance with ICMJE authorship and competing interests disclosure guidelines and, where applicable, compliance with legal and ethical guidelines on human and animal research participants.

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