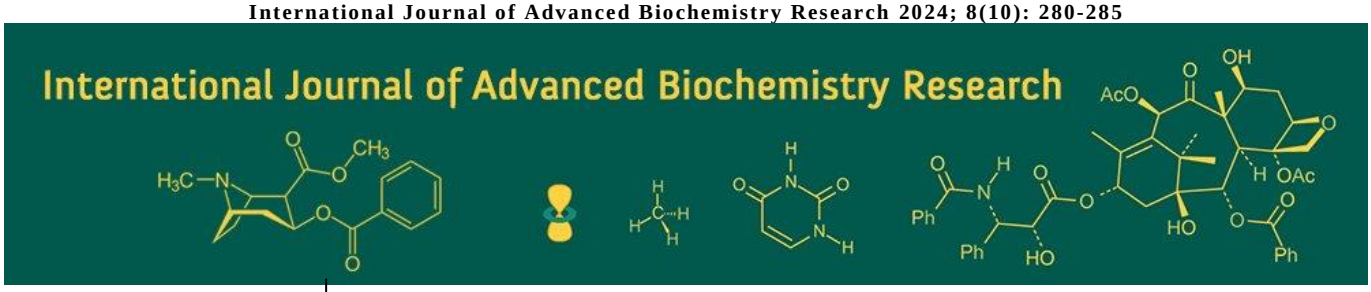

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Assessment of the soil rhizospheric microbiome of maize as influenced under various fertigation levels and weed management practices

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Abstract
A field investigation was carried out at the experimental field of the AICRP on Weed Management of the Department of Agronomy, Dr. P.D.K.V., Akola (M.S.), India, during *kharif* 2021-22, to study the impact of various fertigation levels and weed management practices on the microbial population dynamics of the rhizospheric soil of maize. The experiment was designed in a split-plot design with four main plot treatments of fertigation levels and five sub-plot weed management treatments, which were replicated three times. The recommended dose of fertilizer applied was 120:60:60 NPK kg/ha. Among the weed management treatments, results clearly showed that treatment, weedy check, recorded significantly higher microbial population at all stages of observation, which was found to be on par with the farmer's practice of 2 hoeing at a 15-20 days interval after sowing *fb* 2 HW. Whereas, among weed management treatments using herbicides, atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS, recorded a higher soil microbial count, which was found at par with treatment of atrazine 0.75 kg/ha PE *fb* tembotrione 0.120 kg/ha PoE 20 DAS at all stages of observation after application of treatments. However, the effect of various fertigation levels as well as the interaction of fertigation levels and weed management treatments on the microbial count was found to be non-significant.

Keywords: Maize, herbicide, topramezone, bacteria, fungi and actinomycetes

Introduction
Maize (*Zea mays* L.) is a cereal crop with tremendous potential, earning it the titles "Queen of Cereals" and "King of Fodders." Maize is an important staple crop in India and has significant economic value. It is known as the "Queen of cereals" due to its exceptional yield-producing potential and its unparalleled adaptability to a wide range of environmental conditions (Rani *et al.* 2024) [9].

Weeds are detrimental or objectionable and impede the proper growth and development of maize. They are in significant rivalry with crops for water, nutrients, sunlight and space, among other growth factors (Tyagi *et al.* 2018) [12].

Weed management is crucial for achieving higher productivity, especially during the early stages of maize growth. Traditional and cultural practices alone are expensive to control the weed problem. Weed control by physical labour is not only expensive and time-consuming, but also labour availability is not readily accessible when needed. Therefore, the use of appropriate herbicides would be a more advantageous alternative from an economic standpoint (Ramesh and Nadanassababady, 2005) [8].

Traditional weed management methods may be time-consuming and financially expensive. So, utilizing herbicides during the early phases of crop growth or in a sequential manner might effectively address weed issues that arise later in the crop's development (Devi *et al.* 2017) [1].

Since herbicides are chemicals, using them without studying their effects could have unintended consequences on soil such as altering the population and activity levels of soil bacteria. The microbial population of soil bacteria, fungus and actinomycetes were significantly reduced after application of herbicide treatments, which corroborated the results of Jing *et al.* (2010) [5] and Ramesh and Nadanassababady (2005) [8]. The widespread use of herbicides in agriculture poses a risk to disrupt the fragile biological equilibrium inside the

soil (Grossbard, 1976) [3]. For this reason, understanding how herbicides impact soil microbial populations is crucial for assessing soil health. Keeping this view, the current experiment was carried out to obtain insight into the impacts of varying fertigation levels and weed management treatments over soil microbial population.

Materials and Methods

A field investigation was conducted at the experimental field section of the AICRP on Weed Management, Department of Agronomy, Dr. P.D.K.V., Akola (M.S.), India, during the *kharif* season of 2021–22, to examine the effects of different fertigation levels and weed management practices on the microbial population dynamics within the soil rhizosphere of maize. The experimental site was situated at 20.703494° latitude and 77.030027° longitude. The experiment was laid out in a split-plot design with three replications. There were four main plot treatments (three different fertigation levels and one treatment of soil application of 100% RDF) and five sub-plot treatments, which were different weed management treatments. The main plot treatments comprised of different levels of fertigation, *viz.*, (F_2) drip fertigation with 75% RDNK ha^{-1} in 5 splits, (F_3) drip fertigation with 100% RDNK ha^{-1} in 5 splits and (F_4) drip fertigation with 125% RDNK ha^{-1} in 5 splits and were compared with (F_1) 100% RDF soil application (120:60:60) NPK kg/ha (N in 3 splits). P as basal dose applied to all main-plot treatments. 5 splits in fertigation treatments were applied at Basal, 20, 40, 60 and 80 DAS. Whereas, sub-plot treatments comprised of five weed management practices, *viz.*, (W_1) atrazine 0.75 kg/ha PE *fb* tembotrione 0.120 kg/ha PoE 20 DAS; (W_2) atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS; (W_3) atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS; (W_4) farmer's practice of 2 hoeing 15-20 days interval after sowing *fb* 2 HW and (W_5) weedy check. The maize variety "Syngenta NK 6240 plus Hybrid Corn" was used for experimentation. The recommended dose of fertilizers applied was 120:60:60 NPK kg/ha. It was sown on June 16th, 2021 and harvested on October 6th, 2021. Pre-emergence and postemergence herbicides were applied as per the respective treatments. Observations were recorded, before the sowing stage, the next day after the spraying of the pre-emergence herbicide stage, five days after the spraying of the post-emergence herbicide stage and at harvest, respectively.

Dynamics of microbial populations

Composite soil samples were obtained from each treatment plot before sowing, following the application of pre-emergence herbicide, subsequent after the application of post-emergence herbicide, and at harvest to determine the soil microbial populations of fungi, actinomycetes, and bacteria *via* a serial dilution technique.

The medium was prepared and sterilized by autoclaving. Starting with a 1-gram soil sample, we used the serial dilution approach to dilute it in 10 ml of sterilized water (Wollum (1982) [13], Dhingra and Sinclair (1995) [2] and Sharma (2012) [10].

Bacterial count

After spreading pre-prepared Nutrient Agar (NA) medium on petri plates, we collected the supernatant (1 ml) in a pipette and diluted it serially to a concentration of 10^{-6}.

Fungi Count

Once the supernatant (1 ml) was diluted by a factor of 10^{-5}, it was pipetted onto petri plates containing Potato Dextrose Agar (PDA). The bacterial growth in the PDA medium was inhibited by the addition of Rose Bengal.

Actinomycetes count

The supernatant (1 ml) from a serial dilution of 10^{-4} was collected and spread out in a uniform manner on petri plates of Munairs and Kenknight's medium. The poured plates (NA, PDA, and Munairs and Kenknight) were incubated for a period of 72 hours at 28±1 °C. A colony counter was used to count the number of fungal, actinomycetes, and bacterial colonies expressed in CFU.

Results and Discussion

Soil microbial analysis (bacteria, fungi and actinomycetes count)

Soil micro-flora was observed at different stages, *viz.*, before sowing, after pre-emergence herbicide spraying, after post-emergence herbicide spraying and at harvest, to evaluate the impact of different fertigation levels and herbicide application regimes on microbial count (fungi, bacteria and actinomycetes). Different treatments of weed control had a considerable impact on the microbial dynamics. The findings demonstrate that there was a non-significant difference in microbial count before sowing, but the microbial count dropped precipitously when pre-emergence herbicide treatments were applied. However, after 20 DAS, population growth rate picked up might be due to the rhizospheric environment provided by the crop and weed densities as well as increased moisture availability in the soil and microbial count increased at harvest due to increased moisture availability due to rains at harvest. The current research clearly shows that microorganism populations were temporarily suppressed but subsequently rebounded in the soil depending upon moisture availability and herbicide impact over a period of time.

1. Microbial count (bacteria)

The data pertaining to microbial count (bacteria) influenced by different fertigation levels and weed management treatments (CFU g^{-1} 10^{-6}) at periodical growth stages in maize during 2021-22 was presented in Table 1 and expressed graphically in Fig. 1. The mean microbial count (bacteria), *viz.*, before sowing, after pre-emergence herbicide spraying, after post-emergence herbicide spraying and at harvest was 52.80 CFU g^{-1} 10^{-6}, 48.77 CFU g^{-1} 10^{-6}, 53.05 CFU g^{-1} 10^{-6} and 59.47 CFU g^{-1} 10^{-6}, respectively.

Effect of various fertigation levels

The findings demonstrated that at any stage of crop development, the microbial count was not noticeably affected by the four varied fertigation levels. This result agrees with those of Thakare (2019) [11] and Patil *et al.* (2024) [7].

Effect of various weed management practices

The bacterial count was observed to be considerably higher at all stages in the treatment of weedy check, i.e., W_5, which was found to be on par with the treatment W_4, i.e., the farmer's practice of 2 hoeing at a 15-20 days interval after sowing *fb* 2 HW at all stages after application of weed management treatments till harvest. It's possible that the

high bacterial count in both treatments as compared to herbicide treatments might be due to the fact that no chemical herbicides were applied here. Tyagi *et al.* (2018) [12] and Xian *et al.* (2019) [14] found similar outcomes. One possible explanation for the declining periodic soil bacterial count in herbicide application treatment might be due to the effect that pre-emergence herbicides and post-emergence herbicides have on the metabolic activities of soil bacteria (Milosevic and Govedarica 2000) [6]. Whereas among all herbicide treatments, treatment W_2, i.e., atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS, recorded a higher soil bacterial count, which was found at par with treatment W_1, i.e., atrazine 0.75 kg/ha PE *fb* tembotrione 0.120 kg/ha PoE 20 DAS at all stages of observation after application of treatments. The use of herbicides resulted in a reduction in the microbial population as compared to weedy check and weed-free treatments. Comparable outcomes were reported by Jaybhaye *et al.* (2021) [4] and Patil *et al.* (2024) [7]. However, treatment W_3, i.e., atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS recorded the lowest bacterial count among the weed management treatments at all stages of observation.

Interaction effect

Weed management treatments and fertigation levels were not observed to interact significantly with respect to the bacterial count. All of these findings are consistent with Thakare (2019) [11] and Patil *et al.* (2024) [7].

2. Microbial count (fungi)

The data in respect to soil microbial count (fungi) as influenced by different fertigation levels and weed management treatments (CFU g^{-1} soil 10^{-5}) at periodical growth stages in maize during 2021-22 was presented in Table 2 and illustrated graphically in Fig. 2. The mean microbial count (fungi) before sowing, after pre-emergence herbicide spraying, after post-emergence herbicide spraying and at harvest was 18.37 CFU g^{-1} soil 10^{-5}, 16.98 CFU g^{-1} soil 10^{-5}, 22.22 CFU g^{-1} soil 10^{-5} and 25.96 CFU g^{-1} soil 10^{-5} respectively.

Effect of various fertigation levels

The results showed that the fungal count was not significantly influenced by different fertigation levels at any stage of crop growth. The results of the present research agree with those of Thakare (2019) [11] and Patil *et al.* (2024) [7].

Effect of various weed management practices

The data clearly showed that various weed management treatments have substantial effects on the total number of fungal counts. As may be seen from the data, the findings show that the weedy check (W_5) treatment had the higher fungal count, which was found to be comparable with the treatment W_4, i.e., the farmer's practice of 2 hoeing at a 15-20-day interval after sowing *fb* 2 HW. Thakare (2019) [11] and Patil *et al.* (2024) [7] also discovered comparable results, suggesting that the higher fungal count in both treatments may be attributable to the lack of chemical use in the treatments. Herbicide-treated plots had lower total fungal counts compared to the treatments of weedy check treatment, i.e., W_5 and treatment (W_4) i.e., farmer's practice -2 hoeing at a 15-20 days interval after sowing *fb* 2 HW. Pre- and post-emergence herbicide applications result in a periodic decrease in the microbial count due to their effects on the metabolic activities of soil microbes (Milosevic and Govedarica 2000) [6]. Thakare (2019) [11], Jaybhaye *et al.* (2021) [4] and Patil *et al.* (2024) [7] found similar reductions in the fungal count as compared to weedy check and weed-free treatments.

Among herbicidal treatments, treatment W_2, i.e., atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS, recorded a significantly higher fungal count at all stages as compared to all other herbicide treatments. But it was found on par with treatments W_1, i.e., atrazine 0.75 kg/ha PE *fb* tembotrione 0.120 kg/ha PoE 20 DAS and W_3, i.e., atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS. However, treatment W_3, i.e., atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS, recorded the lowest fungal count as compared to all other weed management treatments at all stages. Possible causes include chemical herbicides' knock-on effect of reducing the fungal population, causing a setback in the fungal population. Thakare (2019) [11] reported similar results.

Interaction effect

An interaction effect with respect to fungal count was found to be non-significant between various fertigation levels and weed management practices. These findings are consistent with those of Thakare (2019) [11] and Patil *et al.* (2024) [7].

3. Microbial count (actinomycetes)

The data pertaining to microbial count (actinomycetes) as influenced by different fertigation levels and weed management treatments (CFU g^{-1} soil × 10^{-4}) at periodical growth stages in maize during 2021-22 was presented in Table 3 and expressed graphically in Fig 3. The mean microbial count (actinomycetes) before sowing, after pre-emergence herbicide spraying, after post-emergence herbicide spraying and at harvest was 18.70 CFU g^{-1} soil × 10^{-4}, 16.42 CFU g^{-1} soil × 10^{-4}, 18.92 CFU g^{-1} soil × 10^{-4} and 21.48 CFU g^{-1} soil × 10^{-4} respectively.

Effect of various fertigation levels

None of the treatments of fertigation levels significantly influenced the actinomycetes count throughout the course of the whole observation period. These findings are consistent with the results of Thakare (2019) [11] and Patil *et al.* (2024) [7].

Effect of various weed management practices

The findings clearly revealed that the total number of actinomycetes was greatly influenced by the various weed management practices. Findings showed that the weedy check (W_5) treatment recorded higher values in respect of actinomycetes counts. Those results of the treatment weedy check (W_5) found to be on par with the treatment (W_4) i.e., the farmer's practice of 2 hoeing at a 15-20 days interval after sowing *fb* 2 HW. It might be because chemical herbicides weren't applied in those treatments, i.e., in treatment W_4 and treatment W_5. Xian *et al.* (2019) [14], Thakare (2019) [11] and Patil *et al.* (2024) [7] also discovered similar results. Whereas, treatment W_2, i.e., atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS, recorded a higher actinomycetes count at all stages of observation as compared to all other herbicide treatments, which was found to be comparable with W_1, i.e., atrazine 0.75 kg/ha PE *fb* tembotrione 0.120 kg/ha PoE 20 DAS and

atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS (W_3). However, treatment W_3, i.e., atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS, resulted in the lowest actinomycetes count as compared to all other weed management treatments. Possible causes include chemical herbicides' effect on reducing the microbial population, causing a setback in the actinomycetes count. Patil *et al.* (2024) [7] reported similar results.

Interaction effect

An interaction effect was found to be non-significant between various fertigation levels and weed management practices with respect to actinomycetes count. These findings are consistent with those of Thakare (2019) [11] and Patil *et al.* (2024) [7].

Table 1: Microbial count (bacteria) as influenced by different fertigation levels and weed management treatments (CFU g^{-1} soil 10^{-6}) at periodical growth stages of *kharif* maize during 2021-22.

Treatments	2021-22			
	Before sowing	After spraying-Pre-emergence herbicide	After spraying-Post-emergence herbicide	At harvest
A. Fertigation levels				
F_1-100% RDF (soil application) (120:60:60) NPK kg ha^{-1} (N in 3 splits)	51.67	47.67	51.40	57.81
F_2-75% RDNK in 5 splits	51.20	47.00	51.20	57.41
F_3-100% RDNK in 5 splits	52.87	48.80	52.67	59.71
F_4-125% RDNK in 5 splits	55.47	51.60	56.93	62.94
SE (m) ±	1.19	1.21	1.28	1.22
CD at 5 %	NS	NS	NS	NS
B. Weed management practices				
W_1- Atrazine 0.75 kg/ha PE *fb* tembotrione 0.120 kg/ha PoE 20 DAS	53.75	44.67	47.83	55.50
W_2- Atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS	52.75	46.08	50.08	57.17
W_3 – Atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS	47.00	39.75	45.00	49.96
W_4- Farmer's practice- 2 hoeing 15-20 days interval after sowing *fb* 2 HW	54.67	55.92	59.58	65.75
W_5- Weedy check	55.83	57.42	62.75	68.97
SE (m) ±	2.10	1.00	1.07	1.54
CD at 5 %	NS	2.88	3.19	4.43
C. Interaction (FL X WM)				
SE (m) ±	4.21	2.00	2.15	3.07
CD at 5 %	NS	NS	NS	NS
GM	52.80	48.77	53.05	59.47

Table 2: Microbial count (fungi) as influenced by different fertigation levels and weed management treatments (CFU g^{-1} soil 10^{-5}) at periodical growth stages of *kharif* maize during 2021-22

Treatments	2021-22			
	Before sowing	After spraying-Pre-emergence herbicide	After spraying-Post-emergence herbicide	At harvest
A. Fertigation levels				
F_1-100% RDF (soil application) (120:60:60) NPK kg ha^{-1} (N in 3 splits)	17.07	15.87	20.67	24.69
F_2-75% RDNK in 5 splits	16.93	15.80	20.53	24.47
F_3-100% RDNK in 5 splits	19.47	18.00	22.73	26.68
F_4-125% RDNK in 5 splits	20.00	18.27	24.93	28.01
SE (m) ±	2.28	0.93	1.22	0.88
CD at 5 %	NS	NS	NS	NS
B. Weed management practices				
W_1- Atrazine 0.75 kg/ha PE *fb* tembotrione 0.120 kg/ha PoE 20 DAS	16.50	12.75	17.00	20.08
W_2- Atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS	17.08	13.50	18.58	22.50
W_3 – Atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS	16.17	12.33	16.67	19.67
W_4- Farmer's practice- 2 hoeing 15-20 days interval after sowing *fb* 2 HW	20.33	22.83	29.00	33.00
W_5- Weedy check	21.75	23.50	29.83	34.57
SE (m) ±	1.61	0.78	1.00	1.07
CD at 5 %	NS	2.25	2.88	3.09
C. Interaction (FL X WM)				
SE (m) ±	3.23	1.56	2.00	2.14
CD at 5 %	NS	NS	NS	NS
GM	18.37	16.98	22.22	25.96

Table 3: Microbial count (actinomycetes) as influenced by different fertigation levels and weed management treatments (CFU g^{-1} soil × 10^{-4}) at periodical growth stages of *kharif* maize during 2021-22

Treatments	2021-22			
	Before sowing	After spraying-Pre-emergence herbicide	After spraying-Post-emergence herbicide	At harvest
A. Fertigation levels				
F_1-100% RDF (soil application) (120:60:60) NPK kg ha^{-1} (N in 3 splits)	18.47	15.93	18.00	20.73
F_2-75% RDNK in 5 splits	17.13	14.40	17.07	20.20
F_3-100% RDNK in 5 splits	18.67	16.67	19.40	21.60
F_4-125% RDNK in 5 splits	20.53	18.67	21.20	23.40
SE (m) ±	1.15	0.98	0.99	1.01
CD at 5 %	NS	NS	NS	NS
B. Weed management practices				
W_1- Atrazine 0.75 kg/ha PE *fb* tembotrione 0.120 kg/ha PoE 20 DAS	17.17	13.17	15.42	17.92
W_2- Atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS	17.92	13.58	15.92	18.67
W_3 – Atrazine 0.75 kg/ha PE *fb* halosulfuron-methyl 0.052 kg/ha PoE 20 DAS	15.50	11.92	13.58	15.75
W_4- Farmer's practice- 2 hoeing 15-20 days interval after sowing *fb* 2 HW	20.17	20.50	23.42	26.00
W_5- Weedy check	22.75	22.92	26.25	29.08
SE (m) ±	1.73	0.91	1.18	1.22
CD at 5 %	NS	2.62	3.40	3.50
C. Interaction (FL X WM)				
SE (m) ±	3.47	1.82	2.36	2.43
CD at 5 %	NS	NS	NS	NS
GM	18.70	16.42	18.92	21.48

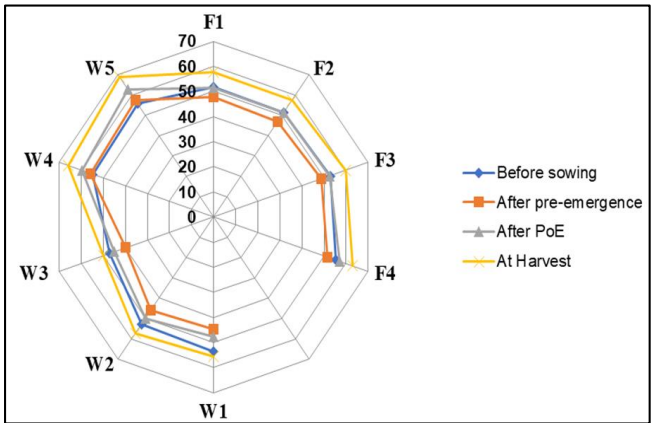

Fig 1: Microbial count (bacteria) as influenced by different fertigation levels and weed management treatments (CFU g^{-1} soil 10^{-6}) at periodical growth stages of *kharif* maize during 2021-22.

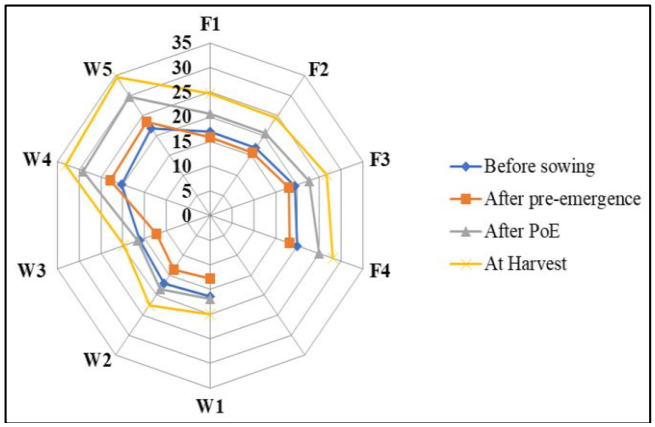

Fig 2: Microbial count (fungi) as influenced by different fertigation levels and weed management treatments (CFU g^{-1} soil 10^{-5}) at periodical growth stages of *kharif* maize during 2021-22

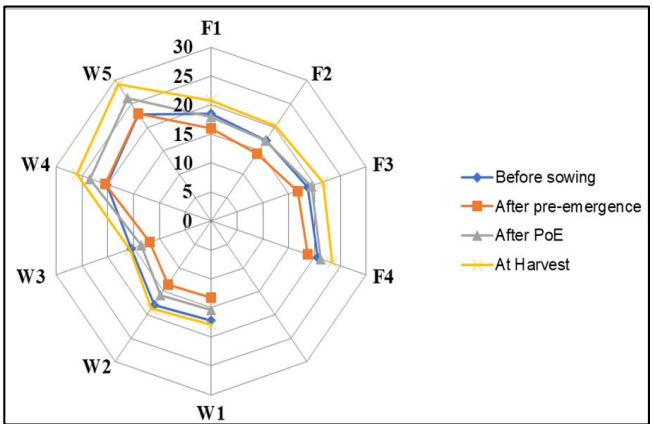

Fig 3: Microbial count (actinomycetes) as influenced by different fertigation levels and weed management treatments (CFU g^{-1} soil × 10^{-4}) at periodical growth stages of *kharif* maize during 2021-22

Conclusions

On the basis of various weed management practices, the weedy check treatment recorded a significantly higher microbial count of bacteria, fungi, and actinomycetes. However, among the various herbicide treatments atrazine 0.75 kg/ha PE *fb* topramezone 0.0252 kg/ha PoE 20 DAS recorded a higher microbial count of bacteria, fungi, and

actinomycetes at all stages as compared to all other treatments.

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