

## EFFECTS OF TREATED POULTRY LITTER ON POTENTIAL GREENHOUSE GAS EMISSION AND FIELD APPLICATION

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### ABSTRACT

*This study examined the effects of different treatments of poultry faecal matter on potential greenhouse gas emission and its field application. Poultry litters were randomly assigned to four treatments viz; salt solution, alum, air exclusion and the control (untreated). Alum treated faeces had higher ( $p < 0.05$ ) percentage nitrogen retention than salt and air-tight treatments, which had higher ( $p < 0.05$ ) moisture content when compared with the control. The pH level was lowest in alum treated faecal matter (6.03,  $p < 0.05$ ), and highest in the control (7.37,  $p < 0.05$ ). Similarly, alum treated faeces had significantly lower mean temperature (28.58°C,  $p < 0.05$ ) control, salt and air-tight treatments, with air-tight treated faeces having the highest temperature (29.44°C,  $p < 0.05$ ). Nitrogen depletion rate was significant lower ( $p < 0.05$ ) in alum treated faecal matter than in salt and air-tight treatments. Post-storage, samples treated with alum increased substantially ( $\geq 46.51\%$ ) in total microbial count, total viable count was lower ( $p > 0.05$ ;  $2.83 \times 10^6$  cfu/ml) in air-excluded treatment. Maize seeds planted on alum treated and air-excluded litter soils had an average germination percentage (GP) range of 65 – 75% and 54 – 75%, respectively. These figures were found to be mildly comparable to the control which averaged a germination index of 75%. Sorghum plots recorded a mean value of 99% GP on alum treated soil two weeks after planting, slightly surpassing air-tight treated soils with mean value of 89% GP. Average maize height was 48cm and 23cm for alum and air-tight treatment, respectively after 21 days of planting, in contrast to mean height of 25cm on the sorghum plots. Seeds planted on salt treated plots did not germinate. The study suggests that alum treated poultry litter was superior in mitigating the tendency for nitrogenous losses as evident in its lower nitrogen depletion rate, pH, weight, temperature and potential field application index.*

**Keywords:** Greenhouse gas, Field application, Poultry litter, Alum, Air exclusion, Germination

### INTRODUCTION

The poultry industry is one of the largest and fastest growing agro-based industries in the world resulting in production of volumes of faecal litter. This litter represents a valuable source of microbial energy and nutrient, livestock feed, crop fertilizer and potential source of bio-fuel (Williams *et al.*, 1999). However, the major problems encountered in the accumulation of large amount of faecal wastes generated by intensive production are

that of proper disposal and pollution, unless environmentally and economically sustainable management technologies are employed (Power and Dick, 2000). Dust, odours and bio-aerosols (e.g. microbes, endotoxins and mycotoxins suspended in air) generated at production, manure storage facilities and during land spreading of poultry litter constitute the most frequent source of complaints against animal-based industries (Millner, 2009). Uncontrolled decomposition of manure produces odorous gases, including amines, amides, sulphides and

disulphide. These noxious gases can cause respiratory diseases in animals and humans (Schiffman and Williams, 2005). Ammonia volatilization from manure causes odour (Williams, 1995; Wheeler *et al.*, 2006) and may also contribute to atmospheric ammonia enrichment and acid rain (Walker *et al.*, 2000a; 2000b). Emission of ammonia ranges from 0.034 – 0.384 kg NH<sub>3</sub>/bird/year for various layer systems (Koerkamp, 1994; Yang *et al.*, 2000) and this could adversely affect the health and welfare of the flock (Dawkins *et al.*, 2004; Ritz *et al.*, 2004), resulting in lower feed efficiencies and increased costs due to the need to remove ammonia, usually through ventilation of the house (Moore *et al.*, 1995). Extended exposure (8 to 10 hours) of humans to these ammonia levels have also been shown to negatively affect the welfare of farm operators (Ritz *et al.*, 2004; Kirychuk *et al.*, 2006; Rylander and Carvalheiro, 2006). In addition to health issues, ammonia can also be a major source of pollution (Koerkamp, 1994; Williams, 1995), causing eutrophication of surface waters (Edwards and Daniel, 1992; Paerl and Fogel, 1994) and acidification of soils (Williams *et al.*, 1999). Addition of poultry manure to soils not only helps to overcome the disposal problems but also enhances the physical, chemical and biological fertility of soils (Friend *et al.*, 2006; McGrath *et al.*, 2009) and likewise increasing the organic matter content, water holding capacity, oxygen diffusion rate and the aggregate stability of the soils (Mahimairaja *et al.*, 1995; Adeli *et al.*, 2009). However, pollution and nuisance problems can occur when manure is applied under environmental conditions that do not favour agronomic utilization of the manure-borne nutrients such as denitrification to release N<sub>2</sub>O, a greenhouse gas (Casey *et al.*, 2006; Kaiser *et al.*, 2009). It is quite expedient, therefore, to subject poultry manure to several treatments in order to improve its storage and handling and to minimize the risk of disease transmission and environmental pollution. This study evaluated the effectiveness of different treatment methods of poultry litters to conserve the nutrients and the potentials of such treated litters for field application.

## MATERIALS AND METHODS

**Materials and Experimental Design:** The study was carried out at the Animal Pavilion, Department of Animal Production, University of Ilorin, Nigeria. Ilorin is located on latitude 08 29'N and longitude 004 35'E with annual temperature range of 22 – 34 °C and annual precipitation range of 80 – 120mm (World Climate, 2013). Fresh faecal samples were collected randomly from laying birds at the Teaching and Research Farm, University of Ilorin, over a three hour period. The samples were assigned to four different treatments each replicated in three units using the completely randomized design (CRD). The treatments were; faecal sample treated with alum, faecal sample kept air tight, faecal sample treated with salt solution and the control (untreated). One kilogram of the faecal matter was weighed into separate pots before subjecting them to the various treatments; salt solution at 350g/L of water was used as salt treatment, while alum at 10% of faecal weight (100g) was used as alum treatment (Moore *et al.*, 1995). Samples were placed under a shed for eight (8) weeks and subsequently applied to soil during the agronomic evaluation of maize and sorghum performance. Three seeds were planted per potting medium while the plant height and number of leaves was taken weekly.

**Physico-chemical Parameters Tested:** Parameters measured include nitrogen retention, moisture content, weight, temperature, pH and microbial population. Nitrogen retention test was carried out bi-weekly by subjecting the samples to acid digestion and distillation (AOAC, 1990), the temperature range was determined using a thermometer and pH determined bi-weekly using a pH meter. The microbial count and identification were carried out fortnightly at the Microbiology Laboratory, University of Ilorin.

### Microbial Population and Identification

**Potato dextrose agar (PDA):** Thirty nine (39) grams of Potato dextrose Agar powder was weighed and suspended in 1 liter of distilled

water. It was shaken properly and heated to dissolve the powder completely. After heating, the flask was plugged with cotton wool and wrapped with aluminum foil, sterilized in the autoclave at the temperature of 121°C for 15 minutes and allowed to cool to about 45°C after which 1% streptomycin powder was added before pouring aseptically into the Petri-dishes.

**Serial dilution technique:** This method was used for the enumeration of all bacteria and fungi. 1ml of each of the samples was transferred to 9mls of sterile distilled water in a test tube. Serially, 1ml was taken from the 10<sup>-1</sup> tube to another tube which make 10<sup>-2</sup> this was done up to 10<sup>-6</sup> dilution. Then 1ml was taken from the 10<sup>-5</sup> dilutions respectively into a sterile Petri-dish and the molten agars that has been prepared earlier for bacterial growth were allowed to cool to about 45°C before pouring into each of the sterile Petri-dishes and allowed to solidify, 10<sup>-2</sup> dilution was taken for the growth of fungal. These steps were followed for the fungal growth, after solidification each of the plates was incubated at 37°C for bacteria and at room temperature for fungi. Incubation period was between 24 – 48 hours and 72 hours for fungi. After incubation period, each of the plates were examined and counted.

**Characterization and identification of bacteria isolates:** The isolates were characterized and identified after obtaining pure culture of isolates through repeated sub-culturing by using their colonial morphology such as colony shape, edge, surface texture elevation, pigmentation, consistency and optics; Cellular morphology tests including Gram staining and motility test and biochemical reactions on the bacteria isolates for possible identification, which includes catalase test (Fawole and Oso, 2007), coagulate test, oxidase test, indole test, starch hydrolysis and sugar fermentation test.

**Statistical Analysis:** Data obtained from the experiment were subjected to analysis of variance using the SPSS version 16.0 and significant means were separated using the Duncan Multiple Range Test (Duncan, 1955).

## RESULTS AND DISCUSSION

**Physico-chemical Profile:** The effects of the various treatments on faecal samples indicated that alum treated faeces had higher ( $p < 0.05$ ) nitrogen retention percentage (Table 1). Alum did not completely stop the process that resulted in gaseous nitrogenous emission (denitrification and ammonification) during storage but have been reported to slow it down (Moore *et al.*, 1995; Malomo *et al.*, 2014). Addition of alum [ $Al_2(SO_4)_3 \cdot 14H_2O$ ] has been used as a cost effective means of reducing ammonia volatilization from poultry litter in houses (Moore *et al.*, 2000; Gilmour *et al.*, 2004). Salt and air-tight treated faeces were lower ( $p < 0.05$ ) in nitrogen content and higher ( $p < 0.05$ ) in moisture compared with the control, which implies high ammonia volatilization and litter quality deterioration (Griffin, 1981; Elliot and Collins, 1983). The pH level was lowest in alum treatment (6.03,  $p < 0.05$ ). This corroborated with previous investigations (Reece *et al.*, 1985; Wheeler *et al.*, 2006) which suggested that litter pH should be below 7 to reduce ammonia volatilization.

Salt and air-tight treated faeces were observed to fall within this range, with the exception of the control (7.37), pH however tends towards alkalinity as the storage duration increases. Similarly, Alum treatment showed a significantly lower temperature (28.58°C,  $p < 0.05$ ) with air-tight treatment being the highest (29.44°C,  $p < 0.05$ ). It has been shown that at higher temperatures and under reduced conditions,  $NO_3$  will be denitrified more rapidly and  $N_2O$  which is harmful to the ozone layer will be formed more quickly (Ghaly and Macdonald, 2012). Nitrogen depletion rate was significant lower ( $p < 0.05$ ) in alum treated poultry manure (Table 2) when compared to salt and air-tight treatment, indicating its superior nitrogen sequestration (Moore *et al.*, 2000).

**Microbial Population:** Bacteria species identified in the treated poultry litter includes; *Staphylococcus aureus*, *Enterobacter aerogenes*, *Proteus vulgaris*, *Escherichia coli*, *Bacillus subtilis*. Identified fungi isolated include *Aspergillus niger*, *Neurospora crassa*, *Penicillium*

**Table 1: Effects of treatments on physico-chemical profile of poultry faecal samples**

Parameters	Control	Salt solution	Alum	Air Exclusion
Nitrogen (%)	2.21 ± 0.02 <sup>b</sup>	1.70 ± 0.06 <sup>a</sup>	2.56 ± 0.03 <sup>d</sup>	1.81 ± 0.05 <sup>c</sup>
pH	7.37 ± 0.04 <sup>d</sup>	6.35 ± 0.11 <sup>b</sup>	6.03 ± 0.16 <sup>a</sup>	6.79 ± 0.15 <sup>c</sup>
Weight (kg)	0.79 ± 0.17 <sup>a</sup>	2.52 ± 0.16 <sup>d</sup>	1.65 ± 0.08 <sup>c</sup>	1.20 ± 0.15 <sup>b</sup>
Temperature (°C)	28.95 ± 0.12 <sup>b</sup>	29.06 ± 0.12 <sup>b</sup>	28.58 ± 0.02 <sup>a</sup>	29.44 ± 0.08 <sup>c</sup>
Moisture (%)	66.78 ± 1.25 <sup>a</sup>	75.36 ± 3.24 <sup>b</sup>	76.96 ± 2.16 <sup>b</sup>	78.24 ± 3.52 <sup>b</sup>

Means with different superscript are significant ( $p < 0.05$ )

**Table 2: Rate of nitrogen depletion in treated poultry faecal samples**

Parameter	Control	Salt solution	Alum	Air Exclusion
N <sub>2</sub> (%)	0.57 ± 0.24 <sup>a</sup>	1.34 ± 0.81 <sup>b</sup>	0.42 ± 0.25 <sup>a</sup>	1.22 ± 0.83 <sup>b</sup>

Means with different superscript are significant ( $p < 0.05$ )

**Table 3: Effect of treatments on microbial population of treated poultry faecal samples**

Parameters (cfu/ml)	Control	Salt solution	Alum	Air Exclusion
<b>Baseline</b>				
TVC (10 <sup>6</sup> )	4.53 ± 1.02 <sup>c</sup>	3.53 ± 1.20 <sup>bc</sup>	2.30 ± 1.23 <sup>a</sup>	2.13 ± 0.19 <sup>a</sup>
TCC (10 <sup>6</sup> )	3.47 ± 1.02 <sup>c</sup>	0.97 ± 0.02 <sup>a</sup>	1.06 ± 0.58 <sup>b</sup>	1.67 ± 0.05 <sup>b</sup>
FC (10 <sup>3</sup> )	1.73 ± 1.25 <sup>b</sup>	1.40 ± 1.46 <sup>a</sup>	1.50 ± 1.24 <sup>a</sup>	1.43 ± 1.56 <sup>a</sup>
<b>Week 4</b>				
TVC (10 <sup>6</sup> )	2.37 ± 0.14 <sup>a</sup>	3.80 ± 0.13 <sup>b</sup>	3.93 ± 0.12 <sup>b</sup>	2.30 ± 1.24 <sup>b</sup>
TCC (10 <sup>6</sup> )	1.70 ± 0.06 <sup>b</sup>	0.13 ± 0.19 <sup>a</sup>	2.83 ± 0.10 <sup>c</sup>	1.73 ± 0.12 <sup>b</sup>
FC (10 <sup>3</sup> )	2.07 ± 0.23 <sup>b</sup>	1.67 ± 0.51 <sup>ab</sup>	1.80 ± 0.68 <sup>ab</sup>	1.13 ± 0.61 <sup>a</sup>
<b>Week 8</b>				
TVC (10 <sup>6</sup> )	4.33 ± 0.25 <sup>b</sup>	4.57 ± 0.32 <sup>b</sup>	4.30 ± 0.34 <sup>b</sup>	2.83 ± 0.15 <sup>a</sup>
TCC (10 <sup>6</sup> )	3.07 ± 0.71 <sup>b</sup>	3.73 ± 0.42 <sup>b</sup>	3.27 ± 0.52 <sup>b</sup>	2.33 ± 0.26 <sup>a</sup>
FC (10 <sup>3</sup> )	1.47 ± 0.32 <sup>b</sup>	1.57 ± 0.42 <sup>b</sup>	1.37 ± 0.35 <sup>b</sup>	0.53 ± 0.016 <sup>a</sup>

Means with different superscript are significant ( $p < 0.05$ ). TVC- Total Viable Count, TCC- Total Coliform Count, FC- Fungal Count.

*chrysogenum*, *Aspergillus flavus*, *Saccharomyces cerevisiae*, *Rhizopus stolonifer*. The baseline analysis had similar ( $p > 0.05$ ) TVC, TCC and FC values across the treatments (Table 3). This trend was observed to be especially true with total viable count (TVC) which tends to persist on to the eight week of study. As the period of storage lingered, faecal samples treated with alum increased substantially ( $\geq 46.51\%$ ) in total microbial count which indicated an ideal environment for microbial proliferation. Microbial population of poultry litter can be as high as  $10^9$  to  $10^{10}$  cells per gram of litter (Lovanh *et al.*, 2007; Rothrock *et al.*, 2007) especially as more than half of the nitrogen in poultry litter is lost as ammonia due to microbial activity (Moore *et al.*, 1995). Fungi, specifically *Aspergillus* spp., are known to

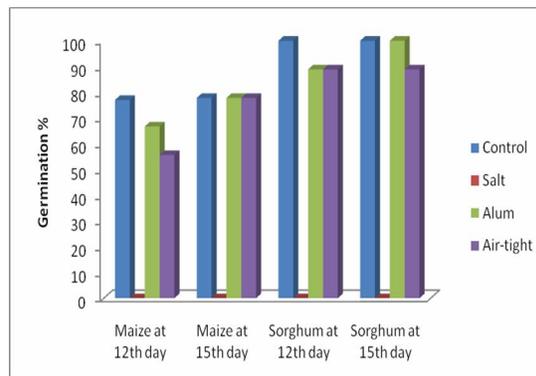
mineralize these predominant forms of organic nitrogen in poultry litter, and this fungal activity tends to proliferate. The microbial population is responsible for both beneficial (carbon mineralization, competitive exclusion, etc.) and detrimental (ammonia and N<sub>2</sub>O (GHG) production, pathogen persistence) effects of litter as was also the case of salt treated faeces (Table 3). At the end of the 8<sup>th</sup> week, TVC was lower ( $p > 0.05$ ;  $2.83 \times 10^6$  cfu/ml) in air-excluded treatment which was indicative of the strict anaerobic condition of the air tight environment which prevented microbial proliferation. Under anaerobic conditions, carbon dioxide production and decomposition rate of organic constituents are generally lower resulting in acid formation, a decrease in pH and ammonium nitrogen accumulation (Acharya, 1935).

### Agronomic Effects of Treated Poultry Feecal Samples:

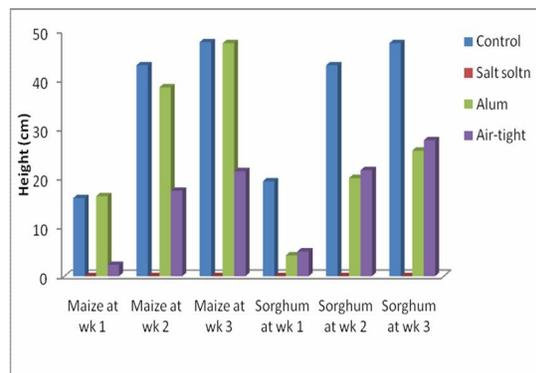
It was observed that maize seeds planted on alum and air-tight treated litter soils had an average germination percentage (GP) range of 65 – 75% and 54 – 75%, respectively (Figure 1). These figures were found to be mildly comparable to the control with averaged a germination index of 75%. Sorghum plots recorded a mean value of 99% GP on alum treated soil two weeks after planting, slightly surpassing air-tight treated soils with mean value of 89% GP. Previous studies have highlighted the beneficial effects of alum treated litter and its field application as this result also suggests. Alum lowers water soluble phosphorous (Moore and Miller, 1994), and soils applied with alum-treated litter have fewer estrogenic compounds (Nichols *et al.*, 1997), pathogens (Gandhapudi *et al.*, 2006), heavy metals and greater nitrogen content (Moore *et al.*, 1995). Long term studies conducted by Moore and Edwards (2005) showed that exchangeable aluminum levels in soils fertilized with normal and alum-treated litter are low (less than 1 mg Al/kg soil) and are not significantly different, whereas plots fertilized with the same amount of nitrogen from ammonium nitrate have very high exchangeable aluminum (up to 1 00 mg Al/kg soil). Moore and Edwards (2005) also showed that tall fescue yields from long-term studies were highest with alum-treated litter, followed by normal litter and lowest with ammonium nitrate. The average height of maize stand was 48 cm for alum treatment after 21 days of planting, air-tight treatment averaged 23 cm per stand, both treatments recorded a mean height of 25 cm on the sorghum plots which suggested slower growth rate compared to the control (with mean plant height of 48 cm) (Figure 2). However, the long-term effects of applying alum treated litter to land have indicated that this practice is sustainable (Moore and Edwards, 2005). Seeds planted on salt treated soil did not germinate, possibly due to exosmosis.

**Conclusion:** The result of this study suggests that alum treated poultry litter was superior in mitigating the tendency for nitrogenous losses as evident in its lower nitrogen depletion rate,

pH, Weight and temperature. Alum treatment supports effective nutrient release in field application for crop production.



**Figure 1: Germination percentage of maize and sorghum cultivated using treated poultry fecal samples**



**Figure 2: Average height of maize and sorghum cultivated using treated poultry fecal samples**

These observations may tend to encourage the use of alum for the treatment and field application of poultry litter, thus, mitigating the twin effects of environmental pollution and emission of GHG, and the use of treated litters for field applications. Further studies on longer storage and field application periods may be necessary for the reduction of GHG and environmental pollution.

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