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Urease and dehydrogenase enzyme activity influenced by Diuron

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Abstract

Temporal dynamics of soil dehydrogenase and urease were studied in field experiments was conducted at Professor Jayashankar Telangana State Agricultural University, Rajendranagar during *kharif* 2017 where diuron was tested for weed control in red and black soils. The soil enzyme activity was higher in black soil over the red soil but however similar results of the chemicals was observed in both soils. Soil dehydrogenase activity were not affected by weed control treatment. However reduced soil urease was observed in diuron 1.0 kg ha⁻¹ fb pyrithiobac sodium + quizalofop p ethyl in both soils.

Keywords: diuron, polymulch, weed control, urease, phosphatase, dehydrogenase

Introduction

Application of herbicides in agriculture is an efficient and economic practice to control weeds. The usage of the herbicides can influence the activity of soil enzymes. Soil enzyme activity indicates the activity of all microorganisms and number of fundamental soil properties such as fertility and structure. The transformation of nutrients, turnover and mineralization of organic substances and their cycling all are dependent upon these enzymes. As dehydrogenase belong to oxydoreductases and catalyse the oxidation of organic compounds, urease belongs to amidohydrolases which catalyses the hydrolysis of urea to carbon dioxide and ammonia. Thus these soil enzymes are important in agriculture. Excessive mineral fertilization and modern cultivation practices are adding to the deterioration of soil fertility status. Diuron has a prolonged soil residual life (80-230 days) making it more suitable for cotton crop due its slow initial growth. However, in the research experiment conducted in PJTSAU, it was observed that diuron 80% WP applied at 1.0 kg ha⁻¹ caused significant reduction in plant stand in black soils which shows the need for the re-evaluation of herbicide dose in black and red soils also. Assessment of the enzymes present in soils offers potential as an integrative index of the soil's biological status. Thus environmental and soil concern have prompted the agricultural research to look for improved management strategies.

Materials and Methods

A field experiment was conducted during *kharif*, 2017 at College farm, College of Agriculture, Professor Jayashankar Telangana State Agricultural University, Rajendranagar, Hyderabad, Telangana State. The farm is geographically located at an altitude of 542.3 m above mean sea level at $17^{\circ}19^{\circ}$ N latitude and $78^{\circ}23^{\circ}$ E longitude in the Southern Telangana agro-climatic zone of Telangana. According to Troll's climatic classification, it falls under semi-arid tropics (SAT). The experiment was conducted in red and black soil and laid out in a randomised block design with three replications. The treatments included three doses of Diuron (0.5, 0.75 and 1.0 kg ha⁻¹), pendimethalin 38.7% CS at 677 g ha⁻¹ as PE followed by sequential application of pyrithiobac sodium 10% EC 62.5 g ha⁻¹ + quizalofop p ethyl 5% EC 50 g ha⁻¹, intercropping of cotton with green manure crop (Sunhemp), mechanical weeding thrice at 20, 40 and 60 DAS (weed free), polymulch and unweeded control.

Mallika *Bt* was sown with a seed rate of 2.5 kg ha⁻¹. One-two seeds per hill were sown at a spacing of 75cm X 75 cm to facilitate the use of power weeder in both directions in case of mechanical weeding. Pre-emergence herbicides were sprayed on the third day after sowing. Diuron 80% W.P. at 0.5 kg ha⁻¹, diuron 80% W.P. at 0.75 kg ha⁻¹, diuron 80% WP 1.0 kg ha⁻¹, pendimethalin 38.7% CS 677 g ha⁻¹ were sprayed on the third day,

pyrithiobac sodium 10% EC 62.5 g ha⁻¹+ quizalofop p ethyl 5% EC 50 g ha⁻¹ were sprayed at 2-3 leaf stage of the weeds. In the intercropping treatment the intercrop sunhemp was sown along with cotton. Polymulch was spread 8 DAS after emergence of the seedling. Mechanical weeding at 20, 40, 60 DAS was done with power weeder and an unweeded check was maintained. The activity of urease and dehydrogenase was evaluated on the day of spray, 3rd day of spray, 7th day of spray, 15th day of spray of the pre emergence and postemergence herbicide spray, at flowering and harvest. The soil was collected from top 5cm at each time.

Urease activity

Urease activity in soil was assayed by quantifying the ratio of release of NH_{4^+} from the hydrolysis of urea (Tabatabai and Bremner, 1972) ^[7]. 5g of soil was taken in a 50 ml volumetric flask, after adding 0.2 ml of toluene and 9 ml THAM buffer, the flask was swirled for a few seconds to mix the contents and 1ml of 0.2M urea solution was added and swirled again for a few seconds.

Then the flask was stoppered and placed in an incubator at 37° C. After 2 hours, the stopper was removed, and approximately 35 ml of KCl-Ag₂SO₄ solution was added, flask was swirled for a few seconds and allowed the flask to stand until the contents have cooled to room temperature (about 5 min). The contents were made to 50 ml by addition of KCl-Ag₂SO₄ solution, the flask was stoppered and inverted several times to mix the contents. NH⁺₄- N was determined in the resulting soil suspension, by pipetting out 20 ml aliquot of the suspension distilling with 0.2 g of MgO for 4 min.

Controls were performed by following the procedure described for assay of urease activity, but for the addition of 1ml of 0.2M urea solution after the addition of KCl-Ag₂SO₄ solution.

Dehydrogenase activity

Dehydrogenase enzyme is a respiratory chain enzyme that plays an important role in the energy production of organisms. This enzyme works as an essential component for enzyme system. DHA could be due to the activity going on in the rhizospheric region during the cropping system such as release of root exudates, mineralization, decomposition etc.

One gram of soil sample was taken in 50ml glass tube to which 50mg of CaCO₃ was added followed by 2.5ml of distilled water and 1ml of 3% TTC and swirled for few minutes and incubated at 37^{0} C for 24 hours. The red precipitate of the TPF was dissolved in 10ml of methanol and the contents were shaken for 30 minutes and then the contents were filtered into 25ml volumetric flask and the volume was made up to 25ml with methanol. Intensity of red colour was measured with spectrophotometer at 485 nm.

Results and Discussion

Urease Activity (µg NH4-N kg⁻¹ soil day⁻¹)

The activity of urease (expressed in μ g NH₄-N kg⁻¹ soil day⁻¹) was significantly influenced by the herbicidal treatments. The activity of the enzyme was increasing till flowering and later there was reduction in the activity was observed due to the decreasing root mass at harvest. The highest amount of the enzyme activity was at peak during the flowering stage which can be attributed to the highest rhizospherical area and there by increased root exudates. But there is spurt observed in the activity of urease enzyme after seventh day after preemergence as basal dose applied at 10 DAS. There was a significant difference in the urease activity among the

treatments upto third day after PoE application in red and black soils.

Red soil

On the day of PE spray (22.6 - 24.5) and third of PE spray (36.2 - 38.5), all the non-herbicidal treatments were on par and recorded significantly highest urease activity. The highest dose of diuron at 1.0 kg ha⁻¹ fb pyrithiobac sodium + quizalofop p ethyl PoE recorded significantly lowest urease activity at on the day of PE spray (7.0), 3rd day of PE spray (14.0), 7th day of PE spray (21.0), 15th day of PE spray (38.3), on the day of PoE spray (42.7) and 7th day of PoE spray (52.2). The dosage of diuron at 0.75 kg ha⁻¹ fb pyrithiobac sodium + quizalofop p ethyl PoE recorded significantly lower activity compared lower dose of diuron and pendimethalin but comparatively higher activity than the highest dosage at on the day of PE spray (12.8) and 3rd day of PE spray (22.2) and later was on par with other chemical treatments. The lower dosage of diuron at 0.5 fb pyrithiobac sodium + quizalofop p ethyl PoE and Pendimethalin treatments also registered significantly lower activity of urease on the day of PE spray and 3rd day of PE spray when compared to the non-herbicidal treatments. There was no significant difference among the treatments at on the 7th day of PoE spray, 15th day of PoE spray, at flowering and at harvest.

Black soil

Similar to that of the red soil the highest amount of urease activity on the day of PE spray (6.00 - 27.17) and 3rd day of PE spray (33.83 - 34.50) was recorded with non-herbicidal treatments. In black soils the significantly lowest amount of activity was observed in case of diuron at 1.0 kg ha⁻¹ and at 0.75 kg ha⁻¹ fb pyrithiobac sodium + quizalofop p ethyl PoE on the day of PE spray (11.3 and 14.00 respectively) and 3rd day of PE spray (19.83 and 21.83 respectively) while diuron at 0.5 kg ha⁻¹ and pendimethalin fb pyrithiobac sodium + quizalofop p ethyl PoE were on par with each other and significantly lower than non-chemical treatments but significantly higher activity compared to higher doses of diuron. From the 7th day of PE spray to the 3rd of PoE spray significantly lowest activity of the urease was registered with diuron at 1.0 kg ha⁻¹ and at 0.75 kg ha⁻¹ fb pyrithiobac sodium + quizalofop p ethyl PoE and other treatments were comparable with eachother. From the 7th day of PoE spray till the harvest there was no significant difference observed among the treatments.

Dehydrogenase activity (µg TPF kg⁻¹ soil day⁻¹)

There was no significant difference among the treatments observed in both red and black soils.

Based on the perusal of the literature varying results were observed which were stimulation, inhibition or no effect. Similar results have been reported by Balasubramanyan *et al.* (1970) ^[3] for simazine, Baruah and Mishra (1986) ^[4] for butachlor, Arya and Ameena (2016) ^[1] for bensulfuron ethyl and pyrazosulfuron ethyl, Baboo *et al.* (2013) ^[2], Kumari *et al.* (2018) ^[5] for various herbicides and Lal *et al.* (2017) ^[6] for imazethpyr for substituted urea herbicides. They were of the opinion that the soil enzymes are protected against inhibition activity decreases. The perusal of the data indicates that the activity of soil urease was supressed with the application herbicide and the suppression increased with the increase in dosage of the herbicide while there was no effect on the soil dehydrogenase activity. With the increase in the

time the activity was regained which may be due to the different reasons i.e., the herbicide effect on microbial population may get stabilized after some time and the herbicides themselves are adsorbed irreversibly on soil colloids with increase in time resulting in decreased inhibition. The partial degradation of the herbicide with time in soil may also be another factor for decrease in inhibition. The recovery from inhibition may also be due to enzyme secreted by plant themselves. The detracting effect of herbicides towards all microbes and enzyme activities decreased with time and this may also be due to microbial population and enzyme activities after initial inhibition due to microbial adaptation to these chemicals or due to their degradation.

Conclusion

From the study the application of the herbicide diuron led to the inhibition of soil urease activity and no effect was shown on the dehydrogenase activity. While the urease enzyme activity was regained with increase in duration of exposure.

		Preeme	ergence			Poste				
Treatments	on the day of spray	on the 3 rd day	on the 7 th day	on the 15 th day	on the day of spray	on the 3 rd day	on the 7 th day	on the 15 th day	Floweri ng	Harve st
Diuron 80% WP 0.5 kg ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	17.5	29.0	36.2	54.5	57.6	63.2	75.33	83.4	106.6	66.2
Diuron 80% WP 0.75 kg ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	12.8	22.2	35.0	42.2	52.5	58.7	74.50	83.6	104.6	64.1
Diuron 80% WP 1.0 kg ha ⁻¹ fb pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	7.0	14.0	21.0	38.3	42.7	52.2	71.50	81.2	103.0	60.7
Pendimethalin 38.7% CS at 677 g ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	16.3	30.3	36.2	52.7	58.6	62.7	75.33	83.6	105.1	61.6
Cotton + sunhemp	24.2	38.3	40.8	57.0	62.3	66.3	78.67	86.2	109.7	68.6
Mechanical weeding at 20, 40, 60 DAS	23.3	38.5	40.8	57.2	61.2	66.3	77.17	85.0	111.2	61.0
Control (unweeded)	24.5	37.5	38.5	57.0	62.7	66.7	77.33	87.3	114.2	70.2
Polymulch of 0.25 mm thickness	22.6	36.2	39.7	56.2	60.0	65.5	76.70	86.1	115.8	69.3
SE(m)±	1.19	1.63	1.78	1.62	2.21	1.88	1.36	5.28	9.00	3.33
C.D. (p=0.05)	3.64	4.99	5.44	4.95	6.76	5.75	NS	NS	NS	NS

Table 1: Soil urease activity (µg NH4-N kg⁻¹ soil day⁻¹) as influenced by weed control options in red soil

Table 2: Soil urease activity (µg NH4-N kg⁻¹ soil day⁻¹) as influenced by weed control options in black soil

	Preemergence					Postem				
Treatments	on the day of spray	on the 3 rd day	on the 7 th day	on the 15 th day	on the day of spray	on the 3 rd day	on the 7 th day	on the 15 th day	Flowering	Harvest
Diuron 80% WP 0.5 kg ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	21.00	27.67	36.17	50.67	72.50	79.2	83.7	90.6	96.6	57.7
Diuron 80% WP 0.75 kg ha ⁻¹ fb pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	14.00	21.83	32.83	48.52	69.00	75.3	82.0	90.3	95.7	56.1
Diuron 80% WP 1.0 kg ha ⁻¹ fb pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	11.83	19.83	22.67	43.33	56.00	63.5	80.5	89.4	92.9	54.7
Pendimethalin 38.7% CS at 677 g ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	21.67	28.50	35.83	50.50	71.33	78.7	86.7	91.0	100.1	58.0
Cotton + sunhemp	27.17	34.33	42.70	51.32	77.67	83.8	85.8	92.3	103.6	59.4
Mechanical weeding at 20, 40, 60 DAS	26.47	34.50	40.50	51.83	77.00	82.3	86.0	91.6	102.5	58.2
Control (unweeded)	26.00	33.83	40.00	52.07	75.87	81.3	84.7	92.0	101.9	56.9
Polymulch of 0.25 mm thickness	26.67	34.67	42.83	51.83	77.33	82.3	85.0	91.0	101.6	57.6
SE(m)±	0.73	1.41	1.10	1.01	1.85	2.51	2.90	2.46	5.89	4.2
C.D. (p=0.05)	2.22	4.31	3.37	3.09	5.68	7.69	NS	NS	NS	NS

Table 3: Soil dehydrogenase activity (µg TPF kg⁻¹ soil day⁻¹) as influenced by weed control options in red soil

		Preem	ergence			Postem				
Treatments	on the day of spray	on the 3 rd day	on the 7 th day	on the 15 th day	on the day of spray	on the 3 rd day	on the 7 th day	on the 15 th day	Flowering	Harvest
Diuron 80% WP 0.5 kg ha ⁻¹ fb pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	3.30	4.24	5.12	5.29	5.45	5.55	5.68	5.75	7.78	4.83
Diuron 80% WP 0.75 kg ha ⁻¹ fb pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	3.22	4.33	5.02	5.20	5.32	5.45	5.62	5.83	7.71	4.84
Diuron 80% WP 1.0 kg ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	3.12	4.44	4.58	5.02	5.34	5.48	5.64	5.82	7.71	4.82
Pendimethalin 38.7% CS at 677 g ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	3.26	4.84	5.07	5.09	5.38	5.52	5.68	5.89	7.79	4.91
Cotton + sunhemp	3.34	5.08	5.23	5.34	5.51	5.65	5.97	6.01	7.88	4.96
Mechanical weeding at 20, 40, 60 DAS	3.33	5.03	5.26	5.31	5.47	5.66	5.77	6.01	7.75	4.98
Control (unweeded)	3.30	5.04	5.17	5.27	5.45	5.59	5.75	5.93	7.70	4.86
Polymulch of 0.25 mm thickness	3.37	5.02	5.28	5.30	5.47	5.61	5.77	5.97	7.74	4.88

SE(m)±	0.11	0.24	0.26	0.08	0.11	0.18	0.20	0.15	0.13	0.05
C.D. (p=0.05)	NS									

Table 4: Soil dehydrogenase activity (µg TPF kg⁻¹ soil day⁻¹) as influenced by weed control options in black soil

		Preem	ergence			Postem				
Treatments	on the day of spray	on the 3 rd day	on the 7 th day	on the 15 th day	on the day of spray	on the 3 rd day	on the 7 th day	on the 15 th day	Flowering	Harvest
Diuron 80% WP 0.5 kg ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	3.39	4.97	5.15	5.32	5.44	5.59	5.77	5.81	7.77	4.82
Diuron 80% WP 0.75 kg ha ⁻¹ fb pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	3.28	4.88	4.91	5.08	5.20	5.68	5.69	5.93	7.81	4.85
Diuron 80% WP 1.0 kg ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	3.12	4.77	4.85	5.03	5.27	5.34	5.56	5.87	7.71	4.81
Pendimethalin 38.7% CS at 677 g ha ⁻¹ <i>fb</i> pyrithiobac sodium 10% EC 62.5 g ha ⁻¹ + quizalofop p ethyl 5% EC 50 g ha ⁻¹	3.44	4.88	4.97	5.15	5.31	5.51	5.66	5.84	7.74	4.82
Cotton + sunhemp	3.55	5.03	5.33	5.50	5.63	5.80	6.01	6.14	7.82	4.74
Mechanical weeding at 20, 40, 60 DAS	3.70	5.14	5.27	5.44	5.57	5.80	5.93	5.87	7.80	4.86
Control (unweeded)	3.47	5.03	5.23	5.41	5.52	5.75	5.91	6.06	7.74	4.80
Polymulch of 0.25 mm thickness	3.49	5.10	5.23	5.41	5.53	5.67	5.84	6.04	7.91	4.87
SE(m)±	0.18	0.14	0.27	0.28	0.28	0.21	0.28	0.22	0.12	0.08
C.D. (p=0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

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