



Conservation Agriculture Provides Greater Resilience to Soil Microorganisms from Perturbation Caused by Pendimethalin Herbicide

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Abstract

Assessment of soil microorganisms under different management practices helps in developing more productive and sustainable agricultural systems. Conservation agriculture (CA) systems are considered vital for soil health, biodiversity and environmental quality but the inevitable repetitive use of herbicides in these systems may have certain negative impacts including phyto-toxicity, carry-over effects on succeeding crops besides contamination of water resources, and hence poses threat to ecosystem. The literature on consequence of herbicides on soil microorganisms is quite controversial, and largely based either on laboratory or short-term field experiments. Therefore, we assessed the impact of pendimethalin herbicide on the population of bacteria, fungi and actinomycetes under CA system in practice for the last 13 years in comparison to conventional system involving intensive tillage in an alluvial sandy loam soil (Typic Haplustept). The experiment included two cropping systems (mungbean (*Vigna radiata* L.) - wheat (*Triticum aestivum* L.) and sorghum (*Sorghum bicolor* L.) - wheat) and three tillage practices, i.e., zero tillage with retention of crop residues in both summer and winter seasons (ZT-ZT), conventional tillage in summer and zero tillage with residue retention in winter (CT-ZT) and conventional tillage in both seasons (CT-CT). In contrast to extensively tillage based conventional, the CA system maintained significantly higher population of microorganisms in the soil at different depths. Bacteria were observed sensitive while fungi and actinomycetes moderate tolerance to pendimethalin but the growth of actinomycetes was quite fast towards the harvest of crops. The ZT-ZT practice increased the population of bacteria, fungi and actinomycetes by 44.5, 46.2 and 33.0%, respectively, over CT-CT practice, but the cropping systems did not influence the population. The study indicated that the CA system provided greater resilience to soil microorganisms from perturbation caused by pendimethalin as compared to the intensively tillage-based conventional system of farming.

Keywords: Conservation Agriculture; Zero Tillage; Pendimethalin; Herbicide; Soil Microorganisms; Cropping Systems

Introduction

Assessment of soil microorganisms under different management practices helps in developing more productive and sustainable agricultural systems. In soil, an array of micro- and macro-organisms performs a variety of natural functions including residue decomposition, nutrient storage and release, improvement in soil structure and its stability, resistance against disease and degradation of pollutants [1]. A soil with high biodiversity is considered

to be in good health inculcating greater resilience to soil from perturbation and maintaining environmental sustainability [2,3]. Conservation agriculture (CA) system involving retention of crop residues on the soil surface, minimum soil disturbance through tillage and diversified cropping systems has largely been proved to be beneficial in different ecosystems in terms of enhanced soil organic matter, better soil physical conditions and biodiversity over the extensively tillage based conventional systems of farming [4-7].

However, the increased and repetitive use of herbicides which is inevitable, particularly, upon shifting from persisting conventional to CA systems may influence soil biodiversity and the environmental quality [8,9]. The tendency of the increased population of weeds under CA further increases their use [10,11]. Therefore, the consequence of applied herbicides in modern agriculture on soil microorganisms is of greater concern. It is an established fact that the agricultural system that enhances the soil organic carbon content has direct control on the population and diversity of soil microorganisms, and in turn, influences the fate of the applied herbicide. The literature available on the effect of herbicides on agriculturally important soil microorganisms is quite controversial [12] varying from short-term transient depressing [13] to non-inhibitory [14] or even stimulatory effect on microbial growth [15,16] depending on the herbicide (type and concentration), the type of soil, prevailing environmental conditions, microbial species, and cultural practices being adopted during crop cultivation [17]. Such information is largely based either on the laboratory or short-term field experiments under different crops [18]. Therefore, to draw significant conclusions, it becomes necessary to conduct such studies under a long-term field experimentation involving major crops/cropping systems in the given region. Among different crops grown in India, wheat (*Triticum aestivum* L.) is the most important food crop after rice, particularly in the northern part, and staple diet for millions of Indians. Mungbean (*Vigna radiata* L.) is another short duration legume crop grown in the summer season fulfilling the requirement of protein in human diet while sorghum (*Sorghum bicolor* L.), though, used for food, fodder and production of alcohol beverages, but its stover often gains importance over grain as fodder for livestock. Both, mungbean and sorghum, being summer (rainy) season crops, face severe weed competition for soil moisture and nutrients throughout the growing period, and thus, chemical weed control has become indispensable for getting good yields. Therefore, the objective of the present study was to assess the impact of the application of pendimethalin herbicide, widely used selective herbicide for controlling annual grasses and broadleaf weeds in cereals, oilseeds, pulses, forage, fruits and vegetable crops [19], on the population of bacteria, fungi and actinomycetes in the soil under conservation tillage over the extensively tillage based conventional system followed in mungbean-wheat and sorghum-wheat cropping systems continuously for the last 13 years.

Materials and Methods

Site, treatments and weather conditions

The study was carried out on an ongoing long-term CA experiment initiated in the year 2006 at Research Farm (29°10' 0" N, 75°46' 0" E, 215.2 m msl) of the Department of Soil Science, CCS Haryana Agricultural University, Hisar, India, during summer season of 2019. The soil is an alluvial sandy loam (coarse loamy, calcareous, mixed, hyperthermic Typic Haplustept). It has 67.6, 11.1 and 21.3 g of sand, silt and clay, respectively, per 100 g of soil; low soluble salt content (EC 0.56 dS m⁻¹); 172.3 kg ha⁻¹ available N; 14.6 kg ha⁻¹ available P; 311.8 kg ha⁻¹ available K and 5.7 g kg⁻¹ of organic carbon content in surface 0.15 m soil depth and alkaline in reaction (pH 8.0). The climate is semiarid with annual rainfall of 429 mm, most of which occurs during July to August. Daily rainfall, pan evaporation and maximum and minimum temperature during the crop growing period (12 July to 17 September, 2019) are depicted in figure 1. The total rainfall received was 229 mm and pan evaporation was 292.5 mm. The maximum and minimum temperatures ranged within 27.9-39.1°C and 20.5- 30.3°C, respectively.

The experiment was laid out in split plot design with three tillage practices in the main plot- zero tillage in both summer and winter seasons (ZT-ZT), conventional tillage in summer and zero tillage in winter (CT-ZT) and conventional tillage in both seasons (CT-CT), and two cropping systems in sub-plots- mungbean-wheat and sorghum-wheat without replications. The main plot was 24m × 72m and the sub-plot was 24 m × 35 m. In ZT plots, the residues of the preceding wheat crop were left on the surface as mulch at the rate of 4 t ha⁻¹. No mechanical seed-bed preparation was done.

Seed and fertilizers were placed in the single slot opened by the zero-till seed drill, while keeping the rest of the area undisturbed. In CT plots, the residues of the previous wheat crop were removed and the seed-bed tilth was prepared by two disc harrowing followed by one cultivation to a depth of ~0.10-0.12 m and then planking of the plots. Mungbean (variety MH 421) and sorghum (variety MSSH 4) were sown on 12 July 2019. Both crops were sown with 0.30 m row-row and 0.10 m plant-plant spacing. For mungbean, 15 kg of N and 40 kg of P₂O₅ ha⁻¹ were applied at the time of sowing; whereas for sorghum, 50 kg of N and 15 kg of P₂O₅ ha⁻¹ were applied as basal and 25 kg N ha⁻¹ top-dressed four weeks after sowing. Due to the appropriate distribution of rainfall during the crop growing sea-

son, no irrigation was needed for mungbean but one irrigation was applied at 45 days after sowing (DAS) for sorghum. The crops were harvested on 17 September 2019.

Herbicide application

Pendimethalin (N-(1-ethylpropyl)-3,4-dimethyl 2,6-dinitro benzamine), commercially available as Penda (30 EC), was applied @ 1.0 kg a.i. ha⁻¹ using a knapsack sprayer with a flat fan nozzle immediately after sowing of the crops in a plot of 24 m × 4 m in each of the treatments.

Soil sampling

Soil samples from 0-0.05, 0.05-0.15 and 0.15-0.30 m depths at 0 (immediately after sowing), 3, 7, 15, 30 and 45 DAS, and at harvest of crops (68 DAS) were collected using 50 mm × 54 mm cylindrical steel cores from three different locations from each treatment plot and were considered as replicates. The samples were packed in polyethylene bags and brought to the laboratory for further processing and analysis.

Soil microbial count

The colony forming units (cfu) of bacteria, fungi and actinomycetes in soil samples collected from different depths from different treatments at different days after application of pendimethalin during the crop season were estimated immediately using serial dilution spread plate technique [20]. The colonies appeared on media plates were counted and microbial population per gram of the sample was calculated by multiplying the number of colonies observed with the dilution factor used.

Statistical analysis

The experimental data were evaluated by analysis of variance and the statistical significance ($P < 0.05$) within factors (tillage and cropping system) using OPSTAT, online statistical analysis software, CCS HAU, Hisar.

Results and Discussion

Impact of tillage practices on soil microorganisms

The viable population of soil microorganisms (bacteria, fungi and actinomycetes) in surface 0-0.05 m soil depth under various treatments measured at different days during the crop season after the application of pendimethalin (Table 1) showed that immedi-

ately after the application of the herbicide (0 day), the population of bacteria was highest ($21.8 \times 10^6 - 16.1 \times 10^6$ cfu g⁻¹ soil) followed by actinomycetes ($19.8 \times 10^5 - 15.7 \times 10^5$ cfu g⁻¹ soil) and least of fungi ($12.7 \times 10^3 - 9.7 \times 10^3$ cfu g⁻¹ soil). The ZT-ZT, *i.e.*, conservation agriculture (CA) system maintained a significantly higher population of the microorganisms at different soil depths throughout the crop season compared to other tillage treatments. The fungal and actinomycetes counts were also observed significantly higher under CT-ZT compared to CT-CT treatment. This was likely because of the proliferation of microorganisms upon relatively higher build up of soil organic matter under zero tillage practice followed either in both the seasons (ZT-ZT) or only in the winter season (CT-ZT) as compared to conventional tillage in both the seasons (CT-CT) which is desirable for better microbial growth [7].

The population decreased substantially with the increase in depth in soil profile due to the reduced amount of organic matter in the lower depths. In general, microorganisms degrade a variety of carbonaceous substances, including the accumulated herbicides in soil and utilize them as a source of energy for their metabolic activities and physiological processes [15,22,23] but before degradation, the targeted deposition of pre-emergent herbicides may lead to direct impact on sensitive soil microorganisms reducing their population and activities, and consequently diversity of their communities. Under field conditions, the large fraction of the applied herbicide accumulates in the soil at the surface few centimeters where most of the microbiological activities occur and there is hardly any possibility of total exposure of soil microorganisms to a biologically active concentration of herbicide, therefore, pendimethalin affected the population of the soil microorganisms significantly in surface 0.05 m depth than in the lower soil depths.

In surface 0.05 m soil depth, the toxic effect of the herbicide persisted up to the 7th day of its application as thereafter, the population started growing. At the 7th day of the application of herbicide, the maximum reduction in the population of microorganisms was observed under ZT-ZT practice as compared to the other tillage practices (Table 2), reducing bacteria the most (47%) followed by actinomycetes (17%) and fungi (12%). The reduction in bacterial population was relatively less under CT-CT (33%) than CT-ZT (41%) practice, but the population of fungi and actinomycetes remained almost similar under these two tillage treatments due to

| Sampling day | Tillage practice* | | | Cropping system | |
|---|-------------------|---------------|--------------|-----------------|---------------|
| | ZT-ZT | CT-ZT | CT-CT | Mungbean-wheat | Sorghum-wheat |
| 0-0.05 m | | | | | |
| 0 | 21.8 ± 1.3 a | 18.8 ± 1.3 b | 16.1 ± 1.3 c | 17.9 ± 1.4 a | 19.8 ± 1.6 a |
| 3 | 12.4 ± 0.9 a | 11.5 ± 1.8 a | 11.1 ± 1.6 a | 11.1 ± 1.6 a | 12.3 ± 1.6 a |
| 7 | 11.5 ± 1.1 a | 11.1 ± 1.6 a | 10.7 ± 1.7 a | 10.5 ± 1.7 a | 11.4 ± 1.5 a |
| 15 | 20.4 ± 1.7 a | 17.2 ± 1.8 ab | 15.8 ± 1.7 b | 16.8 ± 1.7 a | 18.5 ± 1.7 a |
| 30 | 24.8 ± 1.6 a | 19.1 ± 1.3 b | 17.4 ± 1.4 b | 19.8 ± 1.8 a | 20.7 ± 1.4 a |
| 45 | 26.5 ± 0.9 a | 21.3 ± 1.1 b | 18.6 ± 1.2 c | 21.8 ± 1.6 a | 22.5 ± 1.7 a |
| 68 | 26.3 ± 0.8 a | 21.7 ± 1.5 b | 18.2 ± 1.4 c | 21.2 ± 1.5 a | 23.1 ± 1.6 a |
| Mean | 20.5 | 17.2 | 15.4 | 17.0 | 18.3 |
| LSD (<i>P</i> < 0.05): Tillage practice, 2.8; cropping system, NS; interaction, NS | | | | | |
| 0.05-0.15 m | | | | | |
| 0 | 16.2 ± 1.0 a | 13.9 ± 0.6 b | 13.2 ± 0.6 b | 13.6 ± 1.1 a | 15.2 ± 1.3 a |
| 3 | 16.4 ± 0.9 a | 14.6 ± 0.7 b | 13.5 ± 0.8 b | 14.3 ± 1.5 a | 15.4 ± 1.2 a |
| 7 | 16.5 ± 0.8 a | 14.6 ± 0.8 b | 13.5 ± 0.7 b | 14.2 ± 1.2 a | 15.4 ± 1.3 a |
| 15 | 16.2 ± 1.1 a | 14.4 ± 1.0ab | 13.7 ± 0.5 b | 14.3 ± 1.2 a | 15.3 ± 1.3 a |
| 30 | 16.9 ± 0.7 a | 14.5 ± 0.8 b | 13.1 ± 0.7 b | 14.6 ± 1.4 a | 15.2 ± 1.5 a |
| 45 | 18.7 ± 1.1 a | 16.2 ± 0.8 b | 14.3 ± 0.6 c | 15.8 ± 1.2 a | 16.6 ± 1.4 a |
| 68 | 19.5 ± 1.2 a | 16.8 ± 1.0 b | 14.7 ± 0.7 c | 16.6 ± 1.3 a | 18.0 ± 1.3 a |
| Mean | 17.2 | 15.0 | 13.7 | 14.8 | 15.9 |
| LSD (<i>P</i> < 0.05): Tillage practice, 2.1; cropping system, NS; interaction, NS | | | | | |

a) Bacteria (× 10⁶cfu g⁻¹ soil)

| Sampling day | Tillage practice* | | | Cropping system | |
|---|-------------------|--------------|--------------|-----------------|---------------|
| | ZT-ZT | CT-ZT | CT-CT | Mungbean-wheat | Sorghum-wheat |
| 0-0.05 m | | | | | |
| 0 | 12.7 ± 0.6 a | 10.7 ± 0.6 b | 09.7 ± 0.6 b | 10.6 ± 0.9 a | 11.4 ± 1.0 a |
| 3 | 12.3 ± 0.8 a | 10.1 ± 0.3 b | 09.2 ± 0.3 c | 10.4 ± 0.8 a | 10.6 ± 0.9 a |
| 7 | 11.2 ± 0.8 a | 09.9 ± 0.7 b | 09.0 ± 0.7 b | 09.8 ± 1.1 a | 10.2 ± 1.0 a |
| 15 | 12.4 ± 0.7 a | 09.9 ± 0.7 b | 09.0 ± 0.7 b | 09.9 ± 0.8 a | 11.9 ± 0.8 a |
| 30 | 13.0 ± 0.5 a | 10.3 ± 0.5 b | 09.3 ± 0.5 b | 10.6 ± 0.7 a | 11.2 ± 0.8 a |
| 45 | 13.8 ± 0.7 a | 11.3 ± 0.6 b | 10.3 ± 0.5 b | 11.6 ± 0.9 a | 11.1 ± 0.9 a |
| 68 | 14.6 ± 0.8 a | 11.7 ± 0.5 b | 10.6 ± 0.5 b | 12.7 ± 1.1 a | 12.4 ± 1.0 a |
| Mean | 12.9 | 10.6 | 9.6 | 10.8 | 11.3 |
| LSD (<i>P</i> < 0.05): Tillage practice, 2.0; cropping system, NS; interaction, NS | | | | | |
| 0.05-0.15 m | | | | | |
| 0 | 11.1 ± 0.4 a | 10.0 ± 0.6 b | 09.8 ± 0.5 b | 10.1 ± 0.8 a | 10.4 ± 0.7 a |
| 3 | 11.0 ± 0.5 a | 09.8 ± 0.5 b | 09.6 ± 0.6 b | 10.2 ± 0.9 a | 10.0 ± 0.7 a |

| | | | | | |
|---|--------------|---------------|--------------|--------------|--------------|
| 7 | 10.9 ± 0.5 a | 09.7 ± 0.6 b | 09.2 ± 0.4 b | 10.1 ± 0.6 a | 09.8 ± 0.4 a |
| 15 | 10.6 ± 0.6 a | 09.7 ± 0.7ab | 09.1 ± 0.3 b | 09.9 ± 0.5 a | 09.8 ± 0.4 a |
| 30 | 10.8 ± 0.4 a | 10.2 ± 0.5 ab | 09.6 ± 0.6 b | 10.1 ± 0.6 a | 10.3 ± 0.4 a |
| 45 | 11.5 ± 0.7 a | 10.7 ± 0.7 ab | 09.8 ± 0.6 b | 10.7 ± 1.2 a | 10.7 ± 0.8 a |
| 68 | 12.6 ± 0.6 a | 11.3 ± 0.6 b | 10.4 ± 0.5 b | 11.2 ± 1.1 a | 11.7 ± 0.9 a |
| Mean | 11.2 | 10.2 | 9.6 | 10.3 | 10.4 |
| LSD (<i>P</i> < 0.05): Tillage practice, 1.4; cropping system, NS; interaction, NS | | | | | |

b) Actinomycetes (× 10⁵cfu g⁻¹ soil)

| Sampling day | Tillage practice* | | | Cropping system | |
|--|-------------------|---------------|--------------|-----------------|---------------|
| | ZT-ZT* | CT-ZT | CT-CT | Mungbean-wheat | Sorghum-wheat |
| 0-0.05 m | | | | | |
| 0 | 19.8 ± 1.0 a | 16.4 ± 0.9 b | 15.7 ± 0.6 b | 17.0 ± 1.8 a | 17.5 ± 1.6 a |
| 3 | 17.3 ± 1.4 a | 15.2 ± 1.5 ab | 14.7 ± 0.8 b | 15.5 ± 1.4 a | 16.1 ± 1.7 a |
| 7 | 16.5 ± 1.3 a | 14.2 ± 1.4 ab | 13.9 ± 0.9 b | 14.6 ± 1.2 a | 15.2 ± 1.0 a |
| 15 | 17.6 ± 1.2 a | 15.2 ± 1.4 ab | 14.3 ± 0.8 b | 15.0 ± 1.1 a | 16.8 ± 1.6 a |
| 30 | 20.5 ± 1.2 a | 17.9 ± 1.2 b | 16.2 ± 0.8 b | 17.5 ± 1.4 a | 19.1 ± 1.5 a |
| 45 | 22.8 ± 1.3 a | 18.8 ± 1.1 b | 17.0 ± 0.9 b | 19.0 ± 1.9 a | 20.2 ± 1.7 a |
| 68 | 26.6 ± 1.1 a | 21.8 ± 1.2 b | 17.9 ± 0.7 c | 21.4 ± 1.7 a | 22.9 ± 1.9 a |
| Mean | 20.2 | 17.1 | 15.7 | 17.1 | 18.3 |
| LSD (<i>P</i> < 0.05): Tillage practice, 2.9; cropping system, 1.1; interaction, NS | | | | | |
| 0.05-0.15 m | | | | | |
| 0 | 16.8 ± 0.8 a | 15.7 ± 0.9 ab | 15.1 ± 0.6 b | 15.5 ± 0.8 a | 16.2 ± 1.3 a |
| 3 | 17.1 ± 0.7 a | 15.4 ± 0.8 b | 14.7 ± 0.6 b | 15.4 ± 0.9 a | 16.0 ± 1.2 a |
| 7 | 17.0 ± 0.9 a | 15.6 ± 0.7 ab | 14.8 ± 0.8 b | 15.6 ± 0.4 a | 15.9 ± 0.9 a |
| 15 | 16.3 ± 1.0 a | 15.1 ± 0.8 a | 15.0 ± 0.8 a | 15.3 ± 0.8 a | 15.6 ± 1.0 a |
| 30 | 17.7 ± 0.9 a | 16.5 ± 0.8 ab | 15.3 ± 0.9 b | 16.2 ± 1.0 a | 16.8 ± 1.2 a |
| 45 | 20.0 ± 0.7 a | 17.6 ± 0.7 b | 16.3 ± 0.7 b | 17.7 ± 1.6 a | 18.2 ± 1.2 a |
| 68 | 22.1 ± 0.6 a | 19.8 ± 0.8 b | 16.9 ± 0.7 c | 19.5 ± 1.5 a | 19.7 ± 1.2 a |
| Mean | 18.1 | 16.5 | 15.4 | 16.5 | 16.9 |
| LSD (<i>P</i> < 0.05): Tillage practice, 1.9; cropping system, NS; interaction, NS | | | | | |

Table 1: Effect of pendimethalin on population of soil microorganisms at different days of its application under different tillage practices and cropping systems at different soil depths.

* ZT-ZT, zero tillage in both summer and winter seasons; CT-ZT, conventional tillage in summer and zero tillage in winter season; CT-CT, conventional tillage in both the seasons. Means of the values at a given day followed by the same letter do not differ significantly at the 95% confidence level among different tillage practices, and similarly under cropping systems.

the mixing of organic matter in surface 10-12 cm (usually tilled soil layer) soil during tillage operations. The decrease in the bacterial population may be due to competitive influence, and toxic effect as well as different persistence periods of the pendimethalin under different tillage treatments [21]. Higher per cent reduction in the population of microorganisms under ZT-ZT may also be attributed to the relatively higher initial population as compared to the population observed under the other two tillage practices. Under all the tillage treatments, a short time initial depressive effect followed by an increase in the population of microorganisms to initial or even exceeding (Table 1) may be due to the adverse impact of the herbicide on susceptible strains and the subsequent increase in the growth rate of relatively resistant strains during the crop season. The variation in reduction of the population of the microorganisms under different tillage practices indicated that bacteria were very sensitive to pendimethalin while actinomycetes and fungi showed moderate tolerance. The subsequent increase in counts of microorganisms could also be due to the increase in nutrients that come

from weeds killed by the herbicide and utilization of herbicides as a source of nutrients and energy by the resistant strains. Different soil microorganisms have a different degree of sensitivity to the herbicide molecule at different exposure periods. [24] found bacterial population to reduce sharply at the 5th day of the application of pendimethalin (sensitive), sustain up to 15 days and then increased significantly as compared to the initial population. They also reported that pendimethalin moderately toxic to fungal colony development as the colonies showed their ability to recover from the initial inhibition of 54% by 10th day of the application and at 15 days, a full colony recovery was observed. Similarly, actinomycetes' population was also recorded to be reduced but after 15 days of application, the population increased significantly than the population under control. Among soil microbes, Trimurtulu., *et al.* (2015), however, reported a much slower rate of increase in bacterial population after the application of pendimethalin. The initial reduction of soil microorganisms after pendimethalin application into soil and subsequent stimulation of their population was also reported in other studies [14,25-28].

| Soil micro-organisms | Tillage practice* | | | | | | Cropping system | | | |
|----------------------|-------------------|------|-------|------|-------|------|-----------------|------|---------------|------|
| | ZT-ZT | | CT-ZT | | CT-CT | | Mungbean-wheat | | Sorghum-wheat | |
| | A | H | A | H | A | H | A | H | A | H |
| Bacteria | - 47 | + 21 | - 41 | + 15 | - 33 | + 13 | - 41 | + 18 | - 42 | + 17 |
| Fungi | - 12 | + 15 | - 08 | + 09 | - 07 | + 10 | - 08 | + 20 | - 11 | + 09 |
| Actinomycetes | - 17 | + 34 | - 13 | + 33 | - 12 | + 14 | - 14 | + 26 | - 13 | + 31 |

Table 2: Per cent reduction (-) or increase (+) in the population of soil microorganisms at 7th day after the application of pendimethalin (A) and at harvest (H) under different tillage practices and cropping systems in surface 0.05 m soil depth over the initial population.

* ZT-ZT- zero tillage in both summer and winter, CT-ZT- conventional in summer and zero in winter, CT-CT- conventional tillage in both the seasons.

The pattern of increase in the population of microorganisms upon application of the herbicide observed during the crop season indicated that soil microorganisms had the capacity to recover quickly from the initial set-back and exceeding to their initial population in nearly 15 days. The indirect effect of enhanced release of root exudates in the rhizosphere upon herbicide application may have also caused increase in microbial population. After recovering, the actinomycetes kept growing at a relatively faster rate till harvest of crops under all the tillage practices. As a result, under

the CA system (ZT-ZT) at harvest, a maximum increase in population of 34% was observed in actinomycetes followed by bacteria (21%) and least in fungi (15%) over their respective initial population. The exceptionally high augmentation of actinomycetes at harvest could be connected with their ability to better utilize the complex substrates like pendimethalin. Such increase in actinomycetes' population by pendimethalin was also reported by Kocarek., *et al.* (2016) while Trimurtulu., *et al.* (2015) observed the increase in actinomycetes' population in the rhizosphere in a logarithmic

fashion. The rate of such increase in population of microorganisms largely decreased with increase in intensity of tillage in cropping systems, *i.e.*, higher under CT-CT than under CT-ZT. At harvest of the crops, it was observed that the CA system helped increasing the population of bacteria, fungi and actinomycetes by 44.5, 37.7 and 32.7%, respectively, over to that under CT-CT practice in surface 0-0.05 m soil depth. The CA system induces strong changes in soil physico-chemical properties and biological activity, and particularly, the increased organic matter content at the soil surface and its gradual decrease with depth may lead to increased herbicide retention decreasing its availability for biological degradation [21]. The antagonism between retention and degradation escorts higher persistence of herbicide in soils under CA and as a consequence different soil microorganisms may respond differentially as compared to that under conventional systems.

In 0.05-0.15 m soil depth, the population of soil microorganisms started growing largely after 30 days of application of the herbicide when crops were in their growth stages. At harvest, the population of soil microorganisms was significantly higher under ZT-ZT as compared to CT-CT. Under ZT-ZT, the increase in the population of actinomycetes was highest (32%) followed by bacteria (24%) and fungi (14%) over their respective initial population. The corresponding increase under CT-CT practice was 12, 11 and 6%. At this depth, the ZT-ZT practice increased the population of bacteria, fungi and actinomycetes by 37, 21 and 31% as compared to that observed under CT-CT practice. Not much change in the population of soil microorganisms was observed in lower 0.15-0.30 m soil depth under different tillage treatments during the crop season, therefore, data do not show in table 1.

The stabilization of the population of bacteria, fungi and actinomycetes under different tillage practices towards the end of the season as observed at the time of sowing indicated that the ecosystem has achieved a greater degree of equilibrium under the CA system after 13 years of experimentation.

Impact of cropping systems on soil microorganisms

The pattern of the population of soil microorganisms observed at different depths during the season under two cropping systems after the application of pendimethalin remained largely similar to that observed under different tillage practices (Table 1). The

differences in the counts of microorganisms under the two cropping system at different depths during the crop season were non-significant may be due to dominating influence of tillage over the cropping systems. Largely the population remained, numerically, higher under the sorghum-wheat cropping system, which may be attributed to higher biomass production under the sorghum (C4 plant) crop over mungbean in the cropping systems. The interactive effect of tillage practices and cropping systems, however found to be non-significant on the population of soil microorganisms.

Conclusions

- The long-term adoption of CA system maintained a significantly higher population of the soil microorganisms at different depths throughout the crop season as compared to the conventional system.
- The variation in the reduction of population of microorganisms upon the application of pendimethalin under different tillage practices showed bacteria sensitive, and fungi and actinomycetes moderately sensitive to the herbicide at the recommended dose but the growth of actinomycetes remained quite faster.
- At crop harvest, the increase in population of microorganisms decreased with the increase in the intensity of tillage in the cropping systems. The CA system enhanced the population of bacteria, fungi and actinomycetes by 44.5, 46.2 and 33%, respectively, over CT-CT practice.
- Tillage had the significant influence as compared to the cropping system in the CA system on the population of soil microorganisms.
- A reversible change in the equilibrium of the population of microorganisms largely occurred within 15 days of the application of pendimethalin.
- The CA system provided greater resilience to soil microorganisms from perturbation caused by the application of pendimethalin over the extensively tillage based conventional system in practice.

Declaration on Conflict of Interest

The authors declare that they have no conflict of interest.

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