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Persistence Behavior of Fantac Biostimulant in Chili and Soil Under Subtropical Conditions

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Abstract The persistence of the biostimulant Fantac (*N*-ATCA) in chili and soil was evaluated at two application rates (0.25 and 0.50 μg g^{-1}) by high-performance liquid chromatography. At 0.25 μg g^{-1} Fantac persisted up to 3 days in soil and 5 days in chili. However, at 0.50 μg g^{-1} Fantac residues persisted up to 5 days both in soil and chili. The dissipation of the biostimulant from soil and chili appeared to occur in a single phase and conformed to first-order kinetics. Half-lives of Fantac in soil and chili were calculated.

Keywords Fantac · Persistence · Chili · Monophasic · Soil

Chili (*Capsicum annum* L. var. Pant C1) is one of the most valuable crops of India. Different varieties are grown for vegetables spices, condiment, sauces and pickles (Chaudhary 2000). For high yield of this crop, pesticides are used. As these crops invariably retain some residues, the potential health hazards posed by these consumable commodities depend on the quantity of residues present in them. There is, therefore, an urgent need to synthesize and commercialize those compounds which result in minimum or no residues in the agricultural produce at harvest time and at the same time increase the quality and the quantity of the crop yield.

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Fantac, a mixture of 5% *N*-acetyl thiazolidine carboxylic acid (*N*-ATCA) and 0.1% folic acid is one such biostimulant which at recommended rates and in conjunction with well-balanced fertilizers and pesticide programs, has shown to enhance crop yields, both in quantity and quality (Gupta and Macleod 1982). Due to the presence of thiol (–SH) group in Fantac, it provides an anti-stress effect regulating the osmotic equilibrium and enhancing the mitochondrial oxidative phosphorylation under stressing conditions (Direzione et al. 1996). It is used to stimulate seed germination, increase plant growth, improve fruit rating and increase yield on wide range of crops including wheat, corn, rice, sugar beet, potatoes, grapes, strawberry, apples, etc. (Thomas 1982). Fantac (also known as Quantum) when applied as foliar spray on grapes revealed an increase in yield and improve the characteristics of berry (Ramteke and Somkuwar 2004).

Persistence and mobility of agrochemicals are influenced by the agroclimatic conditions such as the soil environment, site conditions, weather, etc., as well as the methods of their application. Recommendations for the use of agrochemicals on a crop cannot be made until its persistence studies have been carried out as information on degradation rate helps to assess and predict their environmental behavior (Laskowski et al. 1983). The present investigation was carried out to determine the residues of Fantac in chili and soil by high-performance liquid chromatography (HPLC) with a view to ensure human and environmental safety.

Materials and Methods

A field trial of chili (cv. Pant C1) was conducted in a randomized block design with three replications during the

winter season of 2006–2007. The plot size was 2.75 m × 3.25 m and the interplant and inter row distance was 45 × 60 cm. The treatments were applied at 0.25 and 0.50 μg g^{-1} in a spray volume of 700 L solution ha^{-1} using a hand sprayer at 22 days after planting without any protection on plants. Two more sprays were given each at 15 days interval. For control treatment (0 μg g^{-1}), equivalent volume of plain water was sprayed on the same dates.

About 500 g fresh weight of whole green chili and surface soil (0–15 cm) were sampled randomly from each plot starting from the last day of spray at different time intervals viz. 0 (1 h after application), 1, 3, 5, 7, 12, and 15 days. Fruits of chili were chopped using Ni coated knife and mixed. Sub-samples (50 g) were then drawn randomly using quartering technique, kept in airtight plastic bags and stored in deep freezer (−20°C) until extraction.

The meteorological data comprising temperature, relative humidity, rainy days, number of bright sunshine hours and wind speed from the first spray to final sampling (28.9.06 to 16.11.06) are presented in Table 1.

Chopped samples (25 g) were mixed with 100 mL methanol in a wide mouth conical flask and shaken for 30 min on a mechanical shaker and subsequently filtered through a Buchner funnel and washed with methanol.

Soil sample (50 g), was also extracted in 100 mL of methanol for 30 min on a mechanical shaker, filtered through buchner funnel and washed with methanol. The combined filtrate was partitioned with isooctane/0.01% *t*-butanol in dichloromethane (1:4) and saturated sodium chloride solution in a separatory funnel. After phase separation, the organic layer was collected. It was then passed through anhydrous sodium sulfate for drying. The mixture was again partitioned and the pooled organic phase was evaporated to dryness in a rotary vacuum evaporator at 40°C. The residue left was dissolved in methanol–acetonitrile (1:1) and then purified. For purification, the column was packed with silica gel (60–120 mesh) using dichloromethane with activated charcoal at the top of the column. The sample solution in methanol–acetonitrile was added to the column and Fantac was eluted. The eluted solution was collected and immediately evaporated to dryness in a rotary vacuum evaporator. The residue was reconstituted in 2 mL acetonitrile–sodium dihydrogen orthophosphate (6:94) pH 2.5, solution for final HPLC analysis.

Fantac residues were determined by HPLC (a Beckman model 322) equipped with UV detector (210 nm). The column used was C_{18} (250 mm × 4.6 mm i.d.) and the mobile phase was acetonitrile–sodium dihydrogen orthophosphate (6:94) pH 2.5 at a flow rate of 1.0 mL min^{-1}. A 5-μL aliquot of each sample was injected every time for residue analysis. The retention time of Fantac under the above conditions was 10.5 min. A calibration curve was plotted between different concentrations of Fantac and peak area. The concentrations of Fantac in replicates of different samples were calculated with the help of the calibration curve. The mean value of the three replicates were computed. When the Fantac residues were below the limit of quantification (LOQ), the sample was spiked with a known concentration of Fantac and the difference of the two taken as the quantity of biostimulant present.

For validating the efficiency of the method, recovery studies were performed by spiking 25 g samples of chili and 50 g soil with Fantac at two concentrations (0.05 and 0.5 μg g^{-1}). The samples were extracted and cleaned up following the procedure described in preceding sections.

Results and Discussion

The percent recovery values of Fantac from soil and chili samples were found to be 90% and 92% at higher fortification rate and 82.6% and 85% at lower fortification rate, respectively. Standard deviation associated with the determinations ranged from 2.6% to 4.8% and the LOQ and LOD values for Fantac were 0.05 and 0.02 μg g^{-1}, respectively.

Fantac residues declined consistently with time in soil as well as in chili (Table 2). At the lower application rate

Table 1 Metereological data during the experimental period

Week	Temperature (°C)		Relative humidity (%)		Rain fall (cm)	Sunshine hours	Wind speed (km h^{-1})
	Max	Min	Morning	Evening			
24–30 September 2006	32.9	22.6	92	56	0.00	09.3	04.4
1–07 October 2006	33.3	21.1	90	51	0.00	09.5	04.7
8–14 October 2006	32.1	20.5	91	56	0.00	07.5	04.1
15–21 October 2006	28.8	17.2	91	52	13.6	07.4	02.9
22–28 October 2006	28.6	14.4	90	57	16.2	08.8	02.9
29–4 November 2006	29.1	14.3	88	44	0.00	08.1	02.7
5–11 November 2006	28.3	14.4	90	52	0.00	07.1	02.7
12–18 November 2006	27.1	13.1	92	52	0.00	05.4	02.7

Table 2 Persistence of Fantac in chili and soil

Days of application	Residues of Fantac (μg g^{-1})			
	Soil		Chili	
	0.25	0.50	0.25	0.50
0 (1 h)	0.23 (0)	0.45 (0)	0.23 (0)	0.46 (0)
1	0.08 (65.8)	0.22 (51.1)	0.14 (39.1)	0.21 (54.4)
3	0.04 (82.6)	0.10 (77.8)	0.05 (78.3)	0.09 (80.4)
5	N.D.	0.05 (88.9)	0.02 (91.3)	0.05 (89.1)
7	N.D.	N.D.	N.D.	N.D.
12	N.D.	N.D.	N.D.	N.D.
15	N.D.	N.D.	N.D.	N.D.

Values in the parenthesis show percent dissipation of the biostimulant
(N.D. < 0.02 μg g^{-1})

(0.25 μg g^{-1}) residues persisted in soil up to 3 days, whereas at the higher rate of application (0.50 μg g^{-1}) Fantac residue was detected up till 5 days. However, in the case of chili, irrespective of application rate, the Fantac residue remained detectable up till 5 days after application.

The data on the amount of Fantac recovered from soil and chili samples for both application rates, were fitted to a first order kinetic equation:

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

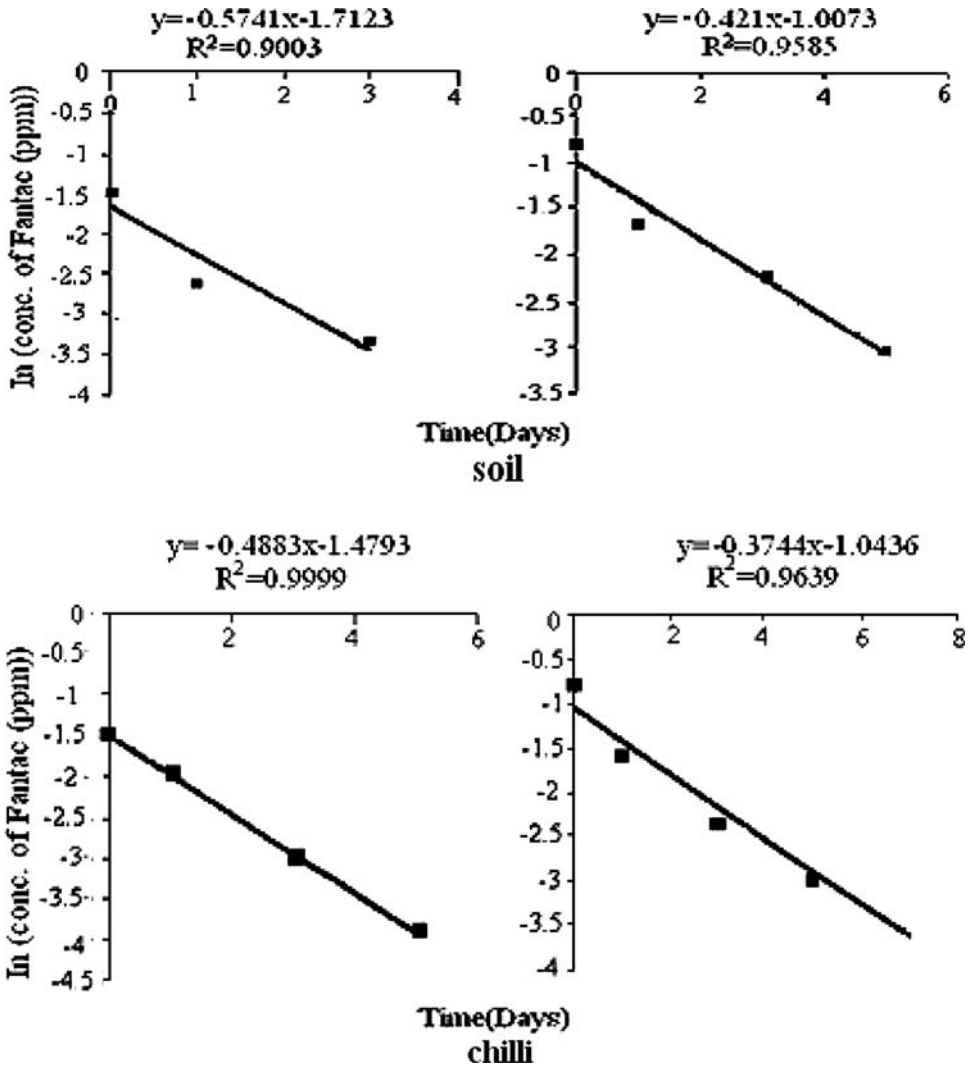

Fig. 1 Plots of natural logarithm of Fantac concentration in soil and chili versus time at 0.25 μg g^{-1} and 0.50 μg g^{-1}, respectively

Table 3 Computed values of degradation rate constant, half-life and correlation coefficient for Fantac in soil as per first-order kinetics

Computed values	Fantac application rate	
	0.25	0.50
Degradation rate constant (λ)	0.5741	0.4121
Half-life ($t_{1/2}$) (days)	1.20	1.68
Correlation coefficient (r)	0.9003	0.9585

where C is amount of Fantac recovered from the samples at time t. C_0 is amount of Fantac recovered at $t = 0$, λ is degradation constant and t is time in days.

For both the rates of Fantac application (0.25 and 0.50 μg g^{-1}), natural logarithm of Fantac residues were plotted against time (Fig. 1). The distribution of points for chili as well as soil at both the levels of treatment suggested that dissipation of Fantac occurs through a single distinct phase conforming to the first-order kinetics. The computed values of coefficient of determination (R^2) between log residues in soil, chili and time varied from 0.900 to 0.999 (all significant at $p = 0.05$) indicating that dissipation of Fantac could be accounted by the first-order kinetics. The half-life values of Fantac in different samples were calculated from the slope of regression equation.

As evident from Table 3, in soil, the half-life values of Fantac were 1.2 days for lower rate of application (0.25 μg g^{-1}) and 1.7 days for higher rate of application (0.50 μg g^{-1}). A relatively higher half-life value of Fantac in soil at 0.50 μg g^{-1} as compared to 0.25 μg g^{-1} in the present study could be attributed to possible deeper movement of Fantac in soil at higher rate of application. A combination of both degradative and dissipative mechanisms control overall persistence of chemicals in the natural environments. The fate of residues in the soil compartments is controlled principally by the primary mechanisms of degradation as well as adsorption–desorption characteristics (Cheng 1990). These aspects are influenced both by the physico-chemical characteristics of the chemical compounds and of the environmental matrix. Therefore, the fate of a compound in the environment is often predictable, if adequate knowledge of key properties of the chemical and matrix with which it is interacting are known (Mackay and Stiver 1991).

The half-life values of Fantac at 0.25 and 0.50 μg g^{-1} application rates (Table 3) were 1.42 and 1.9 days for chili. The possible routes of Fantac dissipation and transformation in the environment include biotransformation via soil microorganisms on soil and decarboxylation and photoconversion to simpler products on plant surfaces.

Thus, it can be concluded that the dissipation of Fantac both in chili crop and soil conformed to first-order kinetics. Fifty percent of Fantac residues in chili and soil dissipate in 1.42 and 1.2 days at lower rate of application (0.25 μg g^{-1}) and 1.9 and 1.7 days at higher rate of application (0.50 μg g^{-1}) under the sub-humid and subtropical climate.

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