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Presence of antibiotics in *Panicum maximum* and soil around poultry manure dumping site in derived savannah zone of Nigeria

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Abstract

The presence of antibiotics in *Panicum maximum* and soil around poultry manure dumping site was investigated. The antibiotics investigated were penicillin, streptomycin, tetracycline and chloramphenicol. *P. maximum* and soil samples were collected at distance of 0, 30 and 60 m from the dumping site. Qualitatively, these antibiotics were found positive in all the samples. Decreases in values (quantitatively) were observed for penicillin ($38.80-18.46 \times 10^{-7}$ µg/kg), streptomycin ($20.84-3.86 \times 10^{-7}$ µg/kg) and tetracycline ($31.58-24.47 \times 10^{-7}$ µg/kg) as the distance increased for the *Panicum maximum* around Poultry manure dumping site. Similar decrease in penicillin ($19.52-12.26 \times 10^{-7}$ µg/kg), tetracycline ($20.53-11.19 \times 10^{-7}$ µg/kg) and chloramphenicol ($16.89-3.38 \times 10^{-7}$ µg/kg) as the distance increased in soil around poultry manure dumping site were also observed. The pH of the soil collected was between the ranged of 6.10-6.30. The organic matter ranged from 1.91 to 2.38% and organic carbon was also between 1.11 to 1.38%. The study however confirmed that up to 60 m distance from the dumping site antibiotics (tetracycline, chloramphenicol, penicillin, and streptomycin) were present in both *P. maximum* and soil.

Key words: *Panicum maximum*, soil, poultry manure, antibiotics.

INTRODUCTION

Antibiotics (prophylaxis and therapeutics) help to prevent disease in exposed animals and to treat diseases (Cromwell, 1999). At sub therapeutic level, antibiotics reduce mortality and morbidity. They also improve reproductive performance (Gracey and Collins, 1992).

Antibiotics used in livestock production usually enter the human food chain as residue in the animal products thereby constituting health hazards to the consumer of such animal products. The major health risk associated with residue of antibiotics in human food is the development of drug resistance in such individuals. In addition hypersensitivity reaction and the development of microorganism's resistances to antibiotics in human have been reported (Dupont and Steele, 1987).

Anti-microbial resistance in pathogens from farm animals can be passed on to human through the exchange of genetic material between microorganisms,

thus increasing public health costs through the use of more expensive drugs for treatment (Steinfeld, 2003).

Panicum maximum prefers fertile soil and is well adapted to a variety of conditions. It grows especially well in shaded damp areas under trees and shrubs and is often seen along river banks, but hardly found in open woodland. It is very palatable to livestock and can be cultivated as pasture and also used to make good quality hay.

Antibiotics have been noted to promote growth and increase feed efficiency. Its possible presence in forages around livestock manure dumping sites apart from being high in nutrient might have effect on animals grazing those areas. The study therefore aimed to evaluate the presence of antibiotics in *P. maximum* and soil around poultry manure dumping sites.

MATERIALS AND METHODS

Study areas

A commercial poultry farm situated in Ladoke Akintola University of

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Table 1. The presence of antibiotics in soil around poultry manure dumping site.

Antibiotics × 10 ⁻⁷ µg/kg	0 m	30 m	60 m	SEM
Penicillin	19.52 ^a	14.99 ^d	12.26 ^c	1.05
Streptomycin	13.39 ^a	8.93 ^c	10.13 ^b	1.61
Tetracycline	20.53 ^a	16.39 ^d	11.19 ^c	1.07
Chloramphenicol	16.89 ^c	12.23 ^d	3.38 ^a	1.43

^{abc} Mean values in the same row are significantly different (p<0.05).

Table 2. The presence of antibiotics in *Panicum maximum* around poultry manure dumping site.

Antibiotics × 10 ⁻⁷ µg/kg	0 m	30 m	60 m	SEM
Penicillin	38.80 ^a	25.14 ^b	18.46 ^c	3.42
Streptomycin	20.84 ^a	9.79 ^b	3.86 ^c	2.44
Tetracycline	31.58 ^a	25.80 ^b	24.47 ^b	1.98
Chloramphenicol	21.40 ^b	25.48 ^a	23.33 ^{ab}	2.17

^{abc} Mean values in the same row are significantly different (p<0.05).

Technology, Ogbomoso was considered for this study. It has over 2000 laying birds supplying table eggs to Ogbomoso environs. Ogbomoso is located in the derived savanna zone of Nigeria. It lies at approximately 8° 7' North of the equator and 4°15' East of Greenwich Meridian. The climate is characterized by a fairly high uniform temperature (33 to 21°C), moderate to heavy seasonal rainfall of 1247 mm annually and high relative humidity (Oguntoyinbo, 1978). The permanent dumping site of this farm was considered and soil samples and forages were collected. The dumping of poultry manure has been on yearly basis for the past fifteen years.

Sample collection

Samples of soil and *P. maximum* were collected for analysis at the dumping site. Soil was collected using soil auger while total plant of *P. maximum* was collected using cutlass, and placed in nylon bag. The samples (soil and the forage) were collected at distances of 0, 30 and 60 m using measuring tape and at a parallel of 2 m at each distance to give three different locations per distance as replicates. The soils were collected to a depth of 15 cm because grasses are fibrously rooted.

Laboratory procedure

Determination of the various antibiotics

The procedure of AOAC (2002) was used for the qualitative and quantitative test.

Qualitative test

A known weight of each sample was extracted with their corresponding solution and make up to 250 ml volume with distilled water in a 250 ml graduated flask. The mixture was placed in a steam bath for 10 min till a corresponding colour precipitate begins to form (white, pink or red, bluish green and yellowish colour for tetracycline, chloramphenicol, penicillin and streptomycin respectively). This confirms the presence of antibiotics.

Quantitative test

The mixture above was filtered through a Whatman No. 42 filter paper into a 100 ml volumetric flask and made up to mark with distilled water. The filtrates were pipetted into a 100 ml beaker, 5 ml of freshly prepared 0.10% sodium solution added and allowed to stand for 3 min. Standard solutions of various antibiotics of specific range in µg/ml were prepared and treated like sample extract above. The absorbencies of sample as well as standard were read against blank on a spectronic 21D spectrophotometer at a wavelength 510 nm.

$$\% \text{ Antibiotics} = \frac{\text{Absorbance of sample} \times \text{G radiant factor} \times \text{Dilution}}{10,000}$$

Statistical analysis

All data collected were subjected to one way analysis of variance (ANOVA) using SPSS (1999) software package. Differences between means were separated using Duncan Multiple Range test of the same package.

RESULTS

The presence of antibiotics in soil around poultry manure dumping site is as shown in Table 1. Qualitatively, all the antibiotics were present in all the treatments. Penicillin, tetracycline and chloramphenicol decreased as the distance increased. Table 2 showed the presence of antibiotics in *P. maximum* around poultry manure dumping site. All the values showed a positive result for the antibiotics at all the distances reported. However, quantitatively, values for penicillin, streptomycin and tetracycline decreased as the distance increased. Values for chloramphenicol were not consistent with increasing

Table 3. The organic content of soil collected at poultry manure dumping site.

Parameter	0 m	30 m	60 m	SEM
% Organic carbon	1.28 ^{ab}	1.11 ^b	1.38 ^a	0.04
% Organic matter	2.20 ^b	1.91 ^c	2.38 ^a	0.61
pH	6.10 ^b	6.20 ^{ab}	6.30 ^a	0.31

^{abc} Mean values in the same row are significantly different ($p < 0.05$).

distance from dumping sites. Lower values were observed for streptomycin as compared with others. Apart from chloramphenicol that recorded a higher ($p < 0.05$) value at 30 m (25.48×10^{-7} $\mu\text{g}/\text{kg}$), the concentrations were generally significantly ($p < 0.05$) higher at the 0 m for all the antibiotics. The result showed a progressive increase in the pH with the increase in the distance from the dumping site (Table 3). The lowest ($p < 0.05$) values were found in 30 m distant from the dumping site for both organic carbon and organic matter and this made the trend to be inconsistent.

DISCUSSION

Quantitatively (Table 1) the presence of the detected antibiotics ranged between $3.38 - 20.53 \times 10^{-7}$ $\mu\text{g kg}^{-1}$. Antibiotics present in the soil was lower than values gotten by De Liguoro et al. (2003) who detected oxytetracycline in soil ($< 5 \mu\text{g kg}^{-1}$ of soil) at a 60 cm depth after treatment with cattle manure while Hamscher et al. (2002) reported the presence of tetracycline ($170 \mu\text{g kg}^{-1}$) at a 30 cm soil depth, 6 month after liquid swine manure was applied. Antibiotics have also been detected in rivers (Kolpin et al., 2002, 2004) and marine sediments (Nygaard et al., 1992).

Boxall et al. (2004) observed that persistence of antibiotics in soil depends on many factors including soil type, climate, and class of antibiotics. Most antibiotics are biodegradable in soils but some of them have a long shelf-life. Antibiotics in manure-applied land may leach to ground water or move to surface waters through surface runoff and this explained why at 60 m away from the dumping site, it was still found present.

Table 2 revealed the presence of antibiotic in *P. maximum* found around poultry dumping sites. Higher values were obtained at 0 m distance in all the antibiotics. This was however, expected since this is the actual site where manure is dumped. It also showed that antibiotics given to animal were not yet fully decomposed. Levy (1992) and Thiele-Bruhn (2003) reported that up to 80% of the orally administered antibiotics may pass through the animals unchanged and end up in manure and later find their way into the terrestrial environment when

manure from antibiotic-fed animals is applied as a source of crop nutrients.

Across the treatments critical observation of the results revealed that values for streptomycin was generally low compared to others at all the distances. This could be as a result that it was less used or it has a low frequency of usage by the farmers or probably the rate of absorption by the grass was low. The value gotten for *P. maximum* around poultry manure dumping site showed a decrease in value as the distance increased, an indication of the topography due to run off effect.

With the findings of this work, the use of antibiotics by livestock's farmers has affected the forages found around the environment and this in turn may affect grazing animals around this area. Mamber and Katz (1985) reported that the rampant use of antibiotics resulted in development and maintenance of populations of antibiotic-resistant gram-negative enteric bacilli in the intestinal tracts of animals making them reservoirs for resistant bacteria.

The soil organic carbon, organic matter and the pH were also affected with the presence of these antibiotics. According to Nygaard et al. (1992), antibiotics have both quantitative and qualitative effects on the native microbial communities in the terrestrial environment. Although antibiotic concentrations in most soils are not at therapeutic levels to cause inhibitory effects on bacterial population, they may still influence the selection of antibiotic resistant bacteria in the environment (Nygaard et al., 1992; Kümmerer, 2003). An increase in antibiotic resistance in soil bacteria after manure application may be due to (i) exchange of genetic elements between soil bacteria and antibiotic resistant manure bacteria, (ii) exchange of genetic elements among antibiotic producing soil microorganisms, or (iii) *in situ* selection pressure from a low level of antibiotics in manure (Onan and LaPara, 2003).

Conclusion

Based on the findings of this work, it could be concluded that antibiotics were present in *P. maximum* and soil collected at poultry dumping sites up to 60 m distance

from dumping site. The quantity for both the forage and soil however decreased as the distance increased.

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