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The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2023; 12(11): 135-140

www.thepharmajournal.com
Received: 19-09-2023
Accepted: 26-10-2023

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Bioassay studies in *Kharif* maize (*Zea mays* L.) as influenced by different weed management practices

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Abstract
A field experiment on maize was carried out during *kharif*, 2022 at Main Agricultural Research Station, University of Agricultural Science, Raichur. The experiment was laid out in Randomized Complete Block Design with three replications and eleven treatments. Among the different herbicide treatments, 2,4-D amine salt 58% SL @ 500 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS had recorded significantly maximum population of bacteria, fungi, actinomycetes and dehydrogenase activity at both first (17.67×10^6 cfu g^{-1} soil, 16.00×10^4 cfu g^{-1} soil, 65.33×10^3 cfu g^{-1} soil and 18.99 µg TPF g^{-1} soil day^{-1}) and second week after herbicide spray (21.33×10^6 cfu g^{-1} soil, 18.33×10^4 cfu g^{-1} soil, 71.33×10^3 cfu g^{-1} soil and 22.35 µg TPF g^{-1} soil day^{-1}) and it was statistically comparable with application of mesotrione 48% SC @ both 96 and 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds alone and also when they were *fb* IC at 45 DAS at first and second week after herbicide spray.

Keywords: Maize, mesotrione, intercultivation (IC), dehydrogenase activity

Introduction
Maize, the Indian and American word for corn means literally which "sustains life". It is the third most important cereal in India in terms of both area and production after rice and wheat, providing nutrients for humans, animals and serving as a basic raw material for the production of starch, oil, protein, alcoholic beverages, food sweeteners and more recently, fuel. The green plant made into silage has been used with much success in the dairy and beef industries. After harvest of the grain, the dried leaves and upper part including the flowers are still used today to provide relatively good forage for ruminant animals owned by many small farmers in developing countries. The erect stalks, which in some varieties are strong, have been used as long-lasting fences and walls. In developed countries more than 60 per cent of the production is used in compounded feeds for poultry, pigs and ruminant animals (Anon., 1992) [1].

Inspite of the production potential, weed growth in the maize fields affects its growth and hence there is a scope for utilizing economic weed management strategy. Sequential application of pre and post-emergent herbicides or combination of herbicides and cultural methods is one such strategy.

Materials and Methods
A field experiment was laid out in Randomized Complete Block Design with eleven treatments and three replications during *kharif*, 2022 at Main Agricultural Research Station, UAS, Raichur. The maize hybrid (NK-6240) seeds were sown on 12th July, 2022 with a spacing of 60 × 20 cm when there was sufficient moisture in *vertisols* of the experimental site. The crop was fertilized with recommended dose of fertilizers (150:75:37.5 kg NPK ha^{-1}, respectively).

The soil from experimental site was powdered and 10 g was mixed in 90 ml sterilized water to give 10^{-1} dilution. Subsequently dilutions upto 10^{-6} were made by transferring serially 1 ml of the dilution into 9 ml water blank. The blanks and the media poured in the petri plates for isolating different microbes were sterilized for 3 hours in an autoclave at 121 °C and then inoculated with 0.1 ml of appropriate dilutions and spread using a sterile glass rod. Then, the inoculated plates were kept for incubation at 34 ± 1 °C for the appropriate time specified for each microbe's growth and emerged colonies were counted.

Each soil sample was sieved through the 1000 micromesh to remove the bigger particles and debris and was used for isolation of bacteria by serial dilution agar plate technique using Nutrient Agar medium, enumeration of fungi using Martin's Rose Bengal Agar (MRBA) medium and enumeration of actinomycetes using Actinomycetes Isolate Agar medium by standard plate count method. The 10^{-6}, 10^{-4} and 10^{-3} dilution of soil suspension was used for isolation, respectively. The plates were incubated for 24 hours, 4 days and 6 days, respectively at 28 °C. The colonies that appeared on the respective media were enumerated and expressed in terms of cfu g^{-1} of soil on dry weight basis.

The dehydrogenase activity in the soil samples was determined by following the procedure as described by Casida *et al.* (1964) [4].

Results and Discussion

Weed parameters

There was significant variation in the weed density and dry weight recorded at 30 DAS than compared to 15 DAS *i.e.*, before herbicide spray as presented in Table 1.

At 15 DAS, significantly higher weed density and dry weight was observed in the treatment receiving mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS (9.65 m^{-2} and 4.82 g m^{-2}, respectively) which was statistically on par with weedy check (9.51 m^{-2} and 4.78 g m^{-2}, respectively), mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds (9.41 m^{-2} and 4.68 g m^{-2}, respectively), mesotrione 48% SC @ 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds (9.34 m^{-2} and 4.64 g m^{-2}, respectively), whilst significantly lower weed density and dry weight was noticed in weed free check (0.71 m^{-2} and 0.71 g m^{-2}, respectively).

At 30 DAS, significantly higher total density and dry weight of weeds was recorded in weedy check (12.08 m^{-2} and 7.37 g m^{-2}, respectively), whereas lower density and dry weight was observed from weed free check (0.71 m^{-2} and 0.71 g m^{-2}, respectively). The second best treatment was farmers' practice wherein significantly lower total weed density and dry weight (5.56 m^{-2} and 2.85 g m^{-2}, respectively) was observed. However, it was statistically on par with other herbicide treatments except application of 2,4-D amine salt 58% SL @ 500 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS (6.85 m^{-2} and 3.33 g m^{-2}, respectively), mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds (7.39 m^{-2} and 3.42 g m^{-2}, respectively) and mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS (7.48 m^{-2} and 3.50 g m^{-2}, respectively).

Application of mesotrione herbicide decreased the total weed density and dry weight by obstructing the weed growth through reduced photosynthetic rates, carotenoids formation, auxin and 4-hydroxy phenylpyruvate dioxygenase (HPPD) inhibition during the critical period of crop-weed competition. These findings are in line with the findings of Dey (2020) [5] in maize, Baban (2022) [3] in *kharif* sweet corn and Sairam *et al.* (2023) [11] in maize. Likewise, Harisha *et al.* (2023) [7] reported that significantly lower density and dry weight of weeds in maize was recorded in the treatment receiving application of mesotrione 48% SC @ 288 ml *a.i.* ha^{-1}.

Table 1: Weed density and dry weight as influenced by different weed management practices in maize

Treatment	Weed density (no. m^{-2})		Weed dry weight (g m^{-2})	
	15 DAS	30 DAS	15 DAS	30 DAS
T_1: Mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	9.41 (88.00)*	7.39 (54.13)	4.68 (21.36)	3.42 (11.21)
T_2: Mesotrione 48% SC @ 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	9.34 (86.67)	6.02 (35.73)	4.64 (21.03)	3.05 (8.82)
T_3: Mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	9.10 (82.40)	5.84 (33.60)	4.59 (20.56)	2.98 (8.40)
T_4: Mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	9.65 (92.53)	7.48 (55.47)	4.82 (22.74)	3.50 (11.77)
T_5: Mesotrione 48% SC @ 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	9.08 (81.87)	5.97 (35.20)	4.51 (19.86)	3.02 (8.63)
T_6: Mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	9.21 (84.27)	5.82 (33.33)	4.58 (20.51)	2.98 (8.35)
T_7: Tembotrione 34.4% SC @ 125 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	8.88 (78.40)	5.68 (31.73)	4.34 (18.36)	2.92 (8.05)
T_8: 2,4-D amine salt 58% SL @ 500 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	8.91 (78.93)	6.85 (46.40)	4.34 (18.30)	3.33 (10.60)
T_9: Farmers' practice (HW at 20 DAS *fb* IC at 40 DAS)	9.02 (80.80)	5.56 (30.40)	4.44 (19.21)	2.85 (7.61)
T_{10}: Weed free check	0.71 (0.00)	0.71 (0.00)	0.71 (0.00)	0.71 (0.00)
T_{11}: Weedy check	9.51 (89.87)	12.08 (145.33)	4.78 (22.34)	7.37 (53.75)
S.Em.±	0.16	0.16	0.07	0.08
L.S.D. at 5%	0.46	0.47	0.22	0.22

*Figures in parentheses indicate original values Original data subjected to square root transformation : $\sqrt{(x+0.50)}$

a.i. = Active ingredient DAS = Days after sowing *fb* = followed by HW = Hand weeding

IC = Intercultivation SC = Suspension concentrate SL = Soluble liquid

Microbial population

The data on the microbial population (bacteria, fungus and actinomycetes) and dehydrogenase activity was recorded before herbicide spray, first week after herbicide spray and second week after herbicide spray which differed significantly except before herbicide spray as there was no complete treatment imposition.

After one week of herbicide application, significant difference was observed in the bacterial, fungal and actinomycetes population among the different weed management practices (Table 2 & 3 and Plate 1). The major reduction in microbial population was noticed because of herbicides application.

Among the different treatments, the maximum bacterial, fungal and actinomycetes population was recorded in weedy check (28.67 × 10^6, 23.00 × 10^4 and 84.00 × 10^3 cfu g^{-1} soil, respectively), while the minimum population of bacteria was observed in the treatment receiving application of mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds (14.00 × 10^6, 13.00 × 10^4 and 58.67 × 10^3 cfu g^{-1} soil, respectively) after first week of herbicides spray. Among the herbicide treatments, application of 2,4-D amine salt 58% SL @ 500 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS had recorded higher bacterial, fungal and actinomycetes population (17.67 × 10^6, 16.00 × 10^4 and 65.33 × 10^3 cfu g^{-1} soil, respectively) after first week of herbicides spray which was statistically on par with weed free check (21.00 × 10^6, 16.67 × 10^4 and 69.67 × 10^3 cfu g^{-1} soil, respectively).

Two weeks after herbicide spray also, notable variation was observed in the microbial population. Among the different treatments, the maximum bacterial, fungal and actinomycetes population (31.33 × 10^6, 25.33 × 10^4 and 89.67 × 10^3 cfu g^{-1} soil, respectively) was recorded in weedy check, whereas minimum microbial populations were observed in the plots receiving application of mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds (17.00 × 10^6, 14.67 × 10^4 and 67.33 × 10^3 cfu g^{-1} soil, respectively) after second week of herbicides spray. Among the herbicide treatments, application of 2,4-D amine salt 58% SL @ 500 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS had recorded higher bacterial, fungal and actinomycetes population (21.33 × 10^6, 18.33 × 10^4 and 71.33 × 10^3 cfu g^{-1} soil, respectively) after second week of herbicides spray which was statistically on par with weed free check (23.00 × 10^6, 18.33 × 10^4 and 72.33 × 10^3 cfu g^{-1} soil, respectively) and farmers' practice (23.67 × 10^6, 18.67 × 10^4 and 73.00 × 10^3 cfu g^{-1} soil, respectively).

Table 2: Bacterial and fungal population in soil as influenced by different weed management practices in maize

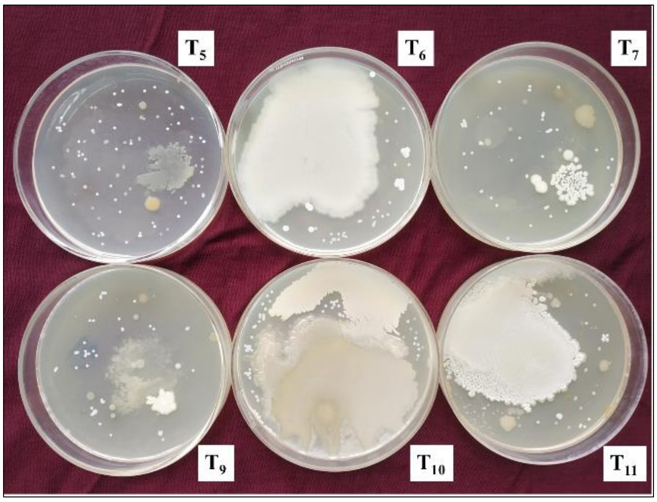
Treatment	Bacterial population (× 10^6 cfu g^{-1} soil)			Fungal population (× 10^4 cfu g^{-1} soil)		
	BHS	1st WAHS	2nd WAHS	BHS	1st WAHS	2nd WAHS
T_1: Mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	25.67	16.33	20.33	20.33	14.67	16.67
T_2: Mesotrione 48% SC @ 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	25.33	16.33	20.00	20.00	14.33	16.33
T_3: Mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	24.67	14.00	17.00	19.33	13.00	14.67
T_4: Mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	27.00	16.67	21.33	21.67	14.67	17.33
T_5: Mesotrione 48% SC @ 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	24.33	15.67	20.33	19.00	14.33	16.00
T_6: Mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	25.00	15.00	18.67	19.67	13.67	15.00
T_7: Tembotrione 34.4% SC @ 125 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	23.00	15.33	19.33	18.00	14.00	15.67
T_8: 2,4-D amine salt 58% SL @ 500 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	23.67	17.67	21.33	18.67	16.00	18.33
T_9: Farmers' practice (HW at 20 DAS *fb* IC at 40 DAS)	24.00	22.33	23.67	19.00	17.33	18.67
T_{10}: Weed free check	26.33	21.00	23.00	20.67	16.67	18.33
T_{11}: Weedy check	26.00	28.67	31.33	20.67	23.00	25.33
S.Em. ±	1.65	1.37	1.24	1.14	1.16	1.22
L.S.D. at 5%	NS	4.04	3.65	NS	3.43	3.61

cfu = Colony forming unit BHS = Before herbicide spray WAHS = Week after herbicide spray
a.i. = Active ingredient DAS = Days after sowing *fb* = followed by HW = Hand weeding
IC = Intercultivation NS = Non-significant SC = Suspension concentrate SL = Soluble liquid

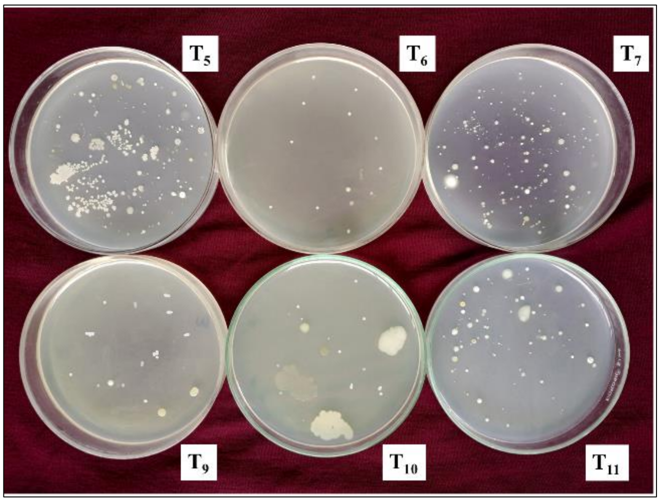
Table 3: Actinomycetes population and dehydrogenase activity in soil as influenced by different weed management practices in maize

Treatment	Actinomycetes population (× 10^3 cfu g^{-1} soil)			Dehydrogenase activity (µg TPF g^{-1} soil day^{-1})		
	BHS	1st WAHS	2nd WAHS	BHS	1st WAHS	2nd WAHS
T_1: Mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	76.67	63.00	70.67	26.21	18.21	21.78
T_2: Mesotrione 48% SC @ 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	76.00	61.33	70.00	26.13	17.28	21.56
T_3: Mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds	75.00	58.67	67.33	25.83	15.99	20.67
T_4: Mesotrione 48% SC @ 96 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	79.33	63.33	71.00	26.47	18.62	21.98
T_5: Mesotrione 48% SC @ 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	75.00	61.33	69.67	25.75	17.03	21.43
T_6: Mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	75.33	59.67	66.67	26.08	16.41	21.01
T_7: Tembotrione 34.4% SC @ 125 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	73.00	60.00	68.33	25.16	16.53	21.11
T_8: 2,4-D amine salt 58% SL @ 500 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS	74.33	65.33	71.33	25.21	18.99	22.35
T_9: Farmers' practice (HW at 20 DAS *fb* IC at 40 DAS)	74.67	70.67	73.00	25.62	22.80	24.12
T_{10}: Weed free check	78.67	69.67	72.33	26.36	22.56	23.89
T_{11}: Weedy check	78.33	84.00	89.67	26.29	28.22	30.87
S.Em. ±	3.09	2.00	1.89	1.06	0.95	0.93
L.S.D. at 5%	NS	5.90	5.57	NS	2.80	2.74

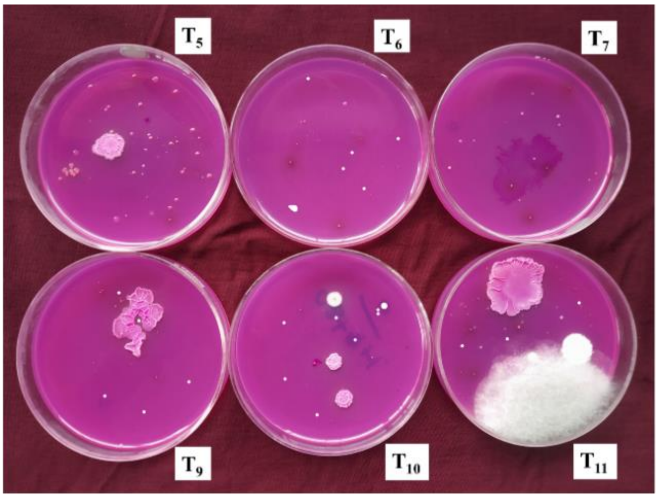
cfu = Colony forming unit TPF = Triphenylformazan BHS = Before herbicide spray WAHS = Week after herbicide spray
a.i. = Active ingredient DAS = Days after sowing *fb* = followed by HW = Hand weeding
IC = Intercultivation NS = Non-significant SC = Suspension concentrate SL = Soluble liquid



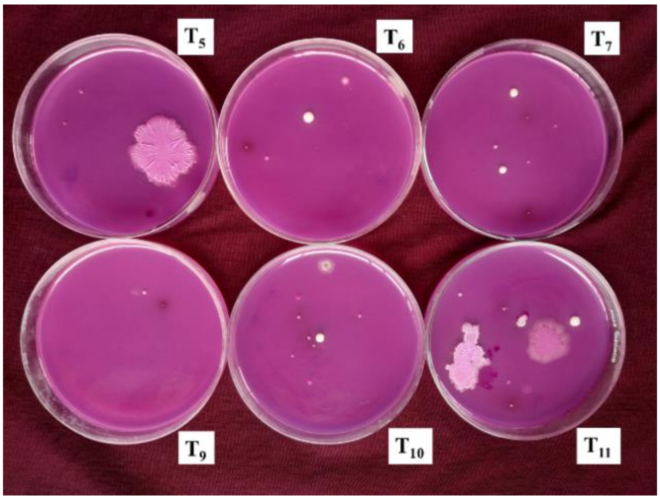
Bacterial population
before herbicide spray



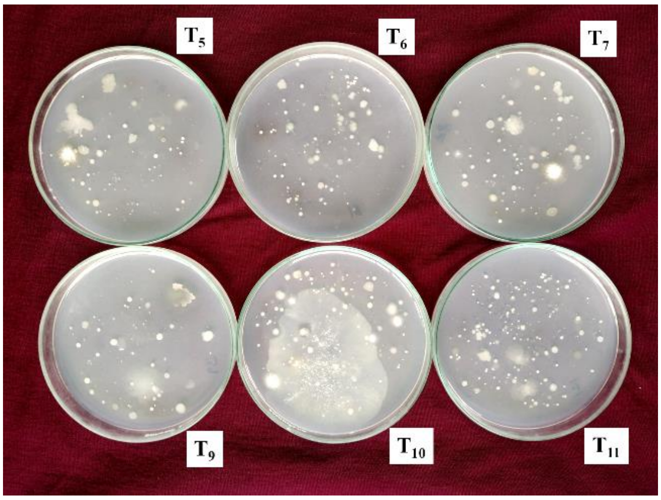
Bacterial population
first week after herbicide spray



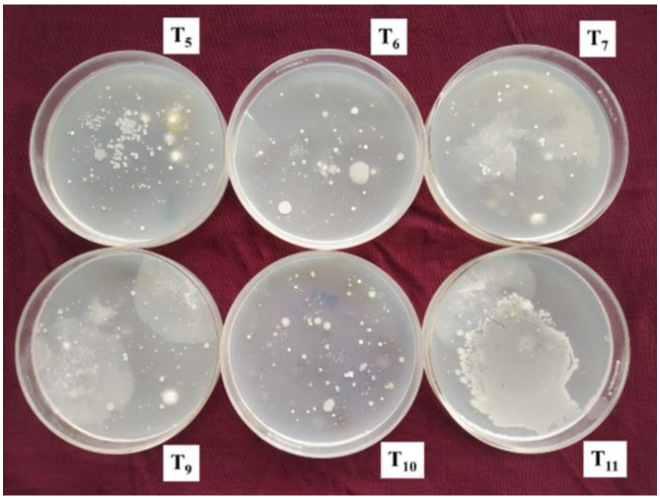
Fungal population
before herbicide spray



Fungal population
first week after herbicide spray

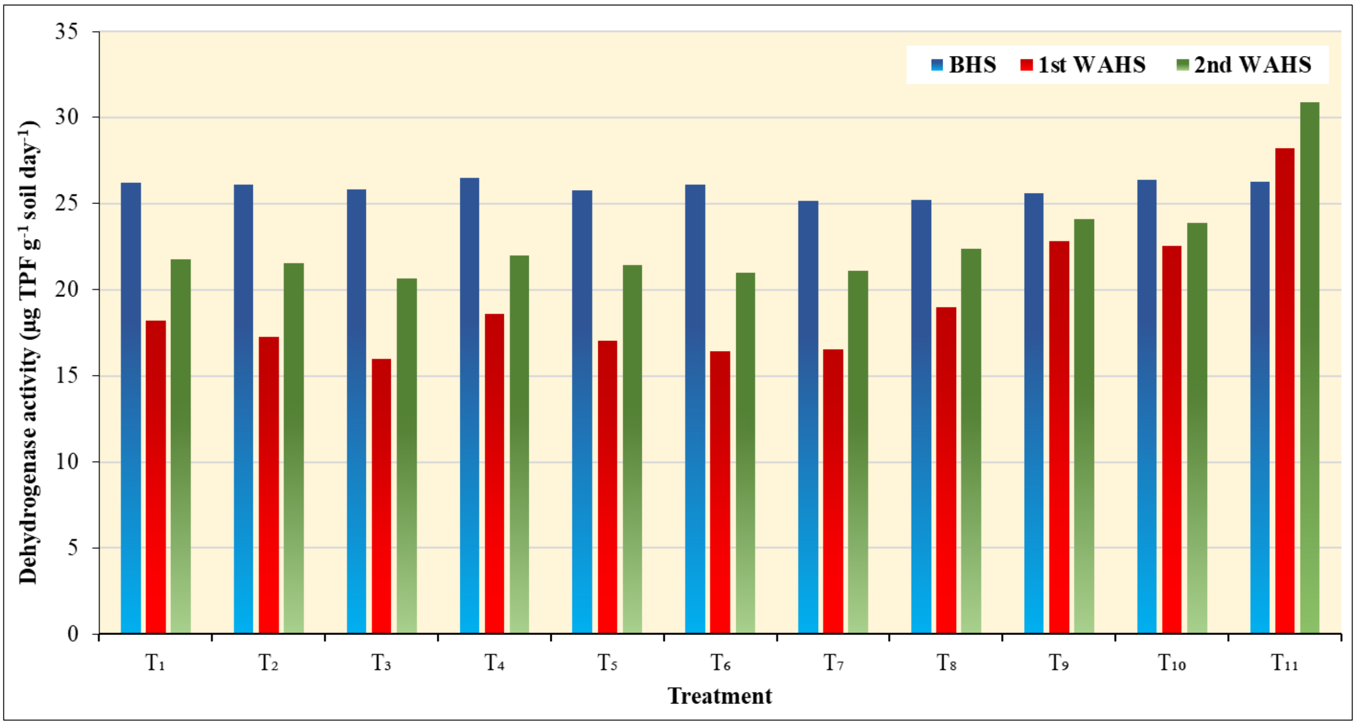


Actinomycetes population
before herbicide spray



Actinomycetes population
first week after herbicide spray

Plate 1: Microbial population before herbicide spray and first week after herbicide spray as influenced by different weed management practices in maize



Note: BHS = Before herbicide spray WAHS = Week after herbicide spray

Fig 1: Dehydrogenase activity (µg TPF g^{-1} soil day^{-1}) as influenced by different weed management practices in maize

The significant difference and also decrease in microbial population after one week of herbicides application might be because of the toxic effect of herbicides on the growth of bacteria in the rhizosphere soil which resulted in the death of these microorganisms. But interestingly, there was rise in the microbial population after two weeks of herbicide spray which could be due to depletion of herbicides in the soil either because of leaching with the rain water into the soil or the toxic effects on the microbial population might be short-lived. It might also be due to consumption of carbon source present in the herbicide molecule by bacteria.

Such outcomes were also noticed by Modak *et al.* (2019) [8] in *rabi* maize and Arunkumar *et al.* (2020) [2] in *kharif* maize. Similarly, Tyagi *et al.* (2018) [12] also observed higher bacterial population at 10 and 20 days after application of herbicides due to application of acetochlor 90% EC @ 1.25 l ha^{-1} in *rabi* maize and higher fungal population at 10 and 20 days after application of 2,4-D ethyl ester 38% EC @ 2.65 l ha^{-1} in *rabi* maize. Paul *et al.* (2023) [9] observed significantly higher actinomycetes population in the application of pendimethalin 38.7% CS @ 1 kg ha^{-1} as pre-emergence at 3 DAS followed by one hand weeding at 25 DAS.

Dehydrogenase activity (µg TPF g^{-1} soil day^{-1})

The data on the dehydrogenase activity analysed at one week after herbicides spray revealed that the maximum dehydrogenase activity (28.22 µg TPF g^{-1} soil day^{-1}) was observed in weedy check, whilst the minimum activity was noticed in the plots receiving mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds (15.99 µg TPF g^{-1} soil day^{-1}). (Table 3 and Figure 1).

The soil samples analysed after two weeks of herbicides application revealed that the maximum dehydrogenase activity was observed in weedy check treatment (30.87 µg TPF g^{-1} soil day^{-1}), whereas the minimum dehydrogenase activity was observed in the plots treated with mesotrione 48% SC @ 144 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds (20.67 µg TPF g^{-1} soil day^{-1}). The next best treatment was farmers' practice with higher dehydrogenase activity (24.12 µg TPF g^{-1} soil day^{-1}) which was statistically on par with the weed free check (23.89 µg TPF g^{-1} soil day^{-1}).

The variation in dehydrogenase activity might be due to the application of herbicides and the spread of those molecules on the soil that led to their toxic effect on the microbial population during first week of herbicide spray. Once the residue of the chemicals was degraded in the soil, there was gradual improvement in the microbial population after second week of herbicide application thereby clearly indicating increase in dehydrogenase activity as the chemicals effect was short-lived and completely depleted thereafter due to the regain in the microbial population. Similar variations in the dehydrogenase activity were also reported in the experiments of Tyagi *et al.* (2018) [12] in *rabi* maize, Sabiry and Babu (2019) [10], Arunkumar *et al.* (2020) [2] and Paul *et al.* (2023) [9] in *kharif* maize. Besides, Goalla *et al.* (2022) [6] also noticed significantly higher dehydrogenase activity in the plots receiving application of tembotrione @ 120 g ha^{-1} at 15 DAS.

Conclusion

The treatment, mesotrione 48% SC @ 120 g *a.i.* ha^{-1} at 3-4 leaf stage of weeds *fb* IC at 45 DAS was the safer dose among the various herbicide treatments both for microbial population and dehydrogenase activity and hence it can be used as an effective weed management practice in maize.

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