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# Effect of 2,4-Dichlorophenoxyacetic Acid (2,4-D) and Dichlorvos (2,2-Dichlorovinyl Dimethyl Phosphate DDVP) on Soil Microorganisms

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## Authors' contributions

This work was carried out in collaboration among all authors. Author SOB designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors MBO and OAA managed the analyses of the study. Author EPC managed the literature searches. All authors read and approved the final manuscript.

#### Article Information

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# ABSTRACT

Effects of 2,4-D(2,4-dichlorophenoxy acetic acid) and Dichlorvos(2,2-dichlorovinyl dimenthyl phosphate DDVP) were investigated on soil microbial population in a 4 weeks experiment. Soil samples were serially diluted and inoculated using pour plate method on different medium and incubated at 37°C for 24 and 48 hrs for enumeration of microbial diversity, colonies that appeared on plates were counted. At pre-application of 2,4-D, bacteria had the highest population ( $9.5 \times 10^7$  Cfu), followed by fungi ( $7.5 \times 10^5$  Cfu), actinomycetes ( $1.6 \times 10^6$  Cfu) and protozoa ( $1.1 \times 10^3$  Cfu). Likewise, the pre-application of DDVP represents the population count of microorganisms in the following manner: Bacteria ( $8.0 \times 10^7$  Cfu)>fungi ( $5.1 \times 10^5$  Cfu)>actinomycetes ( $1.0 \times 10^6$  Cfu)>

protozoa (1.0x10<sup>3</sup> Cfu)f The post-application of 2,4-D and DDVP also had the similar pattern of population count. With percentage difference on each of the microbial counts, 2,4-D of bacteria, fungi, actinomycetes and protozoa (99.5%, 95.3%, 99.9% and 86.0% respectively), DDVP percentage difference (99.2%, 99.4%, 99.3% and 98.6% respectively). Application of these pesticides at recommended rates was followed by the general decline in microbial counts. Therefore, the pesticides had toxic effects on microorganisms which may be beneficial to cultivated plants.

Keywords: Pesticides; microbial; application; bacteria; toxic.

## **1. INTRODUCTION**

Pesticides include a large group of chemical agents that attempt to eliminate destructive biological forces in agriculture. These chemicals supposedly only target specific species, repeated use inevitably kills microbial life that is beneficial to the soil system [1-3]. Microbes that survive can be genetically altered in a way that is no longer beneficial to the soil ecosystem and be resistant to the chemical intended to kill them. The destruction or alteration of first-level microbes can affect the entire soil ecosystem all the way up to the largest mammalian predators.

The impact of different pesticides on the growth of soil microorganisms and its activity are difficult to expect. Even if the pesticides used in low concentration they effect chemical and biological properties, biochemical activity and soil microorganisms [4,5,6]. The effect of pesticides on soil microorganisms and their activity depend upon the type of pesticides used, quantities and soil conditions [7].

extensive and inappropriate use of The pesticides for increased crop yields due to their local formulation and easy availability can be a significant source of surface water, groundwater, air and soil contamination [8]. Agricultural sprays may reach the soil through direct application or by falling to the ground when not intercepted by foliage. The rate of exposure is so high that individual populations of microflora may be seriously affected [9]. The influence of pesticides on the microbial community structure is determined by the particular pesticide, its concentration, and its persistence [10-12]. The present study investigates the adverse effect of widely used pesticides on soil microbial populations. This research aim at investigating the effect of 2,4-D and DDV (herbicides and pesticides on soil microorganisms).

Dichlorvos (2,3-dichlorovinyl dimethyl phosphate) is one of the classes of pesticide referred to as

organophosphates used to control households and stored products insects. It is effective against mushroom flies, aphids, spider mites, caterpillars, thrips and white flies in greenhouse, outdoor fruits, and vegetables crops [13].

2,4-D is a growth regulator which has a similar effect to auxin hormone [8]. It belongs to the phenolic compounds family being salts or ester of high molecular weight and low volatility derived fromphenoxyacetic acid [14]. Flaws in the control of certain weed species has led farmers to use herbicide such as 2,4-D, one of the most used in this association especially in pre-planting desiccation application [15]. 2,4-D is a selective herbicide that kills dicots (but not grasses) by mimicking the growth hormone auxin which causes uncontrolled growth and eventually death in susceptible plants.

In soil, 2,4-D is degraded primarily by microbes. Hemmett and Faust [10] concluded that the size of microbial population, the concentration of 2,4-D, and the ratio of two factors determine 2,4-D degradation rates. Soil conditions that enhance microbial population (warm and moist) facilitate 2,4-D degradation [9]. In addition, 2,4-D, presumably because there was an increase in 2,4-D degrading bacteria after the first application [16,17,18].

#### 2. METHODOLOGY

### 2.1 Experimental Study Area

The sampling area was the research farm of Federal College of Forestry, Ibadan, Oyo state. The soil was silty clay loam and alkaline in their chemical reaction.

#### 2.2 Collection of Soil Samples

Soil samples were collected at the depth of 0–15 cm from the Federal College of Forestry, Ibadan, Farm with no history of prior pesticides exposure and were immediately transferred to the

laboratory. Soil samples were sieved while still moist (moisture content 0.88 g) through a 2-mm sieve.

# 2.3 Herbicides and Pesticides

The herbicides Super Amine (2,4-D amine contains 720 g/l Di-methyl amine salt as an aqueous solution while the pesticide Nopest Insecticide contains 1000 gms DDVP.

# 2.4 Experimental Period

The experiment lasted for four weeks for both pre and post application of pesticides.

# 2.5 Determination of Microbial Population before Pesticides Application

Bacterial and fungal populations were determined in each soil sample before application of pesticides and at the end of the 8 weeks. For this purpose, nutrient agar (NA) media were prepared for determination of bacterial population, potato dextrose agar (PDA) media was used for the enumeration of fungal colonies using the dilution plate technique. What media you used for the determination of other microorganisms like actinomycetes and protozoa.

## 2.6 Pesticides Investigated for Microbial Behaviour

One insecticide and one herbicide were selected for this research. The amount of pesticides used for amendments was determined on the basis of recommended dose for agricultural crops. All amendments were made in solution form.

# 2.7 Treatment for Soil Samples

The soil sample treatments (T) used were as follows (pesticides are labeled A and B): T1—soil (control), T2—soil + A at 0.01 mL/500 g soil, T3—soil + B at 0.01 mL/500 g soil.

# 2.8 Media Preparation

Preparation of the media used was as follows:

- Nutrient agar (NA): 20 g nutrient agar was added to 1 L distilled water. This was then boiled to dissolve the agar. The contents were sterilized by autoclaving for 15 min at 121°C.
- Potato dextrose agar (PDA): 200 g potato was boiled to extract the starch. Distilled water was added to 1 L. Then, 20 g dextrose and 20 g agar were added. The solution was sterilized by autoclaving for 15 min at 121°C.

# 3. RESULTS AND DISCUSSION

Table 1 shows that the effect of pesticides treatment on soil population was determined based on growth of Fungi, Bacteria. Actinomycetes and Protozoa counts in each soil treatment. The microbial population growth shows different degrees of sensitivity to the pesticides at recommended rate in both soil treatments (A and B). The bacteria population for all soil samples at pre and post application of 2,4-D had the highest population of  $9.5 \times 10^7$  and 4.3x10<sup>4</sup> (cfu/g) with percentage reduction of 99.5% while soil treated with DDVP at pre and post application had the highest bacteria population of  $8.0 \times 10^5$  and  $6.1 \times 10^3$  (cfu/g) with percentage reduction of 99.2%. The effect of pesticide treated soil on fungi population at pre and post application of 2,4-D was 7.5x10<sup>5</sup> and 3.5x10<sup>4</sup> (cfu/g)with percentage reduction of 95.3% while DDVP at pre and post was 5.1x10<sup>4</sup> and 3.2x10<sup>2</sup> (cfu/g) with percentage reduction of 99.4%. Effect of pesticide treatment on actinomycetes population at pre and post application of 2,4-D was 1.0x10<sup>6</sup> and 1.2x10<sup>3</sup> (cfu/a) with percentage reduction of 99.3% while the effect of 2.4-D on protozoa population for pre and post application was 1.0x10<sup>3</sup> and 1.4x10<sup>2</sup>(cfu/g) with percentage reduction of 86.0% and finally for DDVP,  $1.1 \times 10^4$  and  $1.5 \times 10^2$ (cfu/g) was recorded with percentage reduction of 98.6%.

Table 1. Effect of soil microbial load at pre and post application period of two pesticides

Organism	DDVP (cfu/g)			2,4-D (cfu/g)		
	Pre	Post	% Reduction	Pre	Post	% Reduction
Bacteria	8.0x10⁵	6.1x10 <sup>3</sup>	99.2%	9.5x10 <sup>7</sup>	4.3x10⁵	99.5%
Fungi	5.1x10 <sup>7</sup>	3.2x10 <sup>2</sup>	99.4%	7.5x10⁵	3.5x10⁴	95.3%
Actinomycete	1.6x10⁵	1.2x10 <sup>3</sup>	99.3%	1.0x10 <sup>6</sup>	1.2x10 <sup>3</sup>	99.9%
Protozoa	1.1x10⁴	1.5x10 <sup>2</sup>	98.6%	1.0x10 <sup>3</sup>	1.4x10 <sup>2</sup>	86.0%

Significant reductions in the effect of both pesticides were observed for four weeks after application.

DDVP reduced bacteria, fungi, actinomycete and protozoa by 99.2%, 99.4%, 99.3% and 98.6% respectively while 2, 4- D reduced these organisms by 99.5%, 95.3%, 99.9% and 86.0% respectively.

DDVP significantly reduced the protozoa at 98.6% than 2, 4-D at 86.0%. This can be supported by statement of Taiwo and Oso (1997) that the decline in microbial counts must have been due to the fact that microbial population that were tolerant of treated pesticides were susceptible to the product of the soil pesticide. The decline observation in post application follows the trend of report by Zain et al. (2013) who reported that both bacteria and population actinomvcete were drastically inhibited by paraquat to about 70-82% at recommended rate. Similarly, Stanley et al. (2003) reported that bacteria population for all soil samples dropped at week four (4) of post herbicide treatment.

However, Aranjo et al. (2003) reported the increase in population of actinomycete with some herbicide like glyphosphate. The pesticides application on soil has help in the reduction of microbial population which likely may lead to biomagnifications in soil ecosystem.

# 4. CONCLUSION AND RECOMMENDA-TION

The results in this study could be concluded that pesticides may have toxic effects on microorganisms, reducing their abundance, activities and consequently, the diversity of their communities which could reduce the beneficial activities of the organisms in soil fertility.

It can be recommended that frequency of pesticides application should be minimized to allow the re-growth beneficial soil microbes after application. Also, pesticides application should be carefully maintained at manufactures recommended rate.

# **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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