



Effect of Crop Establishment Methods on Rhizospheric Soil Microbiota in Mustard Due to Different Land-Use Systems

Komal Bhatt ^{a*}, S. B. Agrawal ^a, R. Bajpai ^a, B. S. Dwivedi ^b,
Poornima Malviya ^c, Deeksha Gupta ^a and Anjali Anand ^a

^a Department of Forestry, College of Agriculture, JNKVV, Jabalpur-482004, (Madhya Pradesh.), India.

^b Department of Soil Science, College of Agriculture, JNKVV, Jabalpur-482004, (Madhya Pradesh), India.

^c Department of Agronomy, College of Agriculture, JNKVV, Jabalpur-482004, (Madhya Pradesh), India.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: <https://doi.org/10.9734/ijecc/2024/v14i94417>

Open Peer Review History:

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: <https://www.sdiarticle5.com/review-history/122881>

Original Research Article

Received: 28/06/2024

Accepted: 31/08/2024

Published: 02/09/2024

ABSTRACT

A field study was conducted to assess the effect of crop establishment methods on rhizospheric soil microbiota in mustard due to different land-use system at the Research Farm, Department of Forestry, College of Agriculture, JNKVV, Jabalpur (MP) for two consecutive years (2022-23 and 2023-24). The experiment was assessed in the double-split plot design with three replications. The

*Corresponding author: E-mail: komalbhatt2797@gmail.com;

experiment consisted of two land-use systems as main plot treatment (S₁: Agroforestry system and S₂: Open system), three crop establishment methods of mustard (M₁: Broadcasting, M₂: Line sowing, and M₃: Transplanting) as sub-plot treatments and four levels of boron (B₀: Control, B₁: 1 kg ha⁻¹ as basal B₂: 2 kg ha⁻¹ and B₃: ½ kg ha⁻¹ as basal + ½ kg ha⁻¹ as foliar spray) as sub-sub plot treatments. The soil samples were collected after harvesting of crop during both the years of study to analyse soil microbiota (total bacteria, azotobacter, rhizobium, total fungi and actinomycetes). The results revealed that, significantly higher counts of azotobacter (31.37 CFU x 10⁵), rhizobium (21.99 CFU x 10⁶), total bacteria (42.78 CFU x 10⁶), total fungi (8.30 CFU x 10⁵) and actinomycetes (12.98 CFU x 10⁵) were higher under agroforestry system than the open system. Meanwhile higher counts of azotobacter, rhizobium, total bacteria, total fungi and actinomycetes were found to be significantly superior in broadcasting (29.59 CFU x 10⁵, 20.28 CFU x 10⁶, 37.52 CFU x 10⁶, 7.97 CFU x 10⁵, 12.68 CFU x 10⁵, respectively) over both line sowing and transplanting crop establishment methods. In case of boron levels, no significant changes occurred in the soil biology under both agroforestry and open systems. Hence, it could be concluded that vegetative cover over soil helps to proliferation of population of soil microbiota in the rhizospheric zone of crop.

Keywords: *Agroforestry systems; crop establishment methods; boron levels; mustard; open system; soil biology and shisham.*

1. INTRODUCTION

Soil serves as the foundation for agriculture, and the presence of microorganisms is crucial for enhancing soil health and promoting robust crop growth. The microbial biosphere is the most extensive reservoir of biodiversity on the planet [1]. Microorganisms play a vital role in establishing an intricate interrelationship between plants and the soil. Soil microorganisms are an active and essential part of the soil, carrying out numerous functions within the soil system.

Microorganisms have the ability to recycle the nutrients present in the soil [2]. Soil microorganisms play a crucial role in enhancing the quality of soil and its maintenance within the soil system. Soil microorganisms have a crucial role in the decomposition of organic matter, such as animal and plant remains. They also contribute to the formation of soil structure and govern the rate of biogeochemical cycling in the soil [3]. The soil harbours a multitude of microorganisms that play a crucial role in augmenting soil fertility and stimulating plant growth [4]. Moreover, it has been shown that numerous bacterial species have been employed for converting organic contaminants into minerals in soil, which is commonly known as bioremediation of soil pollutants [5].

Agroforestry is a holistic approach that provides ecological and environmental benefits. Nutrient cycling in agroforestry leads to an increase in soil productivity through a variety of microbiological and biological processes [6]. The internal recycling of nutrients, litter fall, root rot, and other

processes can all result in nutrient transfers. By increasing the amount of nutrients available to crops, lowering the amount of nutrients lost from the system through leaching and erosion, and increasing the amount of nutrients in the crop root zone, trees in agroforestry moderate the cycle of nutrients [7]. Agroforestry trees, particularly leguminous trees, enrich soil through biological nitrogen fixation, addition of organic matter and recycling of nutrients [8].

Global agricultural practices today incorporate a variety of non-symbiotic bacteria (such as *Azotobacter*, *Azospirillum*, *Bacillus*, and *Klebsiella sp.*) as well as symbiotic bacteria (specifically *Rhizobium sp.*) to enhance plant productivity [9]. Meanwhile, microorganisms play a crucial role in mitigating the issues associated with the use of chemical fertilizers and pesticides. They are widely employed in natural agricultural land and organic farming to address these concerns [10].

Over the past three decades, one of the busiest sectors of the global agricultural economy has been oilseed production. Oilseeds comprise 13% of the gross cultivated area, 3% of the GNP, and 10% of the nation's total agricultural commodities in terms of value (GoI, 2022). The primary oilseed crop in India is mustard because of its outstanding adaptability to conventional agricultural methods. Productivity of mustard can be enhanced due to microorganisms in soil [11]. Keeping these facts in view, there is a need to identify the suitable methods of crop establishment and land-use system for enhanced population of soil microbes under mustard.

2. MATERIALS AND METHODS

2.1 Details of the Experiment

A field experiment took place under *Dalbergia sissoo* agroforestry model and open system at Research Farm, Department of Forestry, College of Agriculture, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur, Madhya Pradesh. The experimental site is positioned at an elevation of 391 meters above sea level. It is situated at a latitude of 23° 12' 50" north and a longitude of 79° 57' 56" east in the Kymore Plateau and Satpura Hills, agroclimatic zones of Madhya Pradesh. The climate is characterized by scorching, arid summers with an average maximum temperature of 46°C and frigid, arid winters with an average minimum temperature of 4°C. Jabalpur experiences an average annual rainfall of 1350 mm. The majority of rain received during mid-June to the end of September. The remaining months, particularly from December to February, receive just 75 mm of rainfall due to the influence of westerly winds. The region is renowned for its elevated relative humidity levels, ranging from 80 to 90%, 60 to 75% and 20 to 23% during the rainy, summer and winter, respectively.

The experiment arranged in a split-split plot design, with land-use systems (S_1 : agroforestry system and S_2 : open system) as the main factors. Three sub factors were crop establishment methods (M_1 : broadcasting, M_2 : line sowing, and M_3 : transplanting) and four sub-sub factors consisted of boron levels (B_0 : 0 kg ha⁻¹, B_1 : 1 kg ha⁻¹ as basal, B_2 : 2 kg ha⁻¹ as basal and B_3 : ½ kg ha⁻¹ as basal + ½ kg ha⁻¹ as foliar just before flowering) during the *Rabi* seasons in the years 2022–23 and 2023–24. Hence, a total number of 24 different treatment combinations ($S_1M_1B_0$, $S_1M_1B_1$, $S_1M_1B_2$, $S_1M_1B_3$, $S_1M_2B_0$, $S_1M_2B_1$, $S_1M_2B_2$, $S_1M_2B_3$, $S_1M_3B_0$, $S_1M_3B_1$, $S_1M_3B_2$, $S_1M_3B_3$, $S_2M_1B_0$, $S_2M_1B_1$, $S_2M_1B_2$, $S_2M_1B_3$, $S_2M_2B_0$, $S_2M_2B_1$, $S_2M_2B_2$, $S_2M_2B_3$, $S_2M_3B_0$, $S_2M_3B_1$, $S_2M_3B_2$, $S_2M_3B_3$) were replicated thrice. The objective of implementing the agrisilviculture system with *Dalbergia sissoo* (tree component) and *Bassica juncea* (crop component) was to optimize productivity across different boron concentrations and crop establishment methods. Mustard was planted using three different spacing methods for crop establishment: 45 x 15 cm for transplanting, 30 x 10 cm for line sowing, and unevenly scattered for broadcasted in the plots. Crop was sown in plots measuring 3.6 x 15 m in the alley, with *Dalbergia*

sissoo trees with the RDF (80:40:40 N: P: K kg ha⁻¹, respectively). The trees were arranged in a square pattern with a spacing of five meters in both directions.

2.2 Soil Sampling and Analysis

After harvesting the crop in a test field, a comprehensive soil samples were collected in order to assess the population of soil microbiota. In order to get spatial diversity, five distinct sampling locations were chosen at random within each treatment plot and using an auger device from the rhizospheric zone (0-15 cm deep). Once the soil samples were gathered and subjected to determination of microbial population.

The required quantity of media was measured and mixed with the prescribed volume of distilled water in a conical flask. The components were completely dissolved in the distilled water through boiling. The autoclave was utilized to sterilize the media for a duration of 15 minutes at a temperature of 121.6°C at a pressure of 15 pounds. And a concentrated cell culture diluted to a more manageable concentration through serial dilution. To prepare solidified agar plates for the purpose of selectively separating different microorganisms, 100 µl of the appropriate dilution were applied onto the plates. The BOD incubator was maintained at a temperature of 37±2°C in order to facilitate the incubation of culture plates. Observations were recorded after a 48-hour period to enumerate the colonies. The number of colony-forming units per gram of soil was counted by tallying each type of microorganisms.

The data collected was analysed statistically as per the standard analysis of various to variance to draw valid conclusion [12].

3. RESULTS AND DISCUSSION

3.1 Effect of Systems

The data presented in Tables 1 and 2 revealed that microbial counts significantly changed due to change of land-use systems. The highest counts of azotobacter (31.37 CFU x 10⁵), rhizobium (21.99 CFU x 10⁶), total bacteria (42.78 CFU x 10⁶), total fungi (8.30 CFU x 10⁵) and actinomycetes (12.98 CFU x 10⁵) were recorded under Agroforestry system. The higher counts of microbes under Agroforestry system might be due to the presence of abundant quantity of

organic matter and moisture favouring the proliferation of microbes. These findings are in accordance with Beule et al. [13] Beule et al. [14] Lori et al. [15] and López-Ramírez, et al. [16].

Table 1. Changes in population of Azotobacter and Rhizobium and Total Bacterial count in rhizospheric soil under different treatments

Microbes	Azotobacter (CFU x 10 ⁵ g ⁻¹ soil)			Rhizobium (CFU x 10 ⁶ g ⁻¹ soil)			Total Bacteria (CFU x 10 ⁶ g ⁻¹ soil)		
	Y ₁	Y ₂	Pooled	Y ₁	Y ₂	Pooled	Y ₁	Y ₂	Pooled
Land-use Systems									
S ₁	30.93	31.80	31.37	21.17	22.80	21.99	43.70	41.85	42.78
S ₂	24.10	25.21	24.66	14.43	16.02	15.23	24.84	23.12	23.98
SEm±	0.50	0.40	0.45	0.31	0.32	0.28	0.59	0.51	0.55
C. D. (P=0.05)	3.03	2.45	2.7	1.91	1.96	1.70	3.58	3.13	3.35
Crop Establishment Methods									
M ₁	29.19	29.98	29.59	19.48	21.08	20.28	38.45	36.59	37.52
M ₂	27.33	28.56	27.94	17.66	19.25	18.45	34.26	32.58	33.42
M ₃	26.03	26.97	26.50	16.27	17.91	17.09	30.09	28.29	29.19
SEm±	0.40	0.48	0.38	0.28	0.31	0.26	0.33	0.40	0.36
C. D. (P=0.05)	1.31	1.58	1.25	0.91	1.01	0.83	1.09	1.32	1.18
Boron Levels (kg ha⁻¹)									
B ₀	26.96	27.91	27.43	17.30	19.29	18.30	34.13	32.05	33.09
B ₁	27.31	28.19	27.75	17.58	19.35	18.47	34.24	32.37	33.3
B ₂	27.70	28.80	28.25	17.96	19.46	18.71	34.31	32.61	33.46
B ₃	28.10	29.11	28.61	18.37	19.54	18.96	34.39	32.93	33.66
SEm±	0.41	0.39	0.31	0.28	0.37	0.24	0.41	0.33	0.27
C. D. (P=0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS

Y₁ – 2022-23; Y₂ – 2023-24

Table 2. Changes in population of Total fungi and Actinomycetes count in rhizospheric soil under different treatments

Microbes	Total Fungi (CFU x 10 ⁵ g ⁻¹ soil)			Actinomycetes (CFU x 10 ⁵ g ⁻¹ soil)		
	Y ₁	Y ₂	Pooled	Y ₁	Y ₂	Pooled
Land-use Systems						
S ₁	6.90	9.70	8.30	11.99	13.97	12.98
S ₂	4.16	7.07	5.62	9.10	11.08	10.09
SEm±	0.11	0.16	0.13	0.18	0.20	0.19
C. D. (P=0.05)	0.65	0.98	0.8	1.08	1.23	1.15
Crop Establishment Methods						
M ₁	6.61	9.34	7.97	11.69	13.66	12.68
M ₂	5.41	8.30	6.85	10.40	12.38	11.39
M ₃	4.59	7.50	6.04	9.55	11.55	10.55
SEm±	0.06	0.09	0.07	0.15	0.26	0.17
C. D. (P=0.05)	0.21	0.31	0.22	0.48	0.86	0.57
Boron Levels (kg ha⁻¹)						
B ₀	5.40	8.26	6.83	10.41	12.37	11.39
B ₁	5.50	8.33	6.91	10.49	12.49	11.49
B ₂	5.56	8.43	6.99	10.59	12.57	11.58
B ₃	5.67	8.52	7.09	10.69	12.68	11.69
SEm±	0.10	0.11	0.08	0.24	0.19	0.17
C. D. (P=0.05)	NS	NS	NS	NS	NS	NS

Y₁ – 2022-23; Y₂ – 2023-24

3.2 Effect of Crop Establishment Methods

According to data in relation to soil microbial population (Tables 1 and 2), the population of soil microbes was significantly influenced due to crop establishment methods. Broadcasting method accommodated to significantly higher counts of azotobacter, rhizobium, total bacteria, total fungi and actinomycetes (29.59 CFU x 10⁵, 20.28 CFU x 10⁶, 37.52 CFU x 10⁶, 7.97 CFU x 10⁵ and 12.68 CFU x 10⁵, respectively) than in the line sowing and transplanting. This might be due to the spatial configuration of the crop in different crop establishment methods had different densities of plants which regulate the evaporation of moisture as well as interception of sunlight and ultimately the population of microbes. Similar conclusions were reached by Romdhane *et al.* [17] Kim *et al.* [18] and dos Santos Cordeiro *et al.* [19,20].

3.3 Effect of the Boron Levels

Data presented in Tables 1 and 2 pertaining to soil microbial population, revealed that the levels of boron did not show any significant impact on the rhizobia, Azotobacter, other bacterial population, total fungi and actinomycetes.

4. CONCLUSION

Based on two-year study it could be concluded that the Agroforestry system proved to be more congenial for the proliferation of rhizospheric microbes as compared to the open. Further, the microbial population counts varied with crop establishment methods beneath the *Dalbergia sissoo*. The marked higher microbial population was found under the broadcasting method over the other methods of crop establishment. The increase in rhizospheric bacteria especially, azotobacter throughout the study indicates a potential enhancement in nitrogen fixation within the soil. Additionally, the observed rise population count of total fungi and actinomycetes could support the decomposition of organic matter, thus contributing to a higher organic carbon content in the soil.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Vibha B, Neelam G. Importance of exploration of microbial biodiversity. *Int. Res. J. Biological Sci.* 2012;1(3):78-83.
2. Javed Z, Tripathi GD, Mishra M, Dashora K. Actinomycetes—the microbial machinery for the organic-cycling, plant growth, and sustainable soil health. *Biocatalysis and Agricultural Biotechnology.* 2021;31: 101893.
3. Tate III RL. Microorganisms, ecosystem disturbance and soil-formation processes. In *Soil Reclamation Processes Microbiological Analyses and Applications.* CRC Press. 2020;1-34.
4. Basu S, Kumar G, Chhabra S, Prasad R. Role of soil microbes in biogeochemical cycle for enhancing soil fertility. In *New and future developments in microbial biotechnology and bioengineering.* Elsevier. 2021;149-157.
5. Sonune N. Microbes: A potential tool for bioremediation. *Rhizobiont in Bioremediation of Hazardous Waste.* 2021;391-407.
6. Pandey A, Tiwari P, Manpoong C, Jatav HS. Agroforestry for restoring and improving soil health. In *agroforestry to combat global challenges: Current prospects and future challenges.* Singapore: Springer Nature Singapore. 2024;147-164.
7. Tilak MR, KR–Vaiyapuri KS, B–Kumar P. The current role and importance of agroforestry– A review article. *Applied ecology and environmental research.* 2024;22(5):3907-3918.
8. Nayak MR, Mishra PJ, Garnyak LM, Maharana JR. *Agroforestry: a land use system for improving soil health; 2024.*
9. Jalal A, Filho MCMT, da Silva EC, da Silva Oliveira CE, Freitas LA, do Nascimento V. Plant growth-promoting bacteria and nitrogen fixing bacteria: Sustainability of non-legume crops. In *nitrogen fixing bacteria: Sustainable growth of non-legumes.* Singapore: Springer Nature Singapore. 2022;233-275
10. Bokade P, Gaur VK, Tripathi V, Bobate S, Manickam N, Bajaj A. Bacterial remediation of pesticide polluted soils:

- exploring the feasibility of site restoration. *Journal of Hazardous Materials*. 2023; 441:129906.
11. Singh A, Singh N, Kalhapure AH, Singh SB, Gupta AK, Gupta D, Gupta D. Effect of Nutrient Management on Physico-chemical Properties of Soil in Indian Mustard. *International Journal of Plant & Soil Science*. 2024;36(7):610-617.
 12. Gomez KA, Gomez AA. *Statistical procedures for agricultural research*. John Wiley & Sons; 1984.
 13. Beule L, Corre MD, Schmidt M, Göbel L, Veldkamp E, Karlovsky P. Conversion of monoculture cropland and open grassland to agroforestry alters the abundance of soil bacteria, total fungi and soil-N-cycling genes. *PloS one*. 2019;14(6):e0218779.
 14. Beule L, Vaupel A, Moran-Rodas VE. Abundance, diversity, and function of soil microorganisms in temperate alley-cropping agroforestry systems: A review. *Microorganisms*. 2022;10(3):616.
 15. Lori M, Armengot L, Schneider M, Schneidewind U, Bodenhausen N, Mäder P, Krause HM. Organic management enhances soil quality and drives microbial community diversity in cocoa production systems. *Science of the total environment*. 2022;834:155223.
 16. López-Ramírez TM, Estrada-Medina H, Ferrer MM, O'Connor-Sánchez A. Divergence in the soil and rhizosphere microbial communities of monoculture and silvopastoral traditional *C. dodecandra* agroforestry systems in Yucatan, Mexico. *Soil use and management*. 2023; 39(3): 1205-1218.
 17. Romdhane S, Spor A, Busset H, Falchetto L, Martin J, Bizouard F, Cordeau S. Cover crop management practices rather than composition of cover crop mixtures affect bacterial communities in no-till agroecosystems. *Frontiers in microbiology*. 2019;10:1618.
 18. Kim N, Zabaloy MC, Guan K, Villamil MB. Do cover crops benefit soil microbiome? A meta-analysis of current research. *Soil Biology and Biochemistry*. 2020;142: 107701.
 19. dos Santos Cordeiro CF, Echer FR, Araujo FF. Cover crops impact crops yields by improving microbiological activity and fertility in sandy soil. *Journal of Soil Science and Plant Nutrition*. 2021;21(3):1 968-1977.
 20. Schulz S, Brankatschk R, Dümig A, Kögel-Knabner, Schloter M. The role of microorganisms at different stages of ecosystem development for soil formation. *Biogeosciences*. 2013;10:3983-3996.

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of the publisher and/or the editor(s). This publisher and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.

© Copyright (2024): Author(s). The licensee is the journal publisher. This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:

The peer review history for this paper can be accessed here:

<https://www.sdiarticle5.com/review-history/122881>