



Chemical and Microbial Analysis of Bedding Material (Wood Shavings) Treated with Graded Levels of Aluminium Sulphate (Alum)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The study was carried out at the poultry unit of the Department of Animal Science teaching and research farm, Ahmadu Bello University, Zaria to determine the chemical and microbial analysis of poultry litter (wood shavings) treated with graded levels of alum. The alum used was obtained from the Sabon-gari market in Zaria, Kaduna State. The rates of alum application (prior to keeping the birds) was as follows: T1 control (normal wood shavings with no alum), T2 (5% alum by kg weight of wood shavings), T3 (10% alum by kg weight of wood shavings) and T4 (15% alum by kg weight of wood shavings). Five sets of litter samples were obtained fortnightly from each pen from different locations i.e. the four corners and center from which the microbial load, pH, total nitrogen (N), soluble reactive phosphorus, VFA and NH_4^+ concentration were measured. The result shows significantly ($P < 0.05$) lower pH value in all the alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to the control. The result showed that significantly ($P < 0.05$) lower total volatile fatty acid level was obtained in all the alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to the control untreated wood shaving group. The results showed a decrease in total bacteria, *E. coli* and *Salmonella spp.* load in alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to

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the control, while mould and yeast load was increased in all the alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to the control. The study conclude that treating wood shavings with alum can reduce microbial load of the litter, hence improve health and reduce mortality. Treating wood shavings with alum tends reduce the microbial load of the litter.

Keywords: Aluminium sulphate; wood shavings; poultry; chemical; microbial.

1. INTRODUCTION

Some of the major environmental issues faced by the poultry industry are the deposition of high amount of wastes especially litter. Phosphorus (P) content in poultry waste has long been recognized as a significant environmental problem in the poultry industry [1]. Manure odour produced by anaerobic decomposition of livestock wastes [2] is also a challenge. However, most of the organic matter in manure is microbially transformed into non-odorous end products under aerobic conditions [3]. Factors that determine odour production from livestock facilities include manure, spilled feed, bedding materials and moisture [4]. The most significant odorous compounds in manures are volatile fatty acids (VFA; C2 to C9), Volatile organic acids and aromatic compounds [5].

Acidifiers are the types of substances that create an acidic condition (pH less than 7) in the litter resulting in more of the ammoniacal-N being retained as ammonium ions (NH_4^+) rather than ammonia (NH_3). The acidity also creates unfavourable conditions for the bacteria and enzymes that contribute to ammonia formation resulting in reduced ammonium ions. Many different types of acidifiers such as aluminium sulphate (alum), sodium bisulphate, ferrous sulphate and phosphoric acid were found to be effective in controlled studies [6]. However, some acidifiers are not recommended for use in poultry houses for reasons such as bird toxicity (ferrous sulphate) or increased phosphorus (P) levels in the already P-rich litter (phosphoric acid) [6].

Aluminium sulphate (alum) is an acidifier that is commonly used in poultry litter treatment under the brand name Al Clear. It is available in either a dry or liquid form. Broiler chickens grown for meat are usually raised on a floor covered with bedding materials. The combination of bedding material, bird excreta and waste feed is referred to as litter. Historically, bedding related research has focused on the effects of type of bedding

materials on broiler performance under various management scenarios [7].

2. MATERIALS AND METHODS

2.1 Experimental Site and Location

The study was carried out at the poultry unit of the Department of Animal Science teaching and research farm, Ahmadu Bello University, Zaria. The pen is located in northern guinea savannah zone of Nigeria, latitude $11^{\circ} 09' 76''$ N and longitude $7^{\circ} 38' 20''$ E at an altitude of 610 mm above sea level. The climate is relatively dry with a mean annual rainfall of 700-1400mm, occurring between the months of April and September [8].

2.2 Experimental Diets and Material

The alum used was obtained from the Sabongari market in Zaria, Kaduna State. Aluminium sulphate (alum) was applied to the wood shavings by mixing it with alum thoroughly using hands covered with hand gloves. The rates of alum application was as follows: T1 control (normal wood shavings with no alum), T2 (5% alum by kg weight of wood shavings), T3 (10% alum by kg weight of wood shavings) and T4 (15% alum by kg weight of wood shavings). Feed and water were supplied *ad libitum* throughout the 56 days study period and routine vaccination schedule was administered.

2.3 Data Collection and Analyses

2.3.1 Litter sample collection

Five sets of litter samples were obtained fortnightly from each pen from different locations i.e. the four corners and center from which the microbial load, pH, total nitrogen (N), soluble reactive phosphorus, VFA and NH_4^+ concentration were measured. Litter samples were taken by removing the first 10mm of the exposed surface from each location set. The samples from each pen were mixed and homogenized to make one sample and was

refrigerated before being taken to the laboratory for analyses.

2.3.2 Microbial analysis of litter

Each homogenous sample mixture from each pen was pooled in one sterile flask as representing sample from one pen and were analysed for microbes in the Department of Microbiology, Ahmadu Bello University Zaria. The flasks were shaken and then serial dilutions were made on each. Each dilution were streaked on plate count containing Nutrient Agar (NA) for total bacteria count, eosine methylene blue Agar (EMB) for *E. coli*, bismuth sulfide agar (BSA) for *Salmonella spp.* and potato dextrose agar (PDA) for mould and yeast and incubated at room temperature. Suspected colonies were inoculated in triple sugar iron agar for confirmation. Standard plate counting techniques were used for total bacteria, *E. coli*, *Salmonella spp.* and mould and yeast counts as described by Yardimci and Kenar [9]. Sample for *Eimeria spp.* was taken to the parasitology laboratory at the Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria for *Eimeria* parasite count using the modified McMaster egg/oocyst count technique [10].

2.3.3 Chemical analysis of litter

The litter samples were analyzed for pH, ammonium ion (NH_4^+) concentration, soluble reactive phosphorus and total nitrogen at the Department of Agronomy, Ahmadu Bello University, Zaria while samples for total VFA were analysed at the chemical laboratory of National Animal Production Research Institute, Zaria, Kaduna State. A 20-g subsample of the litter sample was extracted with 200 ml of deionized water for 2 hours on a mechanical shaker, then centrifuged at $3,687 \times g$ for 15 minutes [11]. Aliquots were taken for pH, total nitrogen, NH_4^+ , soluble reactive phosphorus (SRP), and total VFA. Unfiltered samples were used for pH using a pH meter and were analyzed immediately. Samples for total nitrogen and ammonium ions were filtered through a 0.45- μm membrane filter and were determined using Kjeldahl method with Kjeldahl apparatus as described by [12]. Samples to be tested for soluble reactive phosphorus were filtered through a 0.45- μm membrane filter, acidified to a pH of 2.0 with HCl and frozen until when required for analyses [13]. Soluble reactive phosphorus was determined using the Bray1 method with an auto-analyzer (Spec 20D) according to APHA (1992).

Samples for total VFA were not filtered but frozen until when required for analyses [14]. Total VFA was analyzed using steam distillation technique with steam distillation apparatus as described by Chakrabarty [15].

2.4 Statistical Analyses

All the data collected from the experiment were subjected to analysis of variance (ANOVA) using the general linear model of statistical analysis system [16] software package and the mean separation was done using Duncan multiple range test.

3. RESULTS AND DISCUSSION

3.1 pH Levels of Alum Treated and Untreated Wood Shavings

Fig. 1, shows the pH levels of alum treated and untreated wood shavings fortnightly i.e. week 2, week 4, week 6 and week 8. The result shows significantly ($P < 0.05$) lower pH value in all the alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to the control (0% alum treated wood shaving). The results follow the same trend in week 4 and 6, while in week 8 the pH of Treatment 4 (15% alum treated wood shaving) is significantly lower ($P < 0.05$) compared to the other treatments.

3.2 Total Nitrogen Levels of Alum Treated and Untreated Wood Shavings

The result of total nitrogen levels of alum treated and untreated wood shavings a fortnightly during the research period is presented in Fig. 2. The result showed significantly ($P < 0.05$) higher nitrogen levels in all the alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to the control untreated wood shaving (0% alum treated wood shavings).

3.3 Soluble Reactive Phosphorus Levels of Alum Treated and Untreated Wood Shaving

The fortnightly soluble reactive phosphorus levels of alum treated and untreated wood shavings is presented in Fig. 3. The result showed significantly ($P < 0.05$) lower soluble reactive phosphorus levels in all the alum treated wood shavings (5%, 10% and 15% alum treated wood shavings) compared to the control

untreated wood shavings (0% alum treated wood shaving).

3.4 Total Volatile Fatty Acid Levels of Alum Treated and Untreated Wood Shavings

Fig. 4 shows the fortnightly total volatile fatty acid levels of alum treated and untreated wood

shavings. The result showed that significantly ($P < 0.05$) lower total volatile fatty acid level was obtained in all the alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to the control untreated wood shaving group (0% alum treated wood shaving).

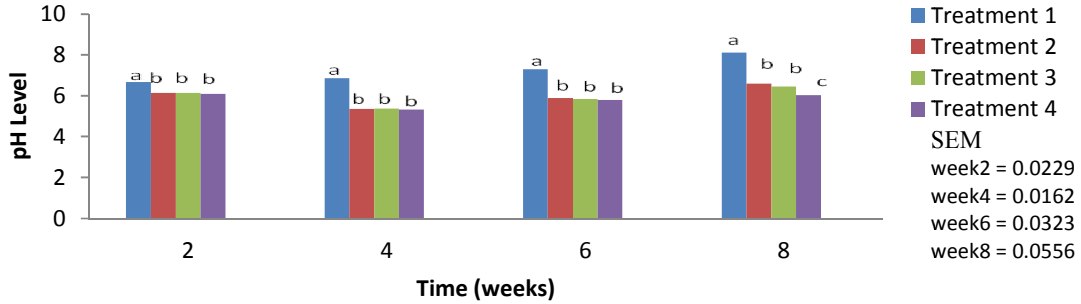


Fig. 1. pH Levels of Alum Treated and Untreated Wood Shavings

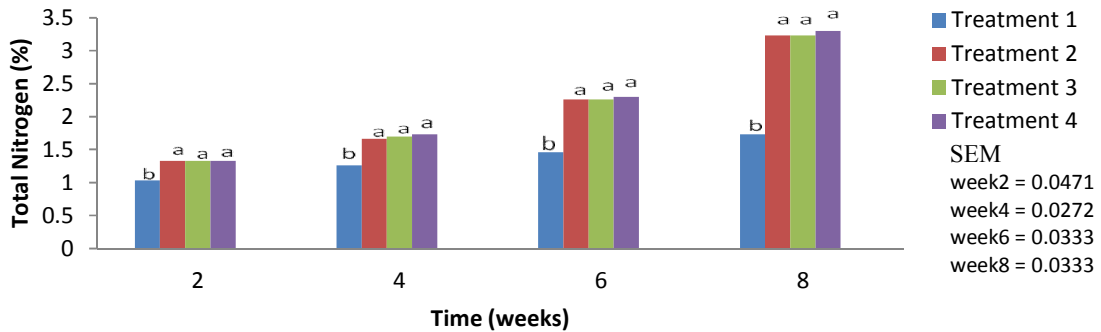


Fig. 2. Total Nitrogen of Alum Treated and Untreated Wood Shavings, ($P < 0.05$)

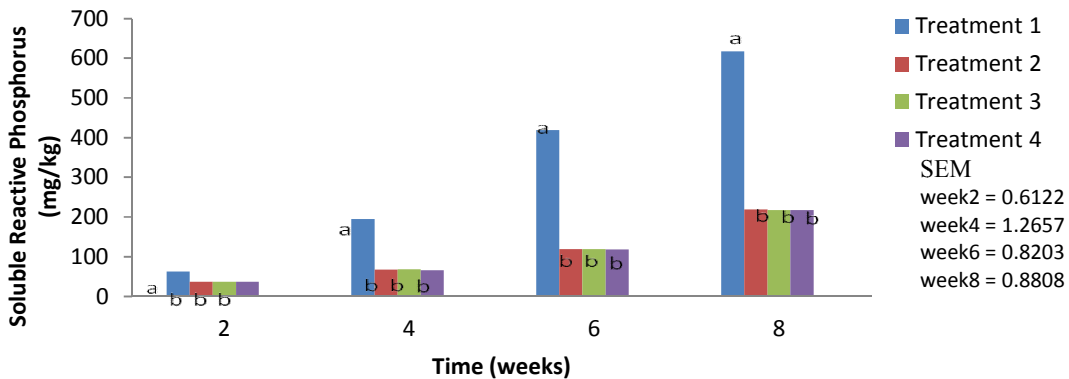


Fig. 3. Soluble Reactive Phosphorus of Alum Treated and Untreated Wood Shavings, ($P < 0.05$)

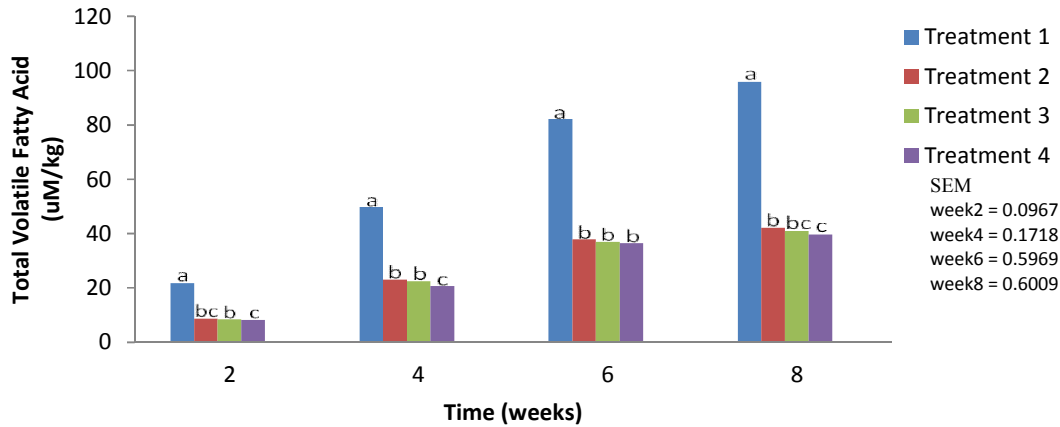


Fig. 4. Total Volatile Fatty Acid Levels of Alum Treated and Untreated Wood Shavings, (P<0.05)

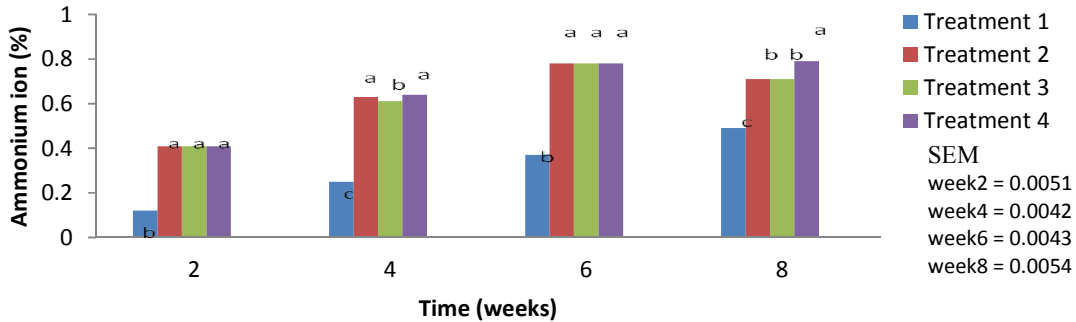


Fig. 5. Ammonium ion (NH₄⁺) of alum Treated and Untreated Wood Shavings, (P<0.05)

3.5 Ammonium ion (NH₄⁺) Levels of Alum Treated and Untreated Wood Shavings

The ammonium (NH₄⁺) ion concentrations of alum treated and untreated wood shavings is presented in Fig. 5. The result shows significantly (P<0.05) higher ammonium ion concentration in the alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to the control (0% alum treated wood shaving).

3.6 Microbial Load in Alum Treated and Untreated Wood Shavings

The results of the fortnightly effect of alum treated wood shavings on the microbial load of the litter are presented in Figs. 6-9. The results showed a decrease in total bacteria, *E. coli* and *Salmonella spp.* load in alum treated wood shavings groups (5%, 10% and 15% alum

treated wood shavings) compared to the control, while mold and yeast load was increased in all the alum treated wood shavings groups (5%, 10% and 15% alum treated wood shavings) compared to the control. The *Eimeria spp* parasite load in the litter remained at below detection levels throughout the period of the study.

The total bacteria load of the litter at week 2 were 2.53x10⁶cfu/g, 2.13x10⁵cfu/g, 2.03x10⁵cfu/g and 1.69x10⁵cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively, at week 4, the total bacteria load of the litter were 9.66x10⁶cfu/g, 3.15x10⁵cfu/g, 3.08x10⁵cfu/g and 2.87x10⁵cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively, at week 6, the total bacteria load of the litter were 8.53x10⁸cfu/g, 4.24x10⁶cfu/g, 4.11x10⁶cfu/g and 3.98x10⁶cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively and at week 8, the total bacteria load of the litter were 9.81x10⁸cfu/g, 8.76x10⁶cfu/g, 8.28x10⁶cfu/g and

7.48x10⁶cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively. The *E. coli* load of the litter at week 2 for 0%, 5%, 10% and 15% alum treated wood shavings were all at below detection levels, the *E. coli* load of the litter at week 4 for 0%, 5%, 10% and 15% alum treated wood shavings were 3.31x10⁶cfu/g, 2.72x10⁴cfu/g, 2.49x10⁴cfu/g and 2.03x10⁴cfu/g respectively, the *E. coli* load of the litter at week 6 for 0%, 5%, 10% and 15% alum treated wood shavings were 9.58x10⁶cfu/g, 6.47x10⁵cfu/g, 6.33x10⁵cfu/g and 5.75x10⁵cfu/g respectively and the *E. Coli* load of the litter at week 8 for 0%, 5%, 10% and 15% alum treated wood shavings were 6.08x10⁷cfu/g, 6.22x10⁶cfu/g, 6.15x10⁶cfu/g and 6.14x10⁶cfu/g respectively. The *Salmonella spp.* load of the litter at week 2 were 1.57x10⁶cfu/g, 1.63x10⁵cfu/g, 1.64x10⁵cfu/g and 1.44x10⁵cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively, at week 4, the *Salmonella spp.* load of the litter were 4.92x10⁶cfu/g, 2.18x10⁵cfu/g, 2.21x10⁵cfu/g and 2.50x10⁵cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively, at week 6, the *Salmonella*

spp. load of the litter were 6.22x10⁶cfu/g, 8.11x10⁵cfu/g, 8.46x10⁵cfu/g and 8.10x10⁵cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively and the *Salmonella spp.* load of the litter at week 8 were 9.94x10⁶cfu/g, 2.43x10⁶cfu/g, 2.29x10⁶cfu/g and 2.23x10⁶cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively. The mold and yeast load of the litter for 0%, 5%, 10% and 15% alum treated wood shavings at week 2 were 1.30x10⁴cfu/g, 2.50x10⁵cfu/g, 2.60x10⁵cfu/g and 2.50x10⁵cfu/g respectively, the mold and yeast load of the litter for 0%, 5%, 10% and 15% alum treated wood shavings at week 4 were 5.13x10⁴cfu/g, 7.86x10⁵cfu/g, 8.80x10⁵cfu/g and 7.80x10⁵cfu/g respectively, the mold and yeast load of the litter for 0%, 5%, 10% and 15% alum treated wood shavings at week 6 were 8.30x10⁴cfu/g, 7.86x10⁶cfu/g, 7.16x10⁶cfu/g and 7.10x10⁶cfu/g respectively and the mold and yeast load of the litter at week 8 were 1.60x10⁵cfu/g, 1.13x10⁷cfu/g, 2.50x10⁷cfu/g and 5.30x10⁷cfu/g for 0%, 5%, 10% and 15% alum treated wood shavings respectively.

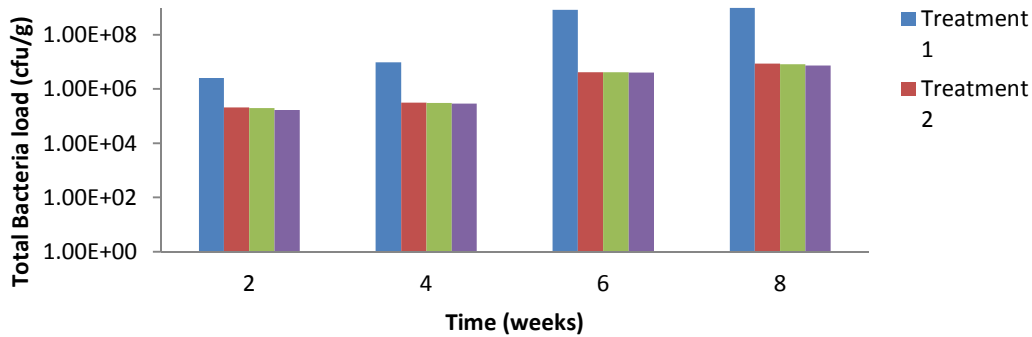


Fig. 6. Total Bacteria Load of Alum Treated and Untreated Wood Shavings

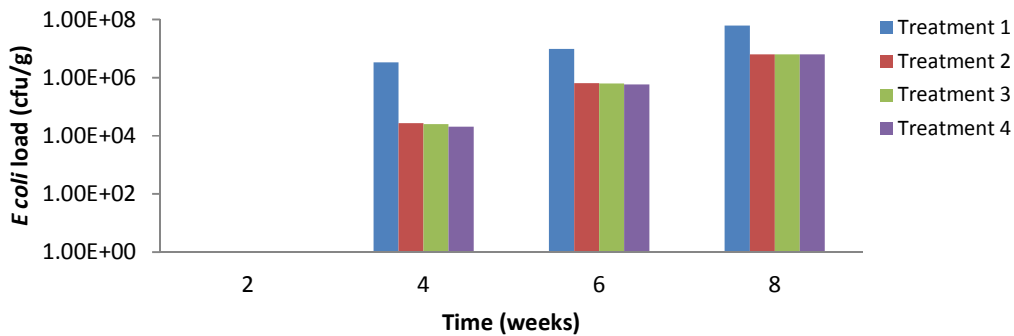


Fig. 7. *E. coli* load of alum treated and untreated wood shavings

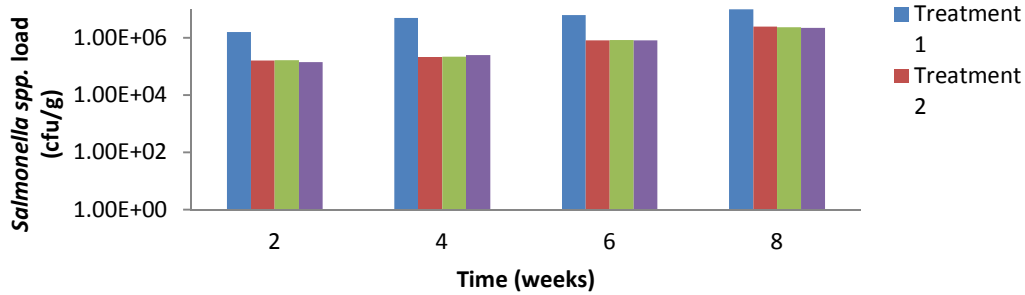


Fig. 8. *Salmonella* spp. load of alum treated and untreated wood shavings

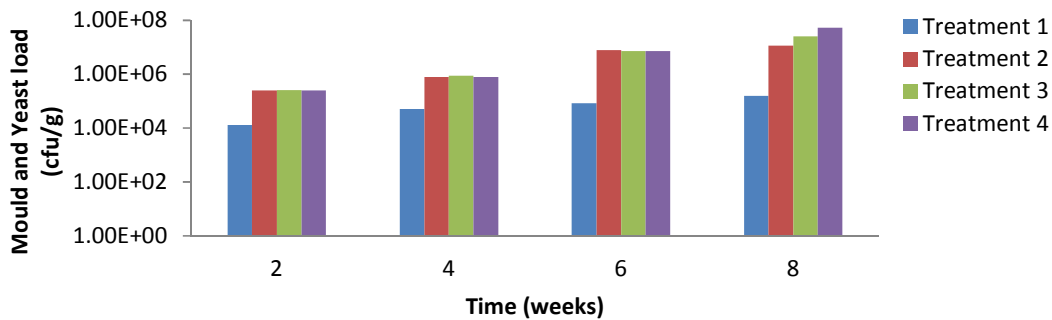


Fig. 9. Mold and Yeast Load of Alum Treated and Untreated Wood Shavings

4. DISCUSSION

4.1 pH of Alum Treated and Untreated Wood Shavings

The reduction in pH level observed in the alum treated wood shavings (5%, 10% and 15% alum treated wood shavings) compared to untreated wood shavings can be attributed to the reaction of alum with H_2PO_4 in the litter resulting in the generation of acidity in the litter as reported by [17]. This reduced pH level in the litter is similar to the result obtained by [18] and [19] who reported that alum additions to litter significantly reduced the pH of the litter. Litter pH was also significantly lowered with liquid $AlCl_3$ treatments to litter for up to 6 weeks [20].

4.2 Total Nitrogen of Alum Treated and Untreated Wood Shavings

The significantly higher nitrogen level observed in the alum treated wood shavings in this study might be due to the conversion ability of alum (aluminium sulphate) for nitrogen from volatile gas to a more stable form in the litter i.e. through

the conversion of NH_3 gas to $(NH_4)_2SO_4$ by the reaction of sulphate from alum with NH_3 in the litter as reported by [21]. The present result on significant higher nitrogen in alum treated litter was similar to various researchers [18], [19] and [17] who reported average total nitrogen contents of alum treated litter to be significantly higher compared to untreated litter. The higher total nitrogen in the litter, indicate that crop yields could be higher with litter treated with alum when used as manure as has been reported by [12] and [22].

4.3 Soluble Reactive Phosphorus Level of Alum Treated and Untreated Wood Shavings

The reduction of Soluble reactive phosphorus levels for 5%, 10% and 15% alum treated wood shavings groups by 64.53%, 64.74% and 64.76% respectively compared to the control (0% alum treated wood shaving group) is similar to the findings of [23] who reported that alum and $AlCl_3$ treatments reduced soluble reactive phosphorus concentrations in runoff by as much as 84% compared to normal manure. The significantly

lower soluble reactive phosphorus level observed in the alum treated wood shavings may be due to the impact of alum on the water solubility of phosphorus in the litter, thereby making the phosphorus in the litter less water soluble and hence reducing phosphorus runoff on land as reported by [18] and [19]. This result is also consistent with the findings of [24] and [25], who reported that Al, Ca, and Fe amendments reduced soluble phosphorus in animal manures. [24] Reported that Alum treated litter lowered phosphorus concentrations in runoff by 87% and 63% compared with untreated litter for the first and second runoff events, respectively. [20] Reported that concentrations of soluble reactive phosphorus were 83% lower for AlCl₃ (200 g/kg of rice hulls) treated litter. [18] Explained that one of the reasons alum was chosen for phosphorus control in poultry litter was because alum was stable over a very wide range of pH conditions.

4.4 Total Volatile Fatty Acid Level of Alum Treated and Untreated Wood Shavings

The significantly lower total volatile fatty acid levels in 5%, 10% and 15% alum treated wood shavings groups over time by as much as 56%, 57.33% and 58.58% respectively compared to the control is slightly higher than the report of [20], who reported 51% of total volatile fatty acid reduction with aluminium chloride treatment on poultry litter, thereby reducing the odour in poultry houses.

The mechanism of alum treatments with respect to reducing VFA production is uncertain [20], though, [26], [27] and [20], hypothesized that it was due to the pH effect of Acidifiers (acid-forming compound), which would inhibit microbial growth and activity in poultry litter. Similar findings have been observed by [28], who reported that when eugenol was added to animal manure it reduced volatile fatty acid production by 70% and 50% in cattle and swine manure, respectively. They suggested that eugenol suppressed microbial activity by lowering manure pH and inhibiting the production of volatile fatty acid that are considered the predominant odour compounds emitted from livestock wastes.

4.5 Ammonium ion (NH₄⁺) Level of Alum Treated and Untreated Wood Shavings

The ammonium ion concentration of the litters correspond to 28.32%, 21.98%, 21.98% and

23.94% of the total nitrogen content of 0%, 5%, 10% and 15% alum treated wood shavings respectively. This result is in agreement to that obtained by [20], and [29] who reported ammonium nitrogen representing 11 to 66% of the total nitrogen contents from control and all liquid AlCl₃ treatments. The significantly higher ammonium ion concentration observed in the alum treated wood shavings was due to the higher nitrogen content of the litter resulting from reduced NH₃ emission as reported by [30]. The content of NH₄⁺ and mineralizable organic nitrogen fraction (plant available nitrogen) in manure, and litter plays an important role in determining the value of animal wastes as nitrogen fertilizer [20].

4.6 Microbial Load in Alum Treated and Untreated Wood Shavings

The total bacteria load of the litter were reduced by two folds magnitude at week 8, *E. coli* and *Salmonella spp* reduced by one fold. However, [31], reported two fold magnitude reduction in the total bacteria load of the litter with alum treatment at 16 weeks. This is also similar to the work of [32], [33] and [28] who reported significant reduction in microbial load in poultry litter treated with alum. The study by [34] also reported significant reduction in total bacteria load in poultry litter treated with metam-sodium. This drastic reduction in the total bacteria, *E. coli* and *Salmonella spp.* load can be associated with the low pH in alum treated litter as reported by [33], [32] and [30]. The mold and yeast load was seen to be two fold magnitude higher in the alum treated wood shavings (5%, 10% and 15%) compared to the control (0% alum treated wood shaving). This is similar to the report by Cook *et al.* [30], who reported mold and yeast load to be 3.5 x 10⁷cfu/g and 5.5 x 10⁴cfu/g in alum treated and untreated litters respectively, indicating a threefold magnitude higher fungal load in alum treated litter compared to the untreated litter. This suggests that the addition of alum to wood shavings potentially shifts the microbial load of the litter from bacterial dominant to fungal dominant as reported by [35]. The ramifications of this shift in dominance are still unknown, and future work will be aimed at characterizing these fungi and elucidating their role in the acidified litter environment [36].

5. CONCLUSION

Treating wood shavings with alum can reduce microbial load of the litter, hence improve health

and reduce mortality. Treating wood shavings with alum tends reduce the microbial load of the litter, hence reducing the risk of most bacterial diseases in broiler chicken. And can increase total nitrogen and ammonium ion concentration of the litter and reduce pH, total volatile fatty acid and soluble reactive phosphorus content of the litter, thereby making the litter to be a better manure for crop production and reduce odour in poultry houses. The study recommends treating wood shavings with 5% inclusion level of alum.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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