### Full Length Research Paper

# Effect of herbicide (pendimethalin) on soil microbial population

Oyeleke, S. B.\*, Oyewole, O. A. and Dagunduro, J. N.

Department of Microbiology, Federal University of Technology, Minna, Nigeria.

Accepted 1 November, 2011

A field study on the effect of herbicide (pendimethalin) on soil microbial community was conducted between August to September (2010) on a sandy-loamy soil located at the Federal University of Technology, Crop Science Departmental Farm Gidan-Kwano, Minna, Nigeria. Samples were collected randomly from the experimental farm before and after application of herbicide; on which cowpea was cultivated. Herbicide (Pendimethalin) was applied at below recommended rate, recommended rate and above recommended rate. The result reveals the following herbicide treatments at the recommended rates resulted in decrease in microbial count. High concentration of herbicide treatment resulted in a much lower microbial count compared to soil treated rate, recommended herbicides and none treated soil. The most frequently isolated bacteria from herbicide treated soil are Bacillus and Pseudomonas sp., while Aspergillus, Rhizopus and Penicillium sp. were the most frequently isolated fungi from treated soils. Cowpea yield was highest in site treated with herbicide at above recommended rate and lowest in the control site. The results revealed that herbicide is an integral part of farming, although it has a significant effect on soil microbial community.

Key words: Herbicide (pendimethalin), recommended rate, recommended rate and above recommended rate.

#### INTRODUCTION

Pendimethaline (N - (I ethylpropoly) - 3, 4 dimethyl - 2, 6- benzamine) is an organic herbicide of the dinitroaniline herbicide used for grass weed control in many crops ranging from maize, sorghum, through cowpea soybean to vegetable crops (Akobundu, 1987; Cork and Krueger, 1992). Although pendimethaline provides improvement in plant growth, it can also have side effects and adversely influence soil microbial activities. Since herbicides are formulated to kill organisms, it is not surprising that some of these compounds are toxic to specific soil organisms such as Cyanobacteria sp., Bacillus sp., Pseudomonas sp. for bacteria and fungi e.g. Penicillium, Mucor, Fusarium and Aspergillus species (MacNaughton et al., 1999) and actinomycetes affecting various soil microbial processes such as nitrification, nitrogen fixation, nitrogen metabolism, respiration and organic matter decomposition (White et al., 1998). The aim and objective of this research is to examine the effects of pendimenthalin on soil microbial community.

#### **MATERIALS AND METHODS**

#### Collection of sample

The farm was sited at the Federal University of Technology, Minna, Crop science farm at Gidan Kwano campus, permanent site, the size of the farm was 26 by 15 m. The experiment was conducted in a split-split plot arrangement of treatments with three replications in a randomized complete block. The replications were spaced in 1 m apart with each replication measuring 4 m. The sites were designated  $T_1$  to  $T_4$ .  $T_1$  is the control plot with no herbicides application, T is the below recommended rate application, T<sub>3</sub> is the site with the recommended rate application, while T4 is the site with the above recommended rate, application. Each sub-plot consisted of five rows, 4 m long and spaced 25 cm apart at the seed rate of 25 kg/ha. Soil samples were collected from the top 10 cm depth by taking 4 random scores (7.5 cm diameter) using a sterile auger. Bulk soil sample were homogenized by passing through a 2 mm sieve. Soil samples were collected for a period of 8 weeks at 2 weeks interval from July to September 2010.

#### Herbicide treatment of soil

Plants in the entire experimental area were cleared manually before ridges were made. Herbicides treatment included pre-only. The herbicide used was pendimenthalin. It was purchased from a local

<sup>\*</sup>Corresponding author. E-mail: droyeleke@yahoo.com.

**Table 1.** Mean Microbial counts of control and pendimenthalin treated soils.

Soil treatment	AHB	Fungi
T <sub>1</sub>	$5.23 \times 10^6$	$11.37 \times 10^4$
$T_2$	$4.02 \times 10^6$	$7.9 \times 10^4$
T <sub>3</sub>	$3.85 \times 10^6$	$7.52 \times 10^4$
T <sub>4</sub>	$3.23 \times 10^6$	$6.80 \times 10^4$

 $T_{1^-}$  Control,  $T_{2^-}$  0.5 kg/L (Below recommended rate);  $T_{3^-}$  1.5 kg/L (Recommended rate);  $T_{4^-}$  2.5 kg/L (Above recommended rate).

**Table 2.** Bacteria Isolated from control and pendimenthalin treated soils.

S/N	Bacteria	Fungi
1	Pseudomonas sp.	Mucor sp.
2	Bacillus sp.	Aspergilus niger
3	Bacillus subtilis	Rhizopus sp.
4	Micrococcus sp.	Aspergillus flavus
5	Acinetobacter sp.	Geotrichum candidum
6	Actinomycetes	Fusarium sp.
7	Rhizobium japonicum	Penicullum italicum
8		Candida albicans
9		Trichoderma sp.

**Table 3.** Bacterial counts  $(\times 10^6)$  of control and pendimenthalin treated soil.

T1	T2	Т3	T4
3.67	3.47	3.57	3.70
6.23	4.40	4.20	4.03
6.20	4.57	4.50	4.20
6.50	4.37	4.10	4.70
5.43	4.10	3.70	2.27
5.03	3.90	3.73	2.50
4.60	3.87	3.67	2.27
4.23	3.57	3.40	2.10
5.23	4.02*	3.85*	3.23*
	3.67 6.23 6.20 6.50 5.43 5.03 4.60 4.23	3.67 3.47 6.23 4.40 6.20 4.57 6.50 4.37 5.43 4.10 5.03 3.90 4.60 3.87 4.23 3.57	3.67 3.47 3.57   6.23 4.40 4.20   6.20 4.57 4.50   6.50 4.37 4.10   5.43 4.10 3.70   5.03 3.90 3.73   4.60 3.87 3.67   4.23 3.57 3.40

<sup>\*</sup> Significant at p<0.05; \*\* insignificant.

agricultural dealer store in Minna. Dosage application for cowpea was 1.5 ml per litre according to the recommended rate. Method of application is by pre-emergence surface spray within 2 days sowing. The herbicides was applied at 0.5 kg/L (below recommended rate) on  $T_2$  plot,  $T_3$  at 1.5 kg/L (at recommended rate) and  $T_4$  at 2.5 kg/L using blue nozzle sprayer as recommended by the manufacturer, while no herbicide was applied to the control plot  $(T_1)$ .

#### Source of cowpea

Cowpea was obtained from Niger State Agricultural Development

Agency and were sowed in 3 m wide rows seeding rate. The variety sowed was BOSA High yield.

#### Enumeration and isolation of microorganisms

Ten grammes (10 g) of each soil sample were suspended in 9 ml of sterile water and serially diluted. The diluted samples were inoculated on nutrient agar and yeast extract agar for the enumeration of total aerobic bacteria and Sabouraud dextrose agar (SDA) amended with 0.2 g chloramphenicol for the enumeration of fundi.

The nutrient agar and yeast extract agar were incubated at  $30^{\circ}\!\!\mathrm{C}$  for 48 h while the SDA was incubated at room temperature (28  $\pm$   $2^{\circ}\!\!\mathrm{C}$ ) for 72 h. Colonies which developed on the plate's were counted and recorded as colony forming units per gramme of soil (cfu/g). The isolates were sub-cultured repeatedly to obtain pure samples, which were maintained on agar slants for further characterization and identification.

#### Characterization and identification of microbial isolates

The characterization and identification of bacterial isolates were based on cell morphology and biochemical tests as described by Cheesbrough (2003), and Oyeleke and Manga (2008). Fungi isolates were identified by microscopic and macroscopic techniques as described by Cowan and Steel (1974).

#### **RESULTS**

### Microbial counts of control and pendimethalin treated soils

Table 1 shows the aerobic heterotrophic bacteria (AHB) and fungi count in the control and treated soils. The counts of AHB in soil ranged from  $6.5 \times 10^4$  to  $2.1 \times 10$  cfu/g. The untreated soil (T1) had the highest counts of AHB and fungi.

### Bacteria isolated from control and pendimethalin treated soils

Table 2 shows the morphology and biochemical characteristics of bacteria isolated from the control and treated soils. *Bacillus, Micrococcus, Pseudomonas* spp. were identified. *Bacillus* sp. was more frequently isolated than *Pseudomonas or Micrococcus*. Fungi isolated and identified include *Aspergillus, Rhizopus, Fusarium, Mucor, Penicillium* and *Candida* spp. (Table 2).

## Bacterial counts of control and pendimethalin treated soil

Table 3 shows the counts of aerobic heterotrophic bacteria (AHB) in control and pendimethalin treated soil over a period of eight weeks. The control soil had higher counts of AHB than those of soil treated with pendimethalin.

**Table 4.** Fungal counts (x10<sup>4</sup>) of control and pendimenthalin treated soil.

Weeks	T1	T2	Т3	T4
1.	10.70	8.10	8.23	6.23
2.	11.30	8.43	8.27	9.77
3.	13.10	8.50	8.67	9.40
4.	13.50	8.60	8.70	9.17
5.	12.17	7.83	7.10	6.43
6.	10.30	7.53	6.83	5.20
7.	10.20	7.10	6.37	4.23
8	9.70	7.10	6.03	4.03
Mean	11.37	7.9*	7.52*	6.80*

<sup>\*</sup> Significant at p<0.05; \*\* insignificant.

### Fungal counts of control and pendimethalin treated soil

Table 4 shows the fungal counts in control and herbicides treated soil. Total fungal populations were highest in control and lowest in soil treated with pendimethalin at above recommended rate.

#### **DISCUSSION**

The results of the research revealed an appreciable difference in the soil microbial community during the period of eight weeks. An initial general rise in bacterial counts was observed in all the soil treated with the herbicide, reaching a maximum between weeks 3 and 4 (Table 3). This agrees with the study of Kunc et al. (1985) who worked on 'Mineralization and changes in the count of bacterial decomposers' and observed an initial rise in microbial count in the treated soils. This could be due to the fact that the soil microflora are able to temporarily mineralize and use the herbicide as energy source (Kunc et al., 1985). While there was an initial increase in the bacterial population from site T4 with above recommended rate treatment immediately after application of the herbicide, a decrease in count was observed by the 5<sup>th</sup> week. This can be appreciated to the microorganism, initially metabolizing the herbicide according to Munier et al. (2002). This led to increase in their number and subsequent reduction when the herbicide disappeared from the soil. Microbial count of sites T2 and T3 were relatively the same throughout the course of the 8 weeks. The bacterial count for T2 at one week of treatment was  $3.47 \times 10^6$  cfu/g before treatment and remained between  $3.57 \times 10^6$  to  $4.57 \times 10^6$  cfu/g. Site T3 at one week of treatment was  $3.57 \times 10^6$  cfu/g and remained between 3.4  $\times$  10<sup>6</sup> to 4.5  $\times$  10<sup>6</sup> cfu/g by the end of the research. Herbicide treatments at recommended rate resulted in lower fungal counts compared to the below recommended rate and control soil.

The mean result obtained after a week of treatment for fungal counts at control and below recommended rate were  $10.7 \times 10^4$  and  $8.1 \times 10^4$  cfu/g respectively. While herbicide treatment at 1.5 recommended rate was  $6.25 \times 10^4$  cfu/g at one week of treatment. By week 4 of herbicide treatment, mean fungal counts at all the sites reached a peak. Control site show counts of  $1.4 \times 10^5$  cfu/g, T2 at  $8.6 \times 10^4$  cfu/g, T3 at  $8.70 \times 10^4$  cfu/g and T4 at  $9.17 \times 10^4$  cfu/g. The fungal count at the control soil was however higher throughout the course of the 8 weeks. The treated soils had lower microbial counts throughout the period of the research.

This agreed with the study of Alexander (1977), that those pesticides which alter the abundance of particular heterotrophic populations are usually the ones present in high concentration. Domsch and Grams (1983), reported that the general initial rise in microbial counts could be due to the fact that the microorganisms benefits from the transformation of the herbicide, since in the process of decomposition of the complex nitrogen containing molecules, many genera benefits, as the proteinaceous material provides the organism with both nitrogen and carbon. While the general decline in count after the initial rise agreed with the work of Taiwo and Oso (1997), which suggest that this decline in microbial counts after each peak must have been due to the fact that the microbial population that were tolerant of the treated pesticides were susceptible to the products of soil-pesticide interactions which could have possibly been bactericidal or fungicidal. The study has evidently shown that herbicides do have inhibitory effect on soil microbial community, while there was no effect on the yield of the planted crop.

#### REFERENCES

Akobundu IO (1987). Weed science in the Tropics- Principles and practice, Wiley Interscience public, pp. 112-153.

Alexander M (1977). Introduction to soil microbiology, Krieger Publishing Company, pp. 56-67

Cheesbrough M (2003). Laboratory Manuals. District Laboratory Practice in Tropical Countries (2) Cambridge University Press. United Kingdom, pp. 146-157.

 Cork DJ, Krueger JP (1992). Pesticide Biodegradation in Encyclopedia of Microbiology (Lederberg, J-ed), 3: 357-36.
Cowan ST, Steel KJ (1974). Manual for the Identification of Medical

Bacteria. "Cambridge University of Press, Cambridge.

Domsch KH, Gams W (1983). Fungal in Agricultural Soils. Halsted Press (Wiley), New York.

Kunc F, Tichy P, Vancura V (1985). 2-4 dichlorophetexoxy acetic acid in the soil: Mineralization and changes in the counts of bacteria decomposer. Versailles Ed.INRA Publ (Les Colloques de l'NRA No. 31).

MacNaughton SJ, Stephen JR, Venosa AD, Davis GA, Chang YJ, White DC (1999). Microbial population changes during bioremediation of an experimental oil spill. Appl. Environ. Microbiol., 65: 3566-3574

Munier LC, Feuvrier MP, Chone T (2002). Degradation of 14C. Atrazine Bound Residue in Brown Soils and Rendzinz Fractions. J. Environ. Qual.. 31: 241-247

Oyeleke SB, Manga SB (2008). Essentials of Laboratory Practical in Microbiology, pp. 20-33.

Taiwo LB, Oso BA (1997). The Influence of some pesticides on soil microbial flora in relation to changes in nutrient level, rock phosphate solubilization and P-release under laboratory conditions. Agric. Ecosyst. Environ., 65: 59-68.

White DC, Flemming CA, Leung KT, MacNaughton SJ (1998). In situ microbial ecology for quantitative assessment, monitoring and risk assessment of pollution remediation in soils, the subsurface, the rhizosphere and in biofilms. J. Microb. Methods, 32: 93-105.