Effect of Nanoencapsulated Pre-emergence Sulfentrazone Herbicide on Soil Microbiome and Nodulation of Irrigated Blackgram (Vigna mungo L.)

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Abstract

Field experiments were conducted in the wetland farms of Department of Agronomy, Tamil Nadu Agricultural University, Coimbatore during Rabi and Summer 2019-2020. Both the experimental trials consists of nine treatments of randomized block design which were replicated thrice. The treatments comprise of sulfentrazone with and without encapsulation @ 0.30 kg ha⁻¹ applied at 1 DBS and 2 DAS followed by general recommended herbicides and weed management methods for blackgram. Sulfentrazone herbicide was encapsulated by using solvent evaporation method for season long weed management and to reduce the leachability. These treatments were tested to know their effect on soil bacterial, fungal and actinomycetes population and also the nodulation ability of blackgram crop. All the herbicide applied treatments were showed reduction in bacterial, fungal and actinomycetes population at 25 DAS compared to initial population, but slight increase in the population of T₇ (Two hand weedicings at 15 and 30 DAS), T₈ (Weed free check)and T₉ (Absolute control)treatments in both the trials. At 50 DAS there was great increase in microbial population compared to 25 DAS in all herbicide applied treatments. There was no significant difference among all the treatments at 50 DAS in microbial population. Higher nodule count and nodule dryweight were noticed at 30 DAS in T₉ (Absolute control), T₈ (Weed free check) and T₇ (Two hand weedicings at 15 and 30 DAS) which is followed by T₁ (Encapsulated sulfentrazone @ 0.30 kg a.i. ha⁻¹ applied at 1 DBS) and T₆ (Pendimethalin @ 1kg a.i. ha⁻¹ applied at 2 DAS fb hand weeding at 20 DAS). But at 60 DAS there was no significant difference among the treatments except with unweeded control.

Keywords
Leachability, Nodulation, Solvent evaporation

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Introduction

Soil is the restless harbour for plant growth and is the mother land for most of the microbes. Use of pesticides and fertilizers in crop protection and production affects soil in many ways. Their concentration, threshold level, half-life, movement and also type of crop that harbours the particular soil influence soil biology and ecology. Blackgram is a nutritious edible seed of leguminous crop, have become an essential part of the human diet. It is an important pulse crop cultivated in tropical and subtropical regions of the world.
These leguminous plants are symbiotically connected with rhizobia and their interaction plays a vital role in crop growth (Vijay et al., 2018). Symbiotic nitrogen fixing ability of this crop helps in enrichment the soil, with this reason blackgram became an important crop for crop rotation. N-fixing bacteria and fungi are accountable up to 80% of nitrogen and up to 75% of phosphorus, that is assimilated by plants annually (Nongmaithem and Pal, 2013). Microorganisms are influenced by several factors including the application of herbicides (Pampulha et al., 2007). Among the different soil microbes, more sensitive microbes to herbicides are bacteria (Ghinea et al., 1998). Sulfentrazone herbicide belongs to the family of phenyl triazolinone, has mean partition coefficient $K_{oc} = 43$ and sorption coefficient $K_d < 1$ and also has high horizontal and vertical leaching potential (Martinez et al., 2008). It has high Groundwater Ubiquity Score (GUS) of 6.75 which is far more than broad spectrum herbicides like pendimethalin and glyphosate which are having GUS of 0.66 and 0.42 respectively (Gustafson, 1989). This is the prime reason for encapsulation of sulfentrazone using solvent evaporation method. This study is mainly aimed to know the effect of sulfentrazone with and without encapsulation and other different treatments on soil microbial population changes with time and also to know nodulation ability of blackgram.

**Materials and Methods**

Field experiments were conducted in the wetland farms of Department of Agronomy, TNAU, Coimbatore during Rabi and Summer 2019-2020. Both the experimental trials consists of nine treatments of randomized block design which were replicated thrice. The treatments are $T_1$-Encapsulated($e^+$) Sulfentrazone @ 0.3 kg a.i. ha$^{-1}$ at 1 DBS, $T_2$-Non-encapsulated($e^-$) Sulfentrazone @ 0.3 kg a.i. ha$^{-1}$ e$^{-}$ at 1 DBS, $T_3$-Sulfentrazone @ 0.3 kg a.i. ha$^{-1}$ e$^{+}$ at 2 DAS, $T_4$-Sulfentrazone @ 0.3 kg a.i. ha$^{-1}$ e$^{-}$ at 2 DAS, $T_5$-Pendimethalin @ 1.0 kg a.i. ha$^{-1}$ at 2 DAS fb Quinalofop-ethyl @ 50 g a.i. ha$^{3}$ and Imazethapyr @ 50 g a.i. ha$^{-1}$ at 20 DAS, $T_6$-Pendimethalin @ 1.0 kg a.i. ha$^{-1}$ at 2 DAS fb 1 HW at 20 DAS, $T_7$-HW twice at 15 and 30 DAS, $T_8$-Weed free check and $T_9$-Absolute control.

The soil type of the field trials is clay loam in texture, slightly basic pH (8.4), low EC (0.43 dSm$^{-1}$), medium in organic carbon (0.70 per cent), low in available N (263.5 kg ha$^{-1}$), medium in available $P_2O_5$ (15.2 kg ha$^{-1}$) and high in available K (891.7 kg ha$^{-1}$). Proper need based crop management practices and plant protection measures were followed in all the treatments as per the crop production guide, TNAU, 2019. Microbial population dynamics in various treatments was studied from the experimental soil before sowing, at 25 and 50 DAS by serial dilution plate count technique. Weighed and transferred 1 gram of soil in to 10 ml sterile distilled water and shaked rigorously. This gives 10$^{-1}$ dilution, from this 1 ml of suspension was transferred to 9 ml of sterile distilled water using a sterile pipette to get 10$^{-2}$ dilution. Consequent 10$^{-3}$, 10$^{-4}$ 10$^{-5}$ and 10$^{-6}$ dilutions were made similarly. The appropriate media viz., nutrient agar, rose bengalagar and kenknightagar for bacteria, fungi and actinomycetes respectively were melted, cooled and poured in to sterile petri plates by pour plate method carrying respective dilution. Petri plates were incubated at 30°C, 2 days, 4 days and 7 days for bacteria, fungi and actinomycetes respectively. After incubation time, emerged colonies were counted and expressed as CFU per gram of soil. For nodule count and dryweight five plants were selected and pulled out after giving irrigation then counted No. of nodules per plant. After that nodules were collected, shade dried and taken dry weight per plant in mg plant$^{-1}$ at 30 and 60 DAS.
**Results and Discussion**

**Effect on soil microbiome**

All the herbicide applied treatments were showed reduction in bacterial, fungal and actinomycetes population at 25 DAS compared to initial population, but slight increase in the population of T7, T8 and T0 treatments in both the trials. At 50 DAS there was great increase in microbial population compared to 25 DAS in all herbicide applied treatments. There was no significant difference among all the treatments at 50 DAS in microbial population. This might be due to carbon released from degraded herbicide leads to an increase of the soil microflora population (Bera and Ghosh, 2013). In sulfentrazone applied plots initially at 25 DAS there was less bacterial, fungal and actinomycetes population compared to control. But at 50 DAS there was gradual increase in population of microbes (Table 1 and Table 2). This was supported by Sulfentrazone applied to sugarcane crop at lower doses of 720 and 840 g a.i. ha⁻¹ did not affect the microflora but in case of higher doses of 1320 and 2400 g a.i. ha⁻¹ initial reduction of microflora was observed and recovered 30 days after application (Kalaiyarasi, 2012).

**Effect on nodulation of blackgram**

Higher nodule count and nodule dryweight were noticed at 30 DAS in T9 (Absolute control), T8 (Weed free check) and T7 (Two hand weedicings at 15 and 30 DAS) which is followed by T1 (Encapsulated sulfentrazone @ 0.30 kg a.i. ha⁻¹applied at 1 DBS) and T6 (Pendimethalin @ 1kg a.i. ha⁻¹applied at 2 DAS fb hand weeding at 20 DAS). But at 60 DAS there was no significant difference among the treatments except with unweeded control. In this experiment it is concluded that in sulfentrazone @ 0.30 kg a.i. ha⁻¹with and without encapsulation and also in other herbicidal treatments even though there was slight decrease in microbial population, nodule count and nodule dryweight at initial stages of blackgram, later due to herbicidal degradation by microbes there was gradual increase in soil microbiome and nodulation ability.
| T. No. | Treatments                                                                 | 25 DAS         | 50 DAS         |
|-------|----------------------------------------------------------------------------|----------------|----------------|
|       |                                                                            | Bacteria (x 10^6 CFU) | Fungi (x 10^4 CFU) | Actinomycetes (x 10^3 CFU) | Bacteria (x 10^6 CFU) | Fungi (x 10^4 CFU) | Actinomycetes (x 10^3 CFU) |
| T_1  | Sulfentrazone @ 0.3 kg a.i. ha^-1 e@ at 1 DBS                              | 30.96          | 8.33           | 18.18               | 45.99             | 14.72            | 24.98                        |
| T_2  | Sulfentrazone @ 0.3 kg a.i. ha^-1 e@ at 1 DBS                              | 30.31          | 8.17           | 17.86               | 45.25             | 14.70            | 24.46                        |
| T_3  | Sulfentrazone @ 0.3 kg a.i. ha^-1 e@ at 2 DAS                              | 30.95          | 7.58           | 17.58               | 45.35             | 14.59            | 24.27                        |
| T_4  | Sulfentrazone @ 0.3 kg a.i. ha^-1 e@ at 2 DAS                              | 30.92          | 7.53           | 17.51               | 45.24             | 14.51            | 24.29                        |
| T_5  | Pendimethalin @ 1.0 kg a.i. ha^-1 at 2 DAS fb Quizalofop-ethyl @ 50 g a.i. ha^-1 and Imazethapyr @ 50 g a.i. ha^-1 at 20 DAS | 29.75          | 7.79           | 17.05               | 47.51             | 14.86            | 25.47                        |
| T_6  | Pendimethalin @ 1.0 kg a.i. ha^-1 at 2 DAS fb 1 HW at 20 DAS                | 33.85          | 9.25           | 18.37               | 48.41             | 15.39            | 26.47                        |
| T_7  | HW twice at 15 and 30 DAS                                                 | 43.55          | 12.25          | 22.67               | 48.50             | 15.79            | 27.20                        |
| T_8  | Weed free check                                                            | 44.51          | 12.08          | 22.74               | 48.45             | 16.39            | 26.90                        |
| T_9  | Absolute control                                                           | 46.90          | 11.67          | 23.48               | 48.34             | 15.49            | 27.30                        |
| SEd  |                                                                            | 2.31           | 0.84           | 0.83                | 2.74              | 0.79             | 1.51                         |
| CD(P=0.05) |                                                                            | 4.89           | 1.79           | 1.76                | NS               | NS              | NS                           |

*e - with encapsulation  e- without encapsulation  DBS – Day before sowing  DAS – Days after sowing  HW – Hand weeding

*Initial microbial population (Before ploughing): Bacteria – 41.55 x 10^6 CFU  Fungi – 8.50 x 10^4 CFU  Actinomycetes – 15.37 x 10^3 CFU
Table 2: Effect of weed management treatments on microbial population (CFU) of soil in trial II

| T. No. | Treatments                                                                 | 25 DAS                  | 50 DAS                  |
|--------|----------------------------------------------------------------------------|-------------------------|-------------------------|
|        |                                                                            | Bacteria (x 10^6 CFU)   | Fungi (x 10^4 CFU)      | Actinomycetes (x 10^3 CFU) | Bacteria (x 10^6 CFU) | Fungi (x 10^4 CFU) | Actinomycetes (x 10^3 CFU) |
| T1     | Sulfentrazone @ 0.3 kg a.i. ha^-1 e^+ at 1 DBS                             | 27.57                   | 7.33                    | 15.40                  | 42.17               | 14.42               | 21.42                   |
| T2     | Sulfentrazone @ 0.3 kg a.i. ha^-1 e^- at 1 DBS                             | 26.89                   | 7.33                    | 15.04                  | 41.48               | 14.10               | 21.02                   |
| T3     | Sulfentrazone @ 0.3 kg a.i. ha^-1 e^+ at 2 DAS                             | 27.62                   | 6.33                    | 14.58                  | 41.58               | 14.36               | 21.05                   |
| T4     | Sulfentrazone @ 0.3 kg a.i. ha^-1 e^- at 2 DAS                             | 27.54                   | 6.67                    | 14.06                  | 41.19               | 14.08               | 20.55                   |
| T5     | Pendimethalin @ 1.0 kg a.i. ha^-1 at 2 DAS fb Quizalofop-ethyl @ 50 g a.i. ha^-1 and Imazethapyr @ 50 g a.i. ha^-1 at 20 DAS | 26.56                   | 6.00                    | 14.15                  | 43.50               | 14.28               | 21.84                   |
| T6     | Pendimethalin @ 1.0 kg a.i. ha^-1 at 2 DAS fb 1 HW at 20 DAS               | 31.35                   | 7.33                    | 15.60                  | 43.77               | 14.92               | 22.66                   |
| T7     | HW twice at 15 and 30 DAS                                                 | 38.79                   | 10.33                   | 20.70                  | 44.08               | 15.33               | 23.28                   |
| T8     | Weed free check                                                           | 39.63                   | 11.33                   | 20.47                  | 44.03               | 16.17               | 22.92                   |
| T9     | Absolute control                                                          | 41.84                   | 9.33                    | 21.29                  | 44.35               | 15.00               | 23.43                   |
| SEd    |                                                                            | 1.94                    | 0.60                    | 0.86                   | 2.35                | 0.72                | 1.23                    |
| CD(P= 0.05) |                                                               | 4.11                    | 1.27                    | 1.83                   | NS                  | NS                  | NS                      |

e^+ - with encapsulation  e^- - without encapsulation  DBS – Day before sowing  DAS – Days after sowing  HW - Hand weeding

*Initial microbial population (Before ploughing): Bacteria – 35.50 x 10^6 CFU  Fungi – 7.33 x 10^4 CFU  Actinomycetes – 15.55 x 10^3 CFU
Table 3 Effect of weed management treatments on nodule count (No. plant\(^{-1}\)) and nodule dryweight (mg plant\(^{-1}\)) of blackgram in trial I and II

| T. No. | Treatments                                                                 | Trial I |          |          | Trial II |          |          |
|--------|-----------------------------------------------------------------------------|---------|----------|----------|----------|----------|----------|
|        |                                                                             |         | 30 DAS   | 60 DAS   | 30 DAS   | 60 DAS   |          |
|        |                                                                             | Count   | Dry weight | Count   | Dry weight | Count   | Dry weight |
| T1     | Sulfentrazone @ 0.3 kg a.i. ha\(^{-1}\) e\(^+\) at 1 DBS                   | 26.33   | 84.88     | 40.67   | 179.29    | 28.85   | 95.35     | 43.86 | 194.80   |
| T2     | Sulfentrazone @ 0.3 kg a.i. ha\(^{-1}\) e\(^-\) at 1 DBS                   | 18.67   | 77.31     | 39.67   | 179.07    | 20.57   | 84.11     | 42.40 | 194.59   |
| T3     | Sulfentrazone @ 0.3 kg a.i. ha\(^{-1}\) e\(^+\) at 2 DAS                   | 22.33   | 80.47     | 39.00   | 178.39    | 24.68   | 89.64     | 42.02 | 193.78   |
| T4     | Sulfentrazone @ 0.3 kg a.i. ha\(^{-1}\) e\(^-\) at 2 DAS                   | 17.33   | 74.53     | 38.33   | 177.98    | 19.03   | 80.70     | 41.29 | 193.43   |
| T5     | Pendimethalin @ 1.0 kg a.i. ha\(^{-1}\) at 2 DAS fb Quinalofop-ethyl @ 50 g a.i. ha\(^{-1}\) and Imazethapyr @ 50 g a.i. ha\(^{-1}\) at 20 DAS  | 16.67   | 76.42     | 41.67   | 180.16    | 18.33   | 84.40     | 43.78 | 195.75   |
| T6     | Pendimethalin @ 1.0 kg a.i. ha\(^{-1}\) at 2 DAS fb 1 HW at 20 DAS        | 27.67   | 82.28     | 41.67   | 179.94    | 30.28   | 92.11     | 44.32 | 195.66   |
| T7     | HW twice at 15 and 30 DAS                                                 | 33.33   | 98.08     | 42.33   | 180.81    | 39.02   | 103.15    | 45.70 | 202.55   |
| T8     | Weed free check                                                           | 34.33   | 99.21     | 42.33   | 182.13    | 37.75   | 108.26    | 45.95 | 202.04   |
| T9     | Absolute control                                                          | 41.33   | 84.89     | 36.67   | 101.54    | 45.43   | 94.43     | 37.25 | 114.50   |
|        | SEd                                                                         | 2.22    | 7.42      | 2.16    | 17.97     | 2.52    | 7.74      | 2.27  | 17.91    |
|        | CD(P= 0.05)                                                                | 4.70    | 15.74     | 4.58    | 38.10     | 5.34    | 16.40     | 4.82  | 37.97    |

* e\(^+\) - with encapsulation  e\(^-\) - without encapsulation  DBS – Day before sowing  DAS – Days after sowing  HW - Hand weeding
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