

Original Research Article

Influence of new generation herbicide on microbial biomass dynamics in sandy loam soil

ABSTRACT

A laboratory experiment was conducted with two new generation herbicide pyrazosulfuron ethyl treatments, namely, recommended dose 15 g a.i. ha⁻¹ (RD) and 30 g a.i. h⁻¹ (2RD), carfentrazone ethyl 25 g a.i ha⁻¹(RD), 50 g a.i. ha⁻¹ (2RD) along with control applied to sandy loam soil to determine their effect on microbial biomass dynamics in soil. The application of pyrazosulfuron ethyl (T₁ and T₂) and carfentrazone ethyl (T₃ and T₄) did not show any significant effect at 0 day after application. While, at 7 days after application of herbicide declined soil microbial biomass status. Whereas, at 28 days after application significantly increase microbial MBC and MBN except for MBP under herbicidal treatments compared to untreated plot. Hence, pyrazosulfuron and carfentrazone ethyl may cause short term transitory change in microbial biomass dynamics in soil.

Keyword: Microbial biomass Carbon, Nitrogen, Phosphorus, Herbicide.

INTRODUCTION

Weed management is an integral part of crop production, and herbicides continue to be the most common weed management tool in most cropping systems. Because herbicides are usually applied when crops are absent or at early growth stages, most of the spray solution contacts soil. These chemicals may affect non-target soil organisms, including microorganisms. Herbicide-induced changes in abundance, diversity and activity of soil microbial communities may, in turn, influence microorganism-mediated processes that are important to sustainable agriculture, e.g., recycling of plant nutrients and maintenance of soil structure. Biological properties are critically important to the ecosystem functioning since they are involved in soil organic matter decomposition, nutrient cycling, and degradation of pesticides, such as herbicides. Therefore, studies assessing the effect of herbicides on soil biological properties are important for evaluating soil quality and health. In addition, soil biological properties are more effective as indicators of soil quality than physical and chemical properties as they often show a faster response to an environmental impact (Pertile *et al.*, 2020). Most studies on herbicide effects on soil microorganisms have focused on one or two herbicides at a time (Haney *et al.*, 2000). While results of these studies indicate that herbicides applied at recommended rates generally do not have significant effects on soil microorganisms, evaluating only a few herbicides at a time limits comparison amongst herbicides on their relative effects on soil microbial ecology. In addition, traditional methods of evaluating effects of herbicides on microorganisms have focused on microbial biomass (or parameters correlated with biomass) and functional parameters such as carbon and nitrogen mineralization (Johnsen *et al.*, 2001). However, the diversity or structure of

Comment [WU1]: Abstract is made up of these parts: introduction/problem statement, methodology, result/findings and conclusion/impact. Check you abstract these features are lacking. Your spacing should be single line.

Comment [WU2]: Herbicides: and

Comment [WU3]: You did not tell us what you used as your control and what is the safe dose of application

Comment [WU4]: MBC, MBN and MBP write in full

Comment [WU5]: Hence

Comment [WU6]: Did this justify your research?

Comment [WU7]: Rephrase

Comment [WU8]: No references

Comment [WU9]: Is this you aim?

Comment [WU10]: Recommended dose

the soil microbial community may be altered markedly even if total biomass or carbon and nitrogen metabolism appear unaffected by the herbicide. Soil microbial biomass, both the source and sink of available nutrients, plays an important role in nutrient transformations. The direct and indirect effects of toxic chemicals on soil biology include reduction in microbial population and reduced mineralization of organic compounds(). Dipika (2014) reported that the application of herbicides exerted adverse effect on soil microbial biomass carbon. The objective of this work was to screen up to 2 herbicides, representing several chemical families, modes of action and different soil residual properties, for their effects on soil microbial biomass carbon, nitrogen and phosphorus. It was hypothesized that these differences among herbicides would influence their impact on soil microbial activity.

MATERIALS AND METHODS

A bulk soil sample (soil depth, 0-15 cm) from the research farm of the BAU, Sabour was collected to conduct an incubation study of the herbicidal impacts on soil microbial biomass and enzyme activity. The university is located at 25°15'4" N and 78°2'45" E and 37.19 metres above the mean sea level on Indo-Gangetic plains. 200 gms of the air-dried, processed soils are taken in beakers (500 ml). The total number of beakers is 100 (treatments (5) x replications (4) x incubation periods (5)). The treatment details are No herbicide, Pyrazosulfuron ethyl @ 15 g and 30 g a.i. ha⁻¹, and Carfentrazone ethyl @ 25 g and 50 g a.i. ha⁻¹. The incubation periods of this study were 0, 7, 14, 21 and 28 days of application (DAA) at 28 °C. The experiment was arranged in a completely randomized design. Soils containing beakers of different treatments were collected at every incubation period and further examined for the dynamics of microbial biomass carbon such as MBC, MBN and MBP. Two set of fresh soil samples were taken in beakers, the first one was fumigated with chloroform for 24 hours, while the second one was kept unfumigated. Both the fumigated and unfumigated soil samples were extracted with using 0.5 M K₂SO₄. A blank was also run simultaneously. Then, 2 ml K₂Cr₂O₇, 10 ml H₂SO₄ and 5 ml H₃PO₄ were added to 10 ml of the extracts. They were kept at 160 °C for 30 minutes on hot plate, then take out conical flasks and add 250 ml of distilled water immediately after cool at room temperature, add 2-3 drops of ferroin indicator and titrate the contents against 0.2 N ferrous ammonium sulphate to get a brick-red end point (Vance *et al.*, 1987), MBN (Brookes *et al.*, 1985), and MBP (Brookes *et al.*, 1982). Data were analysed using ANOVA for factorial completely randomized design (CRD) as described by (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

Microbial Biomass Carbon (MBC)

The application of Pyrazosulfuron ethyl 10% WP and Carfentrazone ethyl 40% DF was found to have no significant effect on microbial biomass carbon under neither of days of application (Table-1). Then at 7 DAA, maximum MBC recorded (176.45 µg g⁻¹ soil) in the treatment T₃(control). While the application of Pyrazosulfuron ethyl 10% WP (T₁ and T₂) and Carfentrazone ethyl 40% DF (T₃ and T₄) in both doses (single and double recommended doses) significant by declined the microbial biomass carbon in 7 days after application. The percent

Comment [WU11]: No reference

Comment [WU12]: This study tries to evaluate the influence of ... and ... two new generation herbicides ...

Comment [WU13]: Sectionalize your methodology .it's too jam-packed

Comment [WU14]: What is your justification for collecting soils at this level

Comment [WU15]: Write in full

Comment [WU16]: No herbicide (control)

Comment [WU17]: Why did you use only two points, why not go below 15g like 5, 10g and above 30g... same applies to carfentrazone ethyl

Comment [WU18]: Write in full

Comment [WU19]: You have to be consistent with what you are using. Initially you used 15g a.i ha⁻¹ now you are using percentage. Please can u explain. Write WP and DF in full for the first time

decreases by the treatment were 17.04, 14.22, 14.21, and 12.16 as compared to control. (Yang and Zheng, 2006) found that decline microbial biomass carbon application of herbicide. It was also hypothesized that cell division of microbial population hampered upon the first stressful event and it takes time to recover its growth until the stressor was completely removed (Andersson 2016; Kumar *et al.*, 2019). Likewise, the present experiment was conducted in microcosm condition under dark incubation where degradation of Pyrazosulfuron was restricted from many factors such as photolysis, volatilization, leaching and run-off processes (Kanrar and Bhattacharyya, 2019). In our study at 28 days after application of herbicide significant increases in microbial biomass carbon in all herbicidal treatment over untreated plot due to enhanced activity of microorganisms due to application herbicide. The herbicide and days interaction were significant on microbial biomass carbon in the order T₁, T₃, T₂ and T₄ respectively, over control under 28 days after application of herbicide.

Table 1. Effect of herbicidal treatment on microbial biomass carbon under incubation laboratory experiment

Treatments	Microbial biomass carbon ($\mu\text{g g}^{-1}$ soil) (Average of four replications sat)					
	0 DAA	7 DAA	14 DAA	21 DAA	28 DAA	Mean
T ₁	176.43	154.98	150.39	156.42	185.56	164.76
T ₂	176.09	151.37	146.30	151.26	179.75	160.95
T ₃	176.37	151.13	148.22	154.87	181.49	162.41
T ₄	176.53	146.37	144.18	149.87	178.70	159.13
T ₅	176.61	176.45	176.62	176.36	175.53	176.31
Mean	176.40	156.06	153.14	157.76	180.21	-
Particulars				SEm \pm	CD at 5%	CD at 1%
Herbicide (H)				1.12	3.16	4.19
Days (D)				1.12	3.16	4.19
Interaction (HxD)				2.51	5.21	9.37

Treatment Details: T₁ - Pyrazosulfuron ethyl 10% WP 15 g *a.i.* ha⁻¹, T₂ - Pyrazosulfuron ethyl 10% WP 30 g *a.i.* ha⁻¹, T₃ - Carfentrazone ethyl 40% DF 25 g *a.i.* ha⁻¹, T₄ - Carfentrazone ethyl 40% DF 50 g *a.i.* ha⁻¹, T₅ - Control, DDA-Day after application.

Microbial Biomass Nitrogen (MBN)

The application of Pyrazosulfuron ethyl 10% WP and Carfentrazone ethyl 40% DF was found to have no significant effect on microbial biomass nitrogen under at zero days after application (Table-2). While, at 7 DAA significant reduction microbial biomass nitrogen in herbicidal treatment compared to control. Thereafter, at 28 DAA maximum (19.23 $\mu\text{g g}^{-1}$ soil) MBN recorded in T₁ (Pyrazosulfuron ethyl 10% WP 15 g *a.i.* ha⁻¹) followed by T₃ (Carfentrazone ethyl 40% DF 25 g *a.i.* ha⁻¹) as compared to control plot. Singh *et al.*, (2007) showed a strong negative correlation between microbial biomass and crop root biomass through the crop cycles. Similar temporal trend of microbial biomass was also reported by (Bhattacharyya *et al.*, 2005; Ghoshal and Singh 2010). However, contrasting trends showing increase in the size of the microbial

Comment [WU20]: You have to try better with your discussion. It looks non-scientific. Try and throw more light. Does your citations agree or disagree with your findings

biomass with the crop growth. Microbial biomass carbon was highly sensitive to herbicide application compared to dehydrogenase activity. Application of pendimethalin exerted adverse effect on microbial biomass carbon and dehydrogenase activity followed by bispyribacsodium, oxyfluorfen, and cyhalofop-butyl at all the intervals. The adverse effects of herbicides on biological activity were minimal in high organic matter soils (Amrita *et al.*, 2017). The herbicide and days interaction were significant on microbial biomass nitrogen in the order T₁, T₃, T₂ and T₄ respectively, over control under 28 days after application of herbicide.

Table 2. Effect of herbicidal treatment on microbial biomass nitrogen under incubation laboratory experiment

Treatments	Microbial biomass nitrogen ($\mu\text{g g}^{-1}$ soil) (Average of four replications sat)					
	0 DAA	7 DAA	14 DAA	21 DAA	28 DAA	Mean
T ₁	17.34	14.43	15.76	17.09	26.54	19.23
T ₂	17.65	12.56	14.76	16.60	23.65	17.84
T ₃	17.43	12.76	15.76	16.66	24.43	18.61
T ₄	17.59	11.34	14.89	16.59	25.89	17.26
T ₅	17.44	17.79	17.91	17.98	21.43	17.11
Mean	17.49	13.78	15.82	17.38	25.59	-
Particulars				SEm \pm	CD at 5%	CD at 1%
Herbicide (H)				0.09	0.26	0.26
Days (D)				0.09	0.26	0.26
Interaction (H×D)				0.21	0.56	0.72

Microbial Biomass Nitrogen (MBN)

It has been observed from the data presented in Table-2 that application of Pyrazosulfuron ethyl 10% WP and Carfentrazone ethyl 40% DF did not show any significant effect on microbial biomass phosphorus up to 28 days after application of herbicide in incubation period. Herbicide application at the recommended dose in this study did not result any significant change in soil MBC, MBN, and MBP compared to the control treatment, probably because the effects of herbicide application on microbial biomass is short-term and relatively insignificant compared with the seasonal variations (Subhani *et al.*, 2000). Hart and Brookes (1996) reported no effect on microbial biomass after 19 years of annual field applications of pesticides (glyphosate, benomyl, chlorfenvinphos, and triadimefon) applied at the recommended rates. Singh and Ghoshal (2010) reported reductions in the microbial biomass due to the application of herbicide, whereas (Moreno *et al.*, 2007) reported higher levels of microbial biomass. The herbicide and days interaction were non significant on microbial biomass phosphorus in the order T₁, T₃, T₂ and T₄ respectively, over control under 28 days after application of herbicide.

Comment [WU21]: Check

Table 3. Effect of herbicidal treatment on microbial biomass phosphorus under incubation laboratory experiment

Treatments	Microbial biomass phosphorus ($\mu\text{g g}^{-1}$ soil) (Average of four replications sat)					
	0 DAA	7 DAA	14 DAA	21 DAA	28 DAA	Mean
T ₁	8.45	8.68	14.65	13.76	14.94	12.10
T ₂	8.43	7.93	14.89	12.54	13.97	11.55
T ₃	8.64	8.94	14.02	13.56	15.43	12.12
T ₄	8.56	8.03	13.43	13.98	14.84	11.77
T ₅	8.38	8.56	13.98	14.87	14.56	12.07
Mean	8.49	8.43	14.19	13.74	14.75	-
Particulars				SEm \pm	CD at 5%	CD at 1%
Herbicide (H)				0.14	0.37	0.67
Days (D)				0.14	0.37	0.67
Interaction (HxD)				0.29	0.73	1.36

Conclusion

The application of Pyrazosulfuron ethyl 10% WP and Carfentrazone ethyl 40% DF single and double doses of herbicide did not show any significant effect on microbial biomass dynamics at 0 DAA. While, at 7 DAA of herbicide significant decline MBC and MBN except MBP. Thereafter, at 28 DAA significant increases microbial biomass carbon and microbial biomass nitrogen as compared to control. Pyrazosulfuron ethyl and Carfentrazone ethyl have a short-term transitory change in the microbial biomass dynamics of the soil.

Reference

- Amrita, K. and Devi, K.M. (2017) Effect of pre and post emergence herbicides on microbial biomass carbon and dehydrogenase activity. *Journal of Tropical Agriculture*. 55 (1): 114-118.
- Amrita, K. and Devi, K.M. (2017) Effect of pre and post emergence herbicides on microbial biomass carbon and dehydrogenase activity. *Journal of Tropical Agriculture*. 55 (1): 114-118.
- Andersson, S.G. (2016) Stress management strategies in single bacterial cell. *Proc.Natl.Acad.Sci.* 113(15), 3921-3923.
- Bhattacharayya, P., Mukherjee, A., Datta, P., Karmakar, P.R. and Kole, R.K. (2005) Dissipation pattern of the herbicide Pyrazosulfuron- ethyl in Alluvial and red lateritic soils of West Bengal. *Journal of crop and weed*. 2(2): 65-69.
- Brookes, P.C., Landman, A., Pruden, G. and Jenkinson, D.S. (1985) Chloroform fumigation and the release of soil nitrogen: A rapid direct extraction method to measure microbial biomass nitrogen in soil. *Soil BiolBiochem*. 17: 837-842.

Comment [WU22]: When we talk microbial biomass I think you should be talking the organisms? I understanding measuring carbon, nitrogen and phosphorus but organisms should also be a measure

- Brookes, P.C., Powlson, D.S. and Jenkinson, D.S. (1982) Measurement of microbial biomass phosphorus in soil. *Soil BiolBiochem.* 14: 319–329.
- Dipika (2014) Effect of different post emergence herbicides on microbiological and biochemical properties of rice soil. M. Sc. (Ag.) thesis, Indira Gandhi Krishi Vishwavidyalaya, Raipur, 99p.
- Ghoshal, N., Singh, K. P. (2010) Impact of addition of various resource quality inputs on soil CO₂ flux and C balance in a tropical dryland agroecosystem. In 19th World Congress of Soil Science on Soil Solutions for a Changing World. Brisbane. pp. 113–116.
- Gomez, K.A. and Gomez, A.A. (1984) Statistical procedures for agricultural research (2 ed.). John Wiley and sons, Newyork, 680p.
- Haney, R. L., Senseman, S. A., Hons, F. M. and Zuberer, D. A. (2000) Effect of glyphosate on soil microbial activity and biomass. *Weed Sci.* 48: 89–93.
- Hart, M.R. and Brookes, P.C. (1996) Soil microbial biomass and mineralization of soil organic matter after 19 years of cumulative field applications of pesticides. *Soil BiolBiochem.* 28: 1641–1649.
- Johnsen, K., Jacobsen, C.S. and Torsvik, V. (2001) Pesticide effects on bacterial diversity in agricultural soils—a review. *Biol. Fertil. Soils.* 33: 443–453.
- Kanrar, S. and Bhattacharyya, S. (2019) Soil microbial population as affected by herbicides. *Madras Agric. J.* 80, 397-99.
- Kumar, U., Panneerselvam, M., Priya, P., Chakraborty, K., Swain, P., Chatterjee, S.N., Sharma, S.G. and Nayak, A.K. (2019) Ascorbic acid for mulation for survivability and diazotrophic efficacy of *Azotobacterchroococcum* Avi2 (MCC 3432) under hydrogen peroxide stress and its role in plant growth promotion in rice (*Oryza sativa* L). *Plant Physiol.Biochem.* 139, 419-427.
- Moreno, J.L., Aliaga, A., Navarro, S., Hern, T., Garcia, C. (2007) Effects of atrazine on microbial activity in semiarid soil. *Appl Soil Ecol.* 35: 120–127.
- Pertile, M., Antunes, J.E., Araujo, F.F., Mendes, L., Brink, J.V., and Araujo, A.S. (2020) Responses of soil microbial biomass and enzyme activity to herbicides imazethapyr and fumioxazin. *Scientific report.* 10:7694 | <https://doi.org/10.1038/s41598-020-64648-3>.
- Singh, K. and Tripathi, H.P. (2007) Effect of nitrogen and weed-control practices on performance of irrigated direct seeded rice (*Oryza sativa*). *Indian Journal of Agronomy.* 52(3): 231-234.
- Subhani, A., Elghamry, A.M., Huang, C.Y., Xu, J.M. (2000) Effects of pesticides (herbicides) on soil microbial biomass, A review. *Pakistan J Biol Sci.* 3: 705-709.

Comment [WU23]: Use recent references at least five years upwards

Vance, E.D. and Brookes, P.C. and Jenkinson, D.S. (1987) An extraction method for measuring soil microbial biomass carbon. *Soil BiolBiochem.* 19: 703–707.

Yang, J.C., Zheng, Mt. (2006) Functional diversity of microbial communities: a quantitative approach. *Soil Biol. Biochem.* 26: 1101–1108.

UNDER PEER REVIEW

UNDER PEER REVIEW

