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Comparison of Microbial Aspects, Ammonia Emission Rates and Properties of Broiler and Layer Litters after Application of Turmeric (*Curcuma longa*) Powder

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***Abstract*— Study it was attempted to compare the microbial dynamics and Ammonia emissions of turmeric powder (TM) applied broiler and layer litters in several applications, as an amendment. Three months old broiler and layer (2kg each) litter samples were analyzed. Initial pH and moisture was determined. Turmeric powder was used to mix with both litters at 5 levels; 0%, 1%, 3%, 5% and 8% (w/w). After mixing, 150g of samples was placed in a container for each level of the 4 replicated samples, incubated 5h and analyzed for Total Plate Count (TPC), Yeast and Mold Count (YMC), total Nematode Count (NC), pH, moisture and ammonia emission. Microbial parameters showed a significant reduction ($p<0.05$) for increment of TM. The reduction TPCs of layer litter were 42.42%, 60.6%, 95.15% and 98.18%, when 1%, 3%, 5% and 8% applications of TM. Reduction of YMC was observed >99% when treated with TM compared with $>10^8$ count of 0%. The NC reduction was 21.87%, 34.37%, 50% and 53.12%. For Broiler litter the reduction of TPC were 20.34%, 46.06%, 95.73% and 96.08%. The YMC reduction against control was 34%, 41.72%, 55% and 65.34%. The NC reduction was 22%, 45.81%, 62.5% and 70.87%. The pH of layer litter was significantly ($p<0.05$) reduced with increment of TM from 0.91, 2.19, 2.3 and 2.83%. Broiler litter also showed significant reduction of pH from 0.1, 2.75, 3.64 and 3.42%. Moisture % of layer litters were significantly ($p<0.05$) increased at 1, 5 and 8% levels from 1.38, 0.12, 3.28% and significantly decreased at 3% from 1.32% and in broiler litters the moisture % were significantly ($p<0.05$) increased along TM levels from 6.23, 0.78, 19.33 and 1.17%. Ammonia emission was significantly decreased ($p<0.05$) by increased TM for both layer (45.58, 50.35, 68.02 and 70.61%) and broiler litters (64.09, 68.31, 73.32 and 84.67%). It was concluded that the TPC, YMC, NC, pH and Ammonia emission of poultry litter (both layer and broiler) can be significantly reduced by application of 8% (w/w) of turmeric. It was more effective in reduction of TPC, YMC and NC in layer litter and pH and NH_3 in Broiler litter.**

***Index Terms*— Turmeric, Total Plate Count, Yeast and Mold Count, Nematode Count, Ammonia**

I. INTRODUCTION

Use of Turmeric (Curcuma longa) [TM] as a disinfectant was seen in many cultures especially in Asia. Several research studies suggested that turmeric can help to combat microbial activities. Aqueous turmeric extracts showed good antimicrobial activity against bacteria, fungus, viruses, yeast, and round worms. Curcumin is the main active ingredient responsible for these numerous activities. It was also noted that another active component named curcumoids which gives its yellow color. Antimicrobial activity of this multipurpose compound frequently seems to depend on the type of extract, and type of infection. Diverse studies reveal different degrees of antimicrobial activity, with essential oils and aqueous extracts of TM showing the greatest prospective. However literature offers promising information for the use of TM in the prevention and treatment of microbes. The antimicrobial activity of TM was used from thousands of years in civilizations over the world to disinfect diverse substances in various ways. Regardless of its anti-inflammatory activities, commonly used as a foodstuff, cosmetic and medicine.

Poultry litter has been shown to harbor bountiful numbers of bacteria. A byproduct of commercial chicken grow out is thousands of tons of litter. This litter is the material on which the chickens spend their entire life. The quality of the in-house environment is highly dependent upon litter quality. The litter environment is ideal for bacterial growth and ammonia production. The two factors that influence litter conditions are manure and moisture. The presence of microbes in litter may leads to contaminated processed carcasses or by increasing the microbial load of skin and feathers or by providing a source for upper gastrointestinal contamination during pre-harvest feed withdrawal both broilers and layers [3].

Jingrang *et. al.,* [10] reported that the total aerobic bacteria in poultry litter were detected by culture at 10^9 Colony Forming Units per Gram of material. Enteric bacteria as *Enterococcus* and coliforms composed 0.1 and 0.01% of the total aerobic bacteria in litter. Twelve families were identified with *lactobacilli* and *Salinococcus* spp. forming the most abundant. Total litter bacteria concentrations fall within the range of 10^{10} to 10^{11} colony forming units per gram of litter [6] [21]. Total aerobic bacteria counts are lower at 10^8 to 10^{10} [11] [12]. These numbers can differ with age of litter and age of birds [12]. Fresh litter were found to have 10^5 cfu/g, as soon as birds were placed, numbers of bacteria increased by several levels of magnitude to 10^8. The bacterial numbers maximized at 10^{10} throughout 6th week for new bedding and 4th week for litter used for more than one flock. Researchers found that after aerobic bacteria peaked at 10^{10} cfu/g of litter bacteria statistics either remained stable or degenerated until the end of every poultry flock grow-out.

Breakdown of nitrogenous compounds in litter is another mission of litter microbes. Through this breakdown, they produce Ammonia. Factors that straightly control NH_3 formation are pH, temperature and moisture level of the litter [9]. Ammonia emissions from poultry litter not only cause environment problems, but also disadvantageous to performance, health, behavior and welfare of birds. Health and welfare problems accompanying with high NH_3 concentrations include damage to the respiratory tract [15] increased vulnerability to Newcastle disease [1], incidence of air sacculitis [16] increased Mycoplasma gallisepticum [20], and incidence of keratoconjunctivitis [5]. High NH_3 concentrations in poultry houses decrease growth rate of birds [14] [18] [19] which reduces meat yield, feed efficiency which lead to smaller animals [7] and egg production [8]. Atmospheric NH_3 pollution plays a vital role in acid rains and the overriding source of NH_3 in Europe is livestock waste [2]. Consequences of high NH_3 concentrations in poultry facilities on human health are also a matter of concern [13].

Basically NH_3 in litter is generated through the microbial breakdown of undigested proteins and excretory uric acid which exists in feces of birds. Conditions that favor microbial growth will result in amplified ammonia production. These conditions include warm temperature, moisture, pH in the neutral range or slightly higher (7.0–8.5) and the presence of organic matter [13]. Ammonia emissions depend on how much of the ammonia-nitrogen in solution reacts to generate ammonia (NH_3) versus ionized ammonium (NH^{4+}), which is nonvolatile and dropping the pH in litter can reduce the NH_3 emission by directing the equilibrium between NH_3 and NH^{4+} towards the NH^{4+} ions [13].

Overall objective of this study was to explore the applicability of TM as a litter amendment to diminish microbial populations, ammonia emission rates from poultry litters by antimicrobial effects and wiping out conditions that favor microbes such as pH, moisture etc.

II. METHODOLOGY

Three months old well managed broiler and layer litters were subjected for the study. Two representative samples of both litters (±2kg) were obtained from the poultry unit of the Faculty of Agriculture, University of Ruhuna, Sri Lanka, put into black polythene bags separately, sealed and brought to laboratory. The litter samples were mixed well keeping inside the poly bags. TM powder available in the local market was used to mix with litters separately (w/w) at 5 different levels; 0%, 1%, 3%, 5% and 8% (1g TM powder /100g litter (1:99). After mixing with TM powder, 150g of mixed litter was placed in a container for each level of the replicated sample and incubated at 30^oC for 5h and analyzed for total plate count (TPC), Yeast and Mold Count (YMC), total Nematode Count (NC), litter pH [4], litter moisture and litter ammonia emission rate [14]. TPC and YMC were determined by using microbial cultures on PCA (Plate Count Agar) and PDA (Potato Dextrose Agar) medias respectively. The PCA and PDA were prepared according to standard procedure and were sterilized at 121 °C for 20 min in autoclave. The 10^{-1} dilution was prepared by mixing the 1 ml of litter sample of each treatment with 9 ml of MRD. Using separate sterile pipettes, serial decimal dilutions of 10^{-2}, 10^{-3} and 10^{-4} were prepared by transferring 1 ml of previous dilution to 9 ml of diluents. Then 15 ml of media (cooled to 45±1ºC) were poured into to each plate and the plates were allowed to solidify. Then the appropriately marked Petri dishes were inoculated with 1 ml of each dilution separately and in duplicates. Then the inoculated Petri dishes were incubated at 37 ºC for 48 hrs (TPC) and at 28 ºC for 5 days (YMC). Colonies were counted using the colony counter. Then the TPC and YMC were calculated for broiler and layer litters separately.

Nematode count was determined using Mc Master Chamber. Ground litter sample of 3g was dissolved in saturated 47ml of NaCl solution for each replicate of both litters. Then a drop of solution was observed under dissecting microscope for Nematodes. Litter NH_3 emissions were determined as described by [14], with slight modifications. The containers used to put litter samples were equipped with air inflow and outflow. Samples were incubated at 30^{o}C for 5h. Air was continuously passed through each flask, and NH_3 volatilized from litter samples in the containers were trapped in 100ml of 0.32N H_3BO_4 solutions. The trap was titrated with 0.1N HCl to determine the NH_3 emission. The emission rate was calculated as milligrams of NH_3 emitted /kilogram of fresh litter/hour. Complete randomize design was used with 4 replicates. Data were analyzed using GLM procedure of SAS (SAS Institute, 1985). Effects were considered significant when p<0.05. Means were compared using Duncan's Multiple Range Test.

III. RESULTS AND DISCISSION

Application of TM to layer litter

Effect of turmeric at different level of applications with Layer litter for TPC, YMC, NC, pH, Moisture % and NH3 was shown in Table: 1. Reduction of total bacteria in layer litter were 42.42%, 60.6%, 95.15% and 98.18% which was significant (p<0.05) along the treatments (1, 3, 5, 8%) than control (0%). A significant (p<0.05) reduction of yeast and mold count (>99%) was observed with all treatments with compared to a too numerous count (10^{8}). Nematode count was also shown a significant (p<0.05) reduction along treatments from 21.87%, 34.37%, 50% and 53.12% and the 5% and 8% levels did not show significant difference among them. Moisture % were significantly (p<0.05) increased at 1, 5 and 8% TM levels from 1.38, 0.12, 3.28% than control and significantly (p<0.05) decreased at 3% TM from 1.32%. This moisture increase of TM may be due to water holding capacity of TM and this did not affect to microbial count reduction. The pH reduction was also showed a significance (p<0.05) from 0.91, 2.19, 2.3 and 2.83% than control. The 3% and 5% levels did not show difference from each other. Total NH_3 reduction was significant (p<0.05) than control from 45.58, 50.35, 68.02 and 70.61% along treatments and the 3, 5 and 8% TM levels did not had significant (p<0.05) difference among them. Based on these observations the application of TM >3% shows significant alterations in microbial dynamics and litter parameters in layer litter.

Application of TM to broiler litter

Effect of turmeric at different level of applications with Broiler litter for TPC, YMC, NC, pH, Moisture % and NH3 was shown in Table: 2. Reduction of total bacteria in broiler litter were 20.34, 46.06, 95.73 and 96.08% which was significant (p<0.05) along the treatments than control. A significant (p<0.05) reduction of yeast and mold count was observed with all treatments from 34, 41.72, 55 and 65.34% along treatments. Total nematode significant (p<0.05) reduction was 22, 45.81, 62.5 and 70.87%.

Moisture % were significantly (p<0.05) increased along all TM levels from 6.23, 0.78, 19.33 and 1.17% than control. The pH reduction was also showed a significance (p<0.05) from 0.1, 2.75, 3.64 and 3.42% than control. The 3% and 8% levels did not show difference from each other. Total NH3 reduction was significant (p<0.05) than control from 64.09, 68.31, 73.2 and 84.67% along treatments and the 1, 3 and 5% TM levels and 5 and 8% levels did not had significant (p<0.05) difference among them. Based on these observations the application of TM >3%

shows significant alterations in microbial dynamics and litter parameters in broiler litter.

Table 1: Effect of turmeric at different level of applications with Layer litter for TPC, YMC, NC pH, Moisture % and NH_3, values with same letters are not significantly ($p<0.05$) different

Parameter	Level of Turmeric application				
	0%	1%	3%	5%	8%
pH	9.34a ±0.01	9.25b ±0.01	9.13c ±0.01	9.12c ±0.01	9.07d ±0
Moisture %	34.06d ±0	34.53b ±0.05	33.61e ±0.02	34.10c ±0.01	35.18a ±0.02
NH_3	0.59a ±0.23	0.21b ±0.16	0.29bc ±0.16	0.19c ±0.09	0.07c ±0.1
TPC	3.3×10^4a ±0	1.9×10^4b ±0	1.3×10^4 c±0	1.6×10^3d ±0	6.0×10^2e ±0
YMC	1×10^8a ±0	2.4×10^4b ±0	1.3×10^4 c±0	2.1×10^3d ±0	1.1×10^2e ±0
NEM	400a ±64	312b ±43.3	262.5c ±56.9	200d ±0	187d ±43.9

Comparison of TM applications to layer and broiler litters

Reduction of Total Plate Counts of broiler litter along with TM% were significantly lower ($p<0.05$) than the layer litter values (Broiler and Layer difference as a percentage: 42.6, 58.51, 58.06, 34.69 and 73.33% along treatments) (Table 3) which proves the higher bacterial count of broiler litter than layer litter at all treatment levels. Both litters show reducing of TPC along TM thus reduction of TPC in layer litter is significantly ($p<0.05$) higher than reduction of TPC in broiler litter. Adlibitum feeding within short life span of broilers, higher N excretion than layers and higher feed intake than layers may cause this. Considering the Yeast and Mold count, the control of layer litter had 10^8 too numerous count and it was 99.95% higher than broiler litter. Other TM levels shows significantly ($p<0.05$) higher YMC in broiler litter along TM Levels 1, 3, 5 and 8% (Broiler and Layer difference as a percentage: 19.73, 50.75, 89.7 and 99.3%) (Table 3).

Restricted feeding and increased spacing than broilers in layer cages results lower nutritive value for bacteria and may favor the growth of yeasts and mold. Reduction of YMC is significantly ($p<0.05$) higher in layer litter than broiler litter. In NEM count 0% and1% TM levels of both litters did not show significance between them (Broiler and Layer difference as a percentage: 0 and 6.2%). Other 3, 5, and 8% of TM applications showed significant reduction intensities of 17.42, 25 and 37.86% (Broiler and Layer difference as a percentage) of layer litter than broiler litter (Broiler and Layer difference as a percentage: 0, 6.2%). According to many past research findings, the volatile oil of turmeric reported for the antibacterial and antifungal activities and use of TM powder shows significant effectiveness for layer litter than the broiler litter.

Table 2: Effect of turmeric at different level of applications with Broiler litter for TPC, YMC, NC pH, Moisture % and NH_3, values with same letters are not significantly ($p<0.05$) different

Parameter	Level of Turmeric application				
	0%	1%	3%	5%	8%
pH	9.06a ±0.01	8.97b ±0.01	8.81c ±0.01	8.73d ±0.01	8.75 c±0
Moisture %	51.34e ±0	54.54b ±0.05	51.74d ±0.02	61.26a ±0.01	51.94c ±0.02
NH_3	0.68a ±0.27	0.24b ±0	0.21b ±0.01	0.18bc ±0.02	0.1c ±0.05
TPC	5.75×10^4a ±0	4.58×10^4b ±0	3.11×10^4c ±0	2.45×10^3d ±0	2.25×10^3e ±0
YMC	4.53×10^4a ±0	2.99×10^4b ±0	2.64×10^4c ±0	2.04×10^4d ±0	1.57×10^4e ±0
NEM	400a ±36.9	333.5b ±21.32	216.75c ±21.32	150d ±0	116.5e ±21.32

Table 3: Comparison of the effect of turmeric at different level of applications with Broiler (B) and Layer (L) litters for TPC, YMC, NC pH, Moisture % and NH_3, values with same letters are not significantly ($p<0.05$) different

Factor	Level of Turmeric application									
	0%		1%		3%		5%		8%	
	B	L	B	L	B	L	B	L	B	L
pH	9.06b ±0.06	9.33a ±0.01	8.97b ± 0.05	9.25a ± 0.01	8.81b ±0	9.13a ±0.01	8.73b ±0.01	9.12a ±0.01	8.75b ±0.02	9.07a ±0
Moist %	51.33a ±0.02	34.06b ±0	54.53a ±0.56	34.53b ±0.05	51.74a ±0.56	33.61b ±0.01	61.26a ±0.5	34.1b ±0.01	51.94a ±0.71	35.18b ±0.01
NH_3	0.68a ±0.27	0.59b ±0.23	0.24b ±0	0.32a ±0.16	0.21b ±0.01	0.29a ±0.16	0.18b ±0.02	0.18a ±0.09	0.1b ±0.05	0.17a ±0.1
TPC	$5.75X10^4$a ±0	$3.3X10^4$b ±0	$4.58X10^4$a ±0	$1.9X10^4$b ±0	$3.1X10^4$a ±0	$1.3X10^4$b ±0	$2.45X10^3$a ±0	$1.6X10^3$b ±0	$2.25X10^3$a ±0	$6X10^2$b ±0
YMC	4.53×10^4b ±0	1×10^8a±0	2.99×10^4a ±0	2.4×10^4b ±0	2.64×10^4a ±0	1.3×10^4b ±0	2.04×10^4a ±0	2.1×10^3b ±0	1.57×10^4a ±0	1.1×10^2b ±0
NEM	400a ±36.9	400a ±64	333.5a ±21.32	312.5a ±43.3	262.5a ±56.9	216.7b ±21.32	200a ±0	150b ±0	187.5a ±43.3	116.5b ±21.32

The pH reduction of layer litter against broiler litter was significantly ($p<0.05$) low (Layer and Broiler difference as a percentage 2.94, 3.02, 3.5, 4.27 and 3.52%) which means application of turmeric powder was more effective at broiler litter samples significantly ($p<0.05$) than layer litter samples. Moisture increase in broiler litters than layer litters also significantly ($p<0.05$) high at TM application (Broiler and Layer difference as %: 33.65, 36.68, 35.04, 44.33 and 32.26%). At 0% application of TM the NH_3 emission was significantly ($p<0.05$) higher in broiler litter than layer litter (Broiler and Layer difference as %: 13.61%) thus increment of TM % (1, 3, 5 and 8%) reduced the emission in both litters and more significantly ($p<0.05$) in broiler litter than layer litter (Layer and Broiler difference as % 23.62, 26.1, 3.42 and 39.63%). Emission of NH_3 from broiler litter is three times higher than that of layer litter [22]. Due to this lower emission from this layer litter samples we can suggest that the application of turmeric powder to broiler type litters is more effective to reduce NH_3 emissions from large scale broiler houses. Further researches in this topic is much needed to apply this technique in large scale operations thus to reduce the environmental pollution and global warming.

IV. CONCLUSION

Application of Turmeric powder as a litter amendment was effective in significant reduction of TPC, YMC, NC, pH and NH3 emission in both broiler and layer litters on more than 3% (w/w) application level. Furthermore TM application was more effective in reducing of TPC, YMC, NC in layer litter and reducing of pH and NH3 emission in broiler litter. Increment of moisture percentage was seen in both litters after application of TM but it did not affect the other parameter enhancements.

REFERENCES

[1] Anderson D.P, Beard C.W and Hanson R.P, 1964, The Adverse Effects of Ammonia on Chickens Including Resistance to Infection with Newcastle Disease Virus. “Avian Diseases”, Vol. 8, No. 3 (Aug), pp. 369-379

[2] Ap Simon H.M, Kruse M, and Bell J.N.P, 1987, Ammonia emissions and their role in deposition, Atmospheric Environment 21:1939-1946

[3] Bennett D.D, Higgins S.E, Moore R.W, Beltran R, Caldwell D.J, Byrd J.A and Hargis B.M, 2003, Effects of lime on Salmonella enteritidis survival in vitro. L. Appl. Polt. Res. 12: 65-68.

[4] Brake J.D, Boyle C.R, Chamblee T.N, Schultz C.D and Peebles E.D, 1992, Evaluation of the chemical and physical properties of hardwood bark used as a broiler litter material. Poult.Sci. 71:467-472.

[5] Bullis K.L, Snoeyenbos G.H, and Van Roekel H, 1950, A kerato-conjunctivitis in chickens. Poult. Sci. 29:386–399.

[6] Carr L.E, Wheaton F.W and Douglass L.W, 1990, Empirical models to determine ammonia concentrations from broiler chicken litter. Trans. ASAE 33:1337-1342.

[7] Caveny D.D, Quarles C.L and Greathouse G.A, 1981, Atmospheric ammonia and broiler cockerel performance. Poult. Sci. 60:513-516.

[8] Deaton I.W, Reece F.N and Lott B.D, 1984, Effect of atmospheric ammonia on pullets at point of lay. Poult. Sci. 63:384-385.

[9] Elliott H.A, and Collins N.E, 1982, Factors affecting ammonia release in broiler litter. Trans. ASAE 25:p.413-424.

[10] Jingrang L, Sanchez S, Hofacre C, Maurer J.J, Harmon B.G and Lee1 M.D, Evaluation of Broiler Litter with Reference to the Microbial Composition as Assessed by Using 16S rRNA and Functional Gene Markers, Applied and Environmental Microbiology, 2003, American Society for Microbiology, Vol. 69, (Feb) p. 901–908

[11] Lu J, Shanchez S, Hofacre C, Maurer J.J, Harmon B.G and Lee M.D, 2003. Evaluation of broiler litter with reference to the microbial composition as assessed by using 16s rRNA and functional gene markers. Appl. Environ. Microb. 69(2):901-908.

[12] Macklin K.S, Hess J.B, Bilgili S.F and Norton R.A, 2005, Bacterial levels of pine shavings and sand used as poultry litter. J. Appl. Poult.

[13] Miles D, Branton S.L, and Lott B.D, 2004. Atmospheric ammonia is detrimental to the performance of modern commercial broilers. Poult. Sci. 83:1650–1654.

[14] Moore Jr. P.A, Daniel T.C, and Edwards D.R, 1999, Reducing phosphorus runoff and improving poultry production with alum. Poult. Sci. 78:692–698.

[15] Nagaraja K.V, Emercy D.A, Jordan K.A, Sivanandan V, Newman J.A, and Pomeroy B.S, 1983, scanning electron microscopic studies of adverse effects of ammonia on tracheal tissues of turkeys. Am. J. Vet. Res. 44:1530–1536.

[16] Oyetunde O.O.F, Thompson R.G, and Carlson H.C, 1976, Aerosol exposure of ammonia, dust and Escherichia coli in broiler chickens. Can. Vet. J. 19:187–193.

[17] Pokharel B.B, 2010, Ammonia emission from poultry industry, its effect and mitigation mechanism, International Veterinary Students Association's Newsletter (December 2010) http://ivsa.org/2011/01/ivsa-newsletter December 2010, accessed on 29 June 2015

[18] Reece F.N, Bates B.J, and Lott B.D, 1979, Ammonia control in broiler houses. Poult. Sci. 58:754–760.

[19] Reece F.N, Lott B.D, and Deaton J.W, 1981, Low concentrations of ammonia during brooding decrease broiler weight. Poult. Sci. 60:937–940.

[20] Sato S, Shaya S, and Kobayashi H, 1973, Effect of ammonia on Mycoplasma gallisepticum infection in chickens. Natl. Inst. Anim. Health Q. (Yatabe) 13:45–53.

[21] Schefferle H.E, 1965. The microbiology of built up poultry litter. J. Appl. Bacteriol. 28(3):403-411

[22] Vitharana J, Atapattu N.S.B.M, Abeywickrama L.M, Nilantha De Silva, 2012, Estimation of Ammonia emission from broiler and layer litters under hot humid small farming conditions in Sri Lanka, The potential health and environmental impacts of exposure to hazardous Natural and manmade chemicals and their proper management, available at http://www.ejustice.lk/ Res. 14:238-245