

Original Research Article

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## Mitigation of Pendimethalin Residues as Influenced by the Organic Sources and Bioagents in Sandy Clay Loam Soil Grown with Greengram

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### ABSTRACT

#### Keywords

Dissipation, Green gram, Herbicide residues, Mitigation, Pendimethalin

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Herbicides use is increasing throughout the globe due to increasing labour cost, choice of application of herbicides, quick weed control in crop and non-cropped areas. Even though the persistence of pendimethalin may be shorter, depends upon the soil, weather and the crop for which its applied will vary in the pendimethalin residues. Hence, pot culture experiments have been carried out with an objective to evaluate various organics and bioagents for their efficiency to degrade the pendimethalin residues faster in the soil. Soil samples were analysed for pendimethalin residues. Residues of pendimethalin were extracted using methanol and determined by High Performance Liquid Chromatography (HPLC). It was found that the dissipation was faster under Farm yard manure (FYM), Arbuscular Mycorrhiza (AM) and biochar applied treatments and the slowest degradation was noticed in control during both the years. The lowest half life of 8.57 days (2018-19) and 8.32 days (2019-20) was observed in farm yard manure applied treatment. Application of farm yard manure at  $10 \text{ t ha}^{-1}$  or vermicompost at  $5 \text{ t ha}^{-1}$  is efficient in reducing the residual concentration of pendimethalin in green gram grown soil. Irrespective of the treatments the pendimethalin residues persisted up to 45 DAHA. On 60 DAHA residues became BDL in FYM and vermicompost applied treatments when compared to others

### Introduction

Herbicides have become one of the most important components for efficient weed control and in turn for increasing the agricultural production and to maintain the non-cropped area free from weeds and pests. Herbicides use is increasing throughout the globe due to increasing labour cost, choice of application of herbicides, quick weed control in crop and non-cropped areas. Globally consumption of herbicides is 44% followed

by the insecticides (22%), fungicides (27%) and others (7%). In India, herbicide use has increased to 30% during the last 10 years in managing weeds in the country (Sondhia, 2014). As herbicides are chemical in nature and thus excessive and repeated use may pose residue problems, phyto-toxicity to crop plants, residual effect on susceptible intercrops or succeeding crops or non targets organisms and ultimately health hazards due to accumulation of herbicide residues in the soil. In general, herbicides have been

formulated in such a way that they degrade from the environment after completion of their intended job of controlling weeds, but a few of the herbicides persist in the environment and pose a serious hazard to the succeeding crop and also to the soil environment. However, the herbicide should persist long enough to check weeds until the end of critical period of weed competition but should not persist beyond the crop harvest, as it would be injurious to the sensitive crops grown in rotation. A herbicide is said to be persistent when it may be found to exist in soil in its original or a closely related but phytotoxic form longer than one crop season after its original application (Sondhia, 2013).

Dinitroaniline herbicides are one of the most widely used herbicide groups and play a major role in managing the weed in the agricultural production systems. These herbicides are valued because of their low costs, moderate persistence, and long season weed control in agroecosystems; Pendimethalin, is used for selective control of complex weed flora in pulses, vegetables and oilseeds (Sondhia, 2012). The use of herbicides offers selective and economic control of weeds from the beginning and gives the crop an advantage of a good start and competitive superiority. However, the intensive utilization of these compounds may cause serious risks including environmental safety, water sources' pollution (Montiel-León *et al.*, 2019) as well as human and animal health and crop safety (Mehdizadeh, 2019). Every year in so many countries excessive amounts of soil-applied herbicides are used in agricultural lands; therefore, it seems that the need to provide safe and effective methods for herbicides removal and degradation from environment are very crucial. Nowadays, non-chemical and biological degradation is an important environment friendly strategy for detoxification and dissipation of herbicides

from environment (Zhang and Quiao, 2002). However, among soil characteristics, organic matter content is a key factor that has important effect on herbicide degradation and movement in the soil (Gamiz *et al.*, 2017). Application of organic-based fertilizers can improve edaphic qualities and change the fate of any herbicide (Marín-Benito *et al.*, 2019). Some researchers reported that the application of organic-based fertilizers under different soil environment, increases the herbicide degradation and movement in soil (Kravvariti *et al.*, 2010). Different studies showed that the addition of organic matter including manures, composts, and plant residues to soils is a widely accepted non-chemical approach for improving the soil characteristics as well as the different crop production (Pampuro *et al.*, 2017). Cox *et al.*, (2000) concluded that the infertile soils had a minimum effect on herbicide movement; therefore, the presence of organic matter can be considered as an important adsorbent for herbicides. However, the influence of different soil organic amendments on herbicide's fate is very complicated (Briceno *et al.*, 2007). Depending on the type of amendments and chemical structure of applied herbicide, the persistence and dissipation responses of these chemicals can be different (Umar *et al.*, 2012). Even though the persistence of pendimethalin may be moderate, depends upon the soil, weather and the crop for which its applied will vary in the pendimethalin residues. Hence, pot culture experiments have been carried out with green gram with an objective to evaluate various materials for their efficiency to mitigate the pendimethalin residues in the soil.

### **Materials and Methods**

The experiments have been conducted under All India Coordinated Research Programme on Weed Management, Department of Agronomy, TNAU, Coimbatore during kharif

seasons of 2018-19 and 2019-20. Soil samples were collected from the field which didn't receive the pendimethalin previously and used for conducting the pot study with the

following treatments. The soil used for the study is sandy clay loam in texture and the initial soil characteristics are furnished below.

**Initial physico-chemicals characteristics of the soil used for the study**

S.No.	Years	Initial Soil Characteristics						
		Texture	pH	EC dS m <sup>-1</sup>	OC %	Available N (kg ha <sup>-1</sup> )	Available P (kg ha <sup>-1</sup> )	Available K (kg ha <sup>-1</sup> )
1.	2018-19	Sandy clay loam	7.92	1.70	0.46	166.8	17.2	674
2.	2019-20	Sandy clay loam	7.81	1.52	0.36	167.2	19.5	542

Treatments were imposed three days before sowing of green gram (Co 8). On 3<sup>rd</sup> day after green gram sowing, the pendimethalin was

applied to each pot @ 1.0 kg ha<sup>-1</sup>. The details of the treatments as follows.

<b>T<sub>1</sub></b>	<b>Farm Yard Manure(FYM) @ 10 t ha<sup>-1</sup></b>	<b>T<sub>6</sub></b>	<b>Arbuscular Mycorrhiza(AM) @ 10 kg ha<sup>-1</sup></b>
<b>T<sub>2</sub></b>	Vermicompost @ 5 t ha <sup>-1</sup>	<b>T<sub>7</sub></b>	Pseudomonas @ 10 kg ha <sup>-1</sup>
<b>T<sub>3</sub></b>	Biochar @ 5 t ha <sup>-1</sup>	<b>T<sub>8</sub></b>	Urea @ 100 kg ha <sup>-1</sup>
<b>T<sub>4</sub></b>	Phosphobacteria @ 10 kg ha <sup>-1</sup>	<b>T<sub>9</sub></b>	Maize straw incorporation @ 5 t ha <sup>-1</sup>
<b>T<sub>5</sub></b>	Trichoderma @ 10 kg ha <sup>-1</sup>	<b>T<sub>10</sub></b>	Control (no manure/bioagents)

Soil samples were collected from a depth of 0-15 cm on 0, 1, 3,5,10, 15, 30 and 45 days after herbicide application (DAHA) and analysed for pendimethalin residues. Residues of pendimethalin were extracted using methanol and determined by High Performance Liquid Chromatography (HPLC). The sample was extracted with 100 ml methanol as extracting solution after shaking in orbital shaker for 1 hr. The suspension was filtered through whatman no.42 filter paper and the extract was evaporated in water bath till the suspension reduced to 10 ml. The concentrated extract was transferred to separating funnel and the pendimethalin residue was portioned with 30 ml of 1% NaCl and 50 ml of hexane. The upper organic layer was collected separately. The lower aqueous layer was re-extracted twice with 25 ml hexane each time. The organic layer was pooled and filtered through

whatman no. 42 filter paper containing a pinch (2 gm) of anhydrous sodium sulphate. The filtered residue extract was concentrated to dryness using rotary vacuum evaporator. The dried residues of pendimethalin compounds were dissolved in 2 ml acetonitrile HPLC grade solvent for HPLC determination.

**Instrumentation**

Pendimethalin residues were determined by HPLC (1200 series) with Diode Array Detector (DAD), binary pump and autosampler with Rheodyne injection system. The pendimethalin compound was separated using Agilent Eclipse XDB – C18, 5µm, 4.6x150mm column maintained at 30°C. The instrument was connected to a computer, which converts the signal response in to peak area using EZChrom software (USA).

Acetonitrile / water (80/20, v/v) was the mobile phase used for the determination. The injection volume of the sample was 20 $\mu$ L with the flow rate of 0.8mL min<sup>-1</sup> and detection at 227nm. Under the given instrumental conditions of HPLC, pendimethalin was resolved at 6.4minutes (Fig. 1) with a response time of 0.20 seconds.

### Method validation

The stock solution of pendimethalin and its working concentrations were prepared in acetonitrile by diluting the stock solution as described by Janaki *et al.*, (2013) for oxyfluorfen. Then 20 $\mu$ l of each working standards were injected in to HPLC and the peak area was measured for linearity check study.

The analytical calibration graph equation obtained by plotting peak areas in y axis versus concentrations pendimethalin in x axis was  $Y= 17819x+3992$  with the range of 0.01 to 1.0  $\mu$ g g<sup>-1</sup>. It showed excellent linearity with the correlation coefficient of 0.997 (Fig. 2). Limit of detection (LOD) and Limit of Quantification (LOQ) of pendimethalin were found to be 0.01and 0.05  $\mu$ g mL<sup>-1</sup>.

Validity of the method was tested by conducting the recovery experiments through spiking of control soil sample (Janaki *et al* 2018) with four known concentration (0.05, 0.1, 0.5 and 1.0  $\mu$ g g<sup>-1</sup>) of pendimethalin in three replicates. Then pendimethalin residues were extracted from the spiked sample as described above.

The concentration of pendimethalin residues was determined by comparing the standard calibration curve and peak area of the sample. The blank soil was used to establish the limit of quantification. A quantification limit of 0.01 ppm was used for calculation.

### Statistical analysis

Linearization of degradation curves was performed by transformation of residue concentration (R) and time (t) using general regression equation  $y = a+bx$ . Based on the decline curves, the intercept (a), slope (b) and half-life (T<sub>1/2</sub>) were arrived using first order reaction kinetics function.

### Results and Discussion

#### Recoveries and detection limit

To make sure the consistency of the results, method validation is a essential requirement. Linearity precision, accuracy, coefficient determination, LOD and (LOQ) are the factors of method validation.

The recovery experiment showed that the mean recovery of pendimethalin at different levels of fortification in soil was 82.32% (Table 1). It indicates the described extraction and cleanup method was found to be satisfactory, since the analytical recovery of the pendimethalin the soil was more than 80 percent.

#### Degradation of pendimethalin in soil

Pendimethalin residue was analysed from soil samples collected from the pot grown with greengram as test crop at periodic intervals and found that the initial deposition on zero day after herbicide application (2hr) ranged from to 0.535 - 0.793 mg kg<sup>-1</sup> during 2018-19 (Table 2) and from 0.582 to 0.741 mg kg<sup>-1</sup> during 2019-20 (Table 3) across different organic sources consisting of microbes application to enhance the degradation of pendimethalin from the soil.

Further it was found that the dissipation was faster under FYM, AM and biochar applied treatments and the slowest degradation was

noticed in control during both the years. The lowest half life of 8.57 days (2018-19) and 8.32 days (2019-20) was observed in farm yard manure applied treatments. Previous studies have shown that atrazine biodegradation was significantly affected by application of different organic manures (Kadian *et al.*, 2008).

Similarly, Rathod *et al.*, (2010) reported that the half-life values in absence of FYM were higher for dinitroaniline herbicides used and dissipation rate of dinitroanilines increased progressively with time.

The degradation equation, correlation coefficient and half lives are given in Table 4 & 5. Irrespective of mitigation measures followed, the pendimethalin persisted up to 45 DAS and after 60 DAHA it became Below Detectable Level (BDL) during both the years of experimentation.

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**Table.1** Recovery (%) of pendimethalin from soil

Sample	Amount fortified ( $\mu\text{g g}^{-1}$ )	Amount recovered ( $\mu\text{g g}^{-1}$ )	Recovery (%)	SD	Average recovery (%)
Soil	0.05	0.039	77.89	$\pm 0.0035$	82.32
	0.1	0.080	79.52	$\pm 0.0106$	
	0.5	0.413	82.63	$\pm 0.0433$	
	1.0	0.892	89.24	$\pm 0.0516$	

**Table.2** Persistence of pendimethalin in soil as influenced by the organic sources and microbial application (2018-19)

Treatments		Pendimethalin residues ( $\text{mg kg}^{-1}$ ) in soil							
		0 day	1 day	3 day	5day	10 day	15 day	30 day	45 day
T <sub>1</sub>	FYM @ 10 t ha <sup>-1</sup>	0.535	0.442	0.364	0.268	0.136	0.080	0.033	0.013
T <sub>2</sub>	Vermicompost @ 5 t ha <sup>-1</sup>	0.543	0.381	0.311	0.222	0.123	0.076	0.042	0.012
T <sub>3</sub>	Biochar @ 5 t ha <sup>-1</sup>	0.633	0.498	0.398	0.226	0.196	0.097	0.037	0.023
T <sub>4</sub>	Phosphobacteria @ 10 kg ha <sup>-1</sup>	0.620	0.459	0.355	0.272	0.142	0.082	0.046	0.028
T <sub>5</sub>	Trichoderma @ 10 kg ha <sup>-1</sup>	0.574	0.492	0.405	0.329	0.211	0.108	0.062	0.035
T <sub>6</sub>	AM @ 10 kg ha <sup>-1</sup>	0.558	0.453	0.329	0.215	0.124	0.093	0.046	0.021
T <sub>7</sub>	Pseudomonas @ 10 kg ha <sup>-1</sup>	0.606	0.541	0.395	0.292	0.165	0.086	0.054	0.024
T <sub>8</sub>	Urea @ 100 kg ha <sup>-1</sup>	0.744	0.616	0.445	0.296	0.172	0.103	0.057	0.034
T <sub>9</sub>	Crop residue (maize straw) incorporation @ 5 t ha <sup>-1</sup>	0.588	0.389	0.348	0.294	0.196	0.115	0.069	0.035
T <sub>10</sub>	Control (no manure/bioagents)	0.793	0.622	0.479	0.346	0.229	0.118	0.064	0.039

**Table.3** Persistence of pendimethalin in soil as influenced by the organic sources and microbial application (2019-20)

Treatments		Pendimethalin residues (mg/kg) in soil							
		0 day	1 day	3 day	5day	10 day	15 day	30 day	45 day
T <sub>1</sub>	FYM @ 10 t ha <sup>-1</sup>	0.582	0.482	0.329	0.226	0.137	0.083	0.037	0.012
T <sub>2</sub>	Vermicompost @ 5 t ha <sup>-1</sup>	0.725	0.598	0.397	0.294	0.165	0.086	0.053	0.018
T <sub>3</sub>	Biochar @ 5 t ha <sup>-1</sup>	0.612	0.453	0.329	0.216	0.127	0.093	0.046	0.019
T <sub>4</sub>	Phosphobacteria @ 10 kg ha <sup>-1</sup>	0.689	0.456	0.329	0.215	0.128	0.093	0.046	0.021
T <sub>5</sub>	Trichoderma @ 10 kg ha <sup>-1</sup>	0.652	0.498	0.355	0.272	0.142	0.082	0.046	0.028
T <sub>6</sub>	AM @ 10 kg ha <sup>-1</sup>	0.741	0.621	0.478	0.347	0.201	0.118	0.064	0.030
T <sub>7</sub>	Pseudomonas @ 10 kg ha <sup>-1</sup>	0.682	0.471	0.329	0.226	0.137	0.083	0.047	0.025
T <sub>8</sub>	Urea @ 100 kg ha <sup>-1</sup>	0.599	0.387	0.348	0.292	0.188	0.115	0.069	0.038
T <sub>9</sub>	Crop residue (maize straw) incorporation @ 5 t ha <sup>-1</sup>	0.637	0.492	0.409	0.328	0.212	0.109	0.062	0.032
T <sub>10</sub>	Control (no manure/bioagents)	0.708	0.613	0.446	0.301	0.173	0.104	0.068	0.041

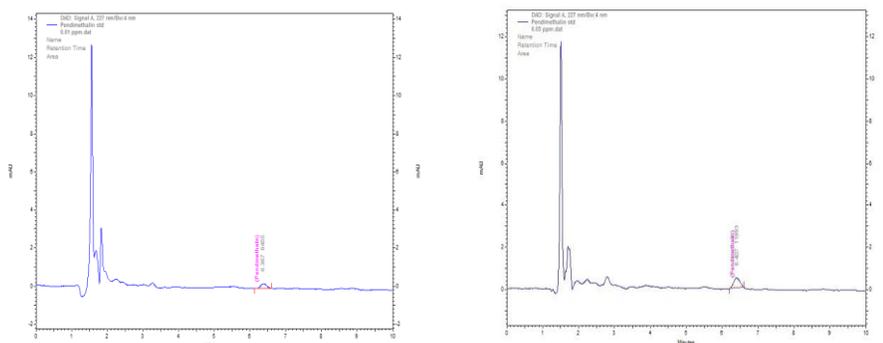
**Table.4** Pendimethalin dissipation equation, correlation coefficient and half lives in soil (2018-19)

Treatments		Regression equation	R <sup>2</sup>	Half life (days)
T <sub>1</sub>	FYM @ 10 t ha <sup>-1</sup>	y = -0.0809x + 0.920	0.970	8.57
T <sub>2</sub>	Vermicompost @ 5 t ha <sup>-1</sup>	y = -0.0782x + 0.997	0.958	8.86
T <sub>3</sub>	Biochar @ 5 t ha <sup>-1</sup>	y = -0.0747x + 0.824	0.931	9.28
T <sub>4</sub>	Phosphobacteria @ 10 kg ha <sup>-1</sup>	y = -0.0669x + 0.925	0.903	10.36
T <sub>5</sub>	Trichoderma @ 10 kg ha <sup>-1</sup>	y = -0.062x + 0.800	0.940	11.18
T <sub>6</sub>	AM @ 10 kg ha <sup>-1</sup>	y = -0.0691x + 0.986	0.931	10.03
T <sub>7</sub>	Pseudomonas @ 10 kg ha <sup>-1</sup>	y = -0.0704x + 0.812	0.930	9.85
T <sub>8</sub>	Urea @ 100 kg ha <sup>-1</sup>	y = -0.0418x + 0.735	0.886	10.66
T <sub>9</sub>	Maize straw incorporation @ 5 t ha <sup>-1</sup>	y = -0.0475x + 0.886	0.956	11.46
T <sub>10</sub>	Control (no manure/bioagents)	y = -0.0657x + 0.618	0.920	10.55

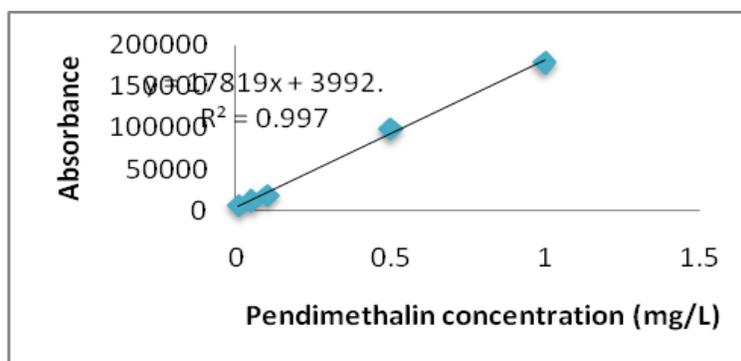
**Table.5** Pendimethalin dissipation equation, correlation coefficient and half lives in soil (2019-20)

Treatments		Regression equation	R <sup>2</sup>	Half life (days)
T <sub>1</sub>	FYM @ 10 t ha <sup>-1</sup>	y = -0.0833x + 0.853	0.955	8.32
T <sub>2</sub>	Vermicompost @ 5 t ha <sup>-1</sup>	y = -0.0741x + 0.739	0.924	9.35
T <sub>3</sub>	Biochar @ 5 t ha <sup>-1</sup>	y = -0.0608x + 0.962	0.927	9.90
T <sub>4</sub>	Phosphobacteria @ 10 kg ha <sup>-1</sup>	y = -0.0696x + 0.969	0.931	9.96
T <sub>5</sub>	Trichoderma @ 10 kg ha <sup>-1</sup>	y = -0.0678x + 0.897	0.897	10.22
T <sub>6</sub>	AM @ 10 kg ha <sup>-1</sup>	y = -0.0649x + 0.654	0.971	10.68
T <sub>7</sub>	Pseudomonas @ 10 kg ha <sup>-1</sup>	y = -0.0712x + 0.921	0.986	10.6
T <sub>8</sub>	Urea @ 100 kg ha <sup>-1</sup>	y = -0.0736x + 0.818	0.933	9.42
T <sub>9</sub>	maize straw Incorporation @ 5 t ha <sup>-1</sup>	y = -0.0648x + 0.763	0.946	10.7
T <sub>10</sub>	Control (no manure/bioagents)	y = -0.0618x + 0.751	0.882	11.22

**Fig.1** HPLC chromatograms of standard pendimethalin at 0.01 and 0.05  $\mu\text{g ml}^{-1}$



**Fig.2** Calibration curve of pendimethalin standard



Pendimethalin dissipated quickly in more neutral soil, but the addition of manure can only increase the dissipation rate in acidic soil, indicating that the amendment of manures exerted different effect in pendimethalin dissipation rates in different pH soils (Wen-Ching Chen *et al.*, 2018). On the other hand, the half-life values in absence of FYM were higher for all three dinitroaniline herbicides used (Rathod *et al.*, 2010). Similarly, pendimethalin herbicide applied in pigeonpea did not show any kind of phytotoxicity on the succeeding green gram even after 150 days after its application indicating that pendimethalin may not persist in the soil beyond 45 days after its application (Chavan *et al.*, 2018) (Table 5).

Based on the present results it was found that the FYM at 10 t ha<sup>-1</sup> or vermicompost at 5 t ha<sup>-1</sup> efficient in reducing the residual

concentration of pendimethalin in greengram grown soil. This could be due to the enhanced adsorption of the compounds by these sources. The application of FYM degraded the pendimethalin very fast due to the increased microbial population, which resulted the half life of 8.57 days (2018-19) and 8.32 days (2019-20). Many studies reported that the addition of organic amendments can also accelerate or enhance herbicide biodegradation by stimulating microbial growth in different soils (Herrero-Hernández *et al.*, 2015). Degradation of metribuzin in 24 different soils was investigated by Juhler *et al.*, (2008) and reported that in soils with higher organic matter content, the removal of herbicide was faster than soils with less organic matter content. Metribuzin residues in amended soils were degraded increasingly over time and the intensity of degradation depended on the

nature of organic amendment. Next to organics pendimethalin degradation was faster in biochar applied treatments. Sugarcane bagasse biochars mixed with clay and a silt loam soils and the pine wood biochar with a loamy sand soil had greater impact in reducing the half life period of pendimethalin (Paul *et al.*, 2015). However, NeetaSoni *et al.*, (2015) observed that biochar application reduced pendimethalin injury for all evaluated crop species and pendimethalin residues can be mitigated by the application of biochar at 0.50 kg/m<sup>2</sup> in soil. The slow degradation by microbes could be ascribed to the low quantity of application when compared to organics. Based on two years of pot culture experiment on mitigation of pendimethalin herbicide residues in sandy clay loam soils grown with green gram, it was observed that the farm yard manure at 10 t ha<sup>-1</sup> or vermicompost at 5 t ha<sup>-1</sup> is efficient in reducing the residual concentration of pendimethalin in green gram grown soil.

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