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Performance, Carcass Yield and Gut Microflora of Broiler Chickens Offered with *Viola odorata* Leaf Extract in Drinking Water

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Authors' contributions

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

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ABSTRACT

The cross resistance of antibiotics from birds to human advocated using of herb extract.120 day old broiler chickens of Arbor Acres strain were used for the study. Birds were randomly allotted into four experimental treatments with three replicate of ten birds per replicate. T1 control without antibiotics, T2 with antibiotics (Amoxycol® wsp containing Amoxycillin 200 mg + collistin sulphate 1,000,000iu) at 1 g/litre of drinking water, T3 contain 10% *Viola odorata* extracts offered at 1 ml extract/1 litre of drinking water, while T4 contain 20% *Viola odorata* extracts offered at 1 ml extract/1 litre of drinking water. The birds were exposed to the same environment, feeds and water were given *ad libitum* and other routine management practices were carried out. Significant

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differences (P>0.05) existed in the daily feed intake, total feed intake and feed conversion ratio. Birds on 20% extracts recorded the highest feed intake while birds on T3 10% extract consumed the least. Broiler chickens on 10% extracts recorded best feed conversion ratio. Increased feed intake and digestive secretions were observed in animals offered phytobiotic-supplemented feed. The gut microflora showed significant difference (P>0.05) existed between the population of *Proteus mirabilis*, *Staphylococcus aureus* and total anaerobic bacteria counts. It was observed that *Proteus mirabilis* colony forming unit in broiler chickens fed diets containing antibiotics and 20% *Viola odorata* were 2.00 $\log_{10}$ of the organisms respectively. The result deduced in this study showed that the inclusions of the leaf extracts at either 10% or 20% concentration has no detrimental effect to broiler chickens.

Keywords: Proteus mirabilis; Staphylococcus aureus; Viola odorata.

1. INTRODUCTION

Antibiotics have been used widely to prevent infections and poultry diseases and for the improvement of meat and egg production. However, use of antibiotics is restricted due to drug resistance in bacteria, drug residue in carcass and also alteration of natural gut micro flora [1]. Recently many countries tend to minimize or prohibit the use of antibiotics because of their deleterious side effects on both animals and human. Consequently, the of use of natural promoters such as probiotics, prebiotics, symbiotics, enzymes, toxic binders, organic acids, oligosaccharides, phytogenics, and other feed additives, to enhance the growth and performance of broiler chickens have been advocated [2]. These alternatives to antibiotics have been used to improve the production parameters of fattening pigs and poultry [3].

Gut micro flora has significant effect on host nutrition, health and growth performance [4] by interacting with nutrient utilization and the development of the gut system of the host. The interaction is very complex and, depending on the composition and activity of the gut micro flora, it can be either positive or negative effects on the health and growth of birds. For instance, when pathogens attach to the mucosa, gut integrity and function is affected [5] and immune system is threatened [6]. The health of organisms largely depends on the composition of the intestinal micro flora which can be significantly supported by probiotics, prebiotics, organic acid, zinc oxide and plant extracts. Feed additives produced from plants have often a significant antibacterial effect, thereby suppressing pathogenic micro flora in the gastrointestinal tract of animals and thus reducing mortality during the fattening period, especially in stress period [7]. Furthermore, it is generally agreed that gut microflora is a nutritional "burden" in fast-growing broiler chickens [8,9] since an active microflora component may have an increased energy requirement for maintenance and a reduced efficiency of nutrient utilization. The focus of alternative strategies has been used to prevent proliferation of pathogenic bacteria and modulation of indigenous bacteria so that the health, immune status and performance are improved [10].

Viola odorata commonly known as 'sweet violet' or 'garden violet' is a perennial ornamental herb, native to Europe, Asia, North America and Australia [11]. In indo-Pakistan, it is called 'banafsha' [12]. It has been used as medicinal herb for the relief of pain due to cancer [13]. It has also been used for chronic bronchial asthma, symptoms of upper respiratory track, headache and insomnia [14]. The aqueous plant extract has been shown to possess anti-bacteria activity [15]. The methanolic extract of *Viola odorata* leaf was found to contain a variety of phenolic (35.4 mg/g) and flavonoid (22.8 mg/g) compounds [16]. Various phytochemical constituents (alkaloids, steroids, tannins, flavonoids, and saponins) have been reported in the butanolic, methanolic and aqueous extracts, including aerial parts of *Viola odorata* [17]. This research work was designed to evaluate the effects of *Viola odorata* on performance, carcass yield and gut microflora of broiler chickens.

2. MATERIALS AND METHODS

2.1 Preparation of Test Ingredient

Viola odorata leaves were collected from Yaba College of Technology farm, Yaba Lagos, Nigeria. The leaves were dried in the sun for 5 days. The dried leaves were pulverized with a blender; 0.2 mm mesh diameter sieve was used to obtain the fine powder. Ten gram leaf powder

was added to 80 ml of distilled water, it was shaken vigorously and allowed to stay overnight at room temperature, filtered and distilled water was added up to 100 ml to make 10% extract. To obtain 20% extract, twenty gram leaf powder was added to 80 ml of distilled water, it was shaken vigorously and allowed to stay overnight at room temperature, filtered and distilled water was added up to 100ml to make 20% extract [18].

2.2 Animals, Diets and Experimental Design

120 day old broiler chickens of Arbor Acres strain were used for the study. The birds were randomly allotted into four experimental treatments with three replicate of ten birds per replicate in deep litter system. T1 (control) group without antibiotics, T2 with antibiotics (Amoxycol® wsp containing Amoxycillin 200 mg + sulphate 1,000,000 iu) at 1 g/litre of drinking water, T3 contain 10% *Viola odorata* extracts offered at 1 ml extract/1 litre of drinking water, while T4 contain 20% *Viola odorata* extracts offered at 1 ml extract/1litre of drinking water. The birds were exposed to the same environment, feeds and water were given *ad libitum* and other routine management practices were carried out. The experimental diets Table 1 were formulated to meet the nutritional recommendations [19].

2.3 Growth Performance

Daily feed intake and weekly weight gain were monitored and recorded. Records of daily mortality were monitored in all phases of the experiments. Feed conversion ratio (FCR) was computed on a weekly basis in all the phases of the study.

2.4 Carcass Yield Evaluation

At the expiration of the experiment (finisher phase), two birds per replicate whose weight is similar or close to the average weight of the birds contained in each replicate was selected for slaughter and designated for carcass yield analysis. Birds selected was fasted overnight and slaughtered by neck cutting, plucked and eviscerated. The live weight, plucked weight and eviscerated weight, weight of cut parts were recorded. Evisceration of the carcass was done manually following standard commercial procedures [20]. Weights of the cut parts was determined and expressed as a percentage of the live weight gain.

Table 1. Percentage composition of experimental diets

Ingredients	Starter (0-28 days)	Finisher (29-56 days)
Maize	54.00	58.00
Soybean meal	30.00	25.00
Vegetable oil	1.00	2.00
MOLM	0.00	0.00
Wheat offal	6.00	7.00
Fish (72% CP)	3.00	2.00
Bone meal	3.00	2.00
Limestone	2.00	3.00
Salt	0.25	0.30
Premix	0.30	0.30
Methionine	0.20	0.20
Lysine	0.25	0.20
Enzyme	-	-
Total	**100**	**100**
Determined analyses (%)		
Crude protein	22.43	20.33
Crude Fibre	2.05	4.08
Ether Extract	3.85	3.91
Calculated analyses (%)		
Calcium	1.78	1.77
Phosphorus	0.46	0.44
Lysine	1.49	1.31
Methionine	0.56	0.54
ME(Kcal/kg)	2832	3100

**Starter premix: - Vit. A 10,000,000 (iu), Vit D_3 2,000,000 (iu), Vit. E 23,000(mg), Vit K_3(mg), Vit B_1 1,800 (mg), Vit. B_2 5,500 (mg), Niacin 27,500 mg, Pantothenic acid 7,500 mg, Vit. B_6 3,000 mg, Vit.B_{12} 15 mg, Folic acid 750 mg, Biotin H2 60 mg, Chlorine chloride 300,000 mg, Cobalt 200 mg, Copper 3,000 mg, Iodine 1,000 mg, Iron 20,000mg, Manganese 40,000(mg), Selenium 200 mg, Zinc 30,000 mg, Anti-oxidant 1,250 mg*

2.5 Evaluation of Gut Micro Flora

At the end of (56 days) feeding trial, two birds per replicate were slaughtered and gut content from ileum, starting from the Meckel's diverticulum to 4 cm above the ileo-caecal junction, was dissected and the digesta contents of this intestinal segment (1 g) were collected into sample bottles and homogenized with 9ml of sterile normal saline. The ileal digesta specimens were sent packed on ice pack to the laboratory (Microbiological laboratory, College of Veterinary Medicine, Federal University of Agriculture,

Abeokuta) for enumeration of total bacteria, *E. coli*, *Pseudomonas fluorescence*, *Salmonella specie*, *Klebsiella spp.*, Clostridium, *Lactobacillus spp.*, *Staphylococcus saprophyticus*, *Bacillus subtilis*, *Proteus mirabilis*, *Citrobacter spp.*, *Streptococcus spp* and *Staphylococcus aureus* as described by [21].

2.6 Statistical Analysis

All data generated were subjected to one-way Analysis of variance (ANOVA) in a completely randomized design; data was analyzed using SAS statistical package [22]. While significant differences among treatment means were separated using Duncan Multiple Range Test [23].

3. RESULTS AND DISCUSSION

3.1 Performance

Tables 2 and 3 shows the performance of broiler chickens fed *Viola odorata* as phytogenic plants at starter phase (0-4 weeks). Significant differences (P<0.05) existed in the daily feed intake, total feed intake and feed conversion ratio. Birds on 20% extracts recorded the highest feed intake while birds on T3 10% extract consumed the least. Broiler chickens on 10% extracts recorded best feed conversion ratio. Increased feed intake and digestive secretions were observed in animals offered phytobiotic-supplemented feed [24]. Growth enhancement through the use of phytobiotics is probably the result of the synergistic effects among complex active molecules existing in phytobiotics [25]. These findings revealed that inclusion of *Viola odorata* in drinking water at either 10% or 20% extract led to increase in weight gained when compared to broiler chicken fed diet containing antibiotics. This showed that *Viola odorata* as phytogenic plant could replace the use of antibiotics in broiler chickens. The result of the present study agreed with the findings of Olobatoke and Oloniruha [26] who reported improved growth performance and better feed conversion ratio for broiler chickens fed diet containing sweet basil meal (*Ocimum basilucum*), Also Oleforuh-Okoleh et al. [27] reported that inclusion of bitter leaf powder in cockerels significantly improved FCR. In a similar study [28], broiler chicken given bitter leaf aqueous extract had better final body weight and weight gain. Inclusion of neem and tulsi was also reported to promote growth and feed efficiency in broiler due to its antibacterial properties [29].

3.2 Carcass Yield

Table 4 shows the effect of oral administration of *Viola odorata* extract on dressed weight, dressing percentage and cut parts. It was observed that inclusion of *Viola odorata* significantly (p<0.05) improved the dressed weight, back and thigh weights of the broiler chickens. The higher body weight gains might be due to growth promoting and antimicrobial properties of *Viola odorata* leaves that helped to reduce the pathogenic microbial load, improve health status and improved feed efficiency of the birds [15]. Dressing percentage of birds fed *Viola odorata* leaves extract were not different from those on the control, though numerical increase in the dressing percentage was observed. This is agreement with the previous report [30] in broiler chickens fed *Azadirachta indica* and *Rauvolfia vomitoria* decoctions. The results of the present study indicated that *Viola odorata* improved significantly the carcass yield. This might be due to the improvement in the digestibility of feed ingredient by the inclusion of herbal supplements [31]. The present findings supported the concept that plant extracts improved the carcass yield of broiler chicken as reported by [32].

3.3 Gut Microflora

The gut microflora is involved in a wide array of physiological, nutritional, and immunological events, which can directly, or indirectly, affect the health and productivity of commercial flocks. The result in Table 5 showed the effect of *Viola odorata* on gut microflora of broiler chickens. From the data collected, significant difference (P<0.05) existed between the population of *Proteus mirabilis*, *Staphylococcus aureus* and total anaerobic bacteria counts. It was observed that *Proteus mirabilis* colony forming unit in broiler chickens fed diets containing antibiotics and 20% *Viola odorata* were 2.00 $\log_{10}$ of the organisms respectively. Several studies have reported effects on intestinal microflora when herbs and essential oils have been included in broiler diets. The dietary supplementation of extracts, an encapsulated product containing capsaicin, carvacrol and cinnamaldehyde reduced the numbers of *E. coli* in broiler rectal contents to the same extent as avilamycin [31]. The antibacterial, anti-fungal and anticoccidial effects of essential or other components from plant extracts may be due to the lipophilic property and chemical structure [33-35]. It was observed that at a higher concentration, there is increase in

Lactobacillus population which confirmed the report of [36] that there is a growing tendency of Lactobacillus species after adding phytogenic additives. The observed trends is similar to Mountzouris, 2014 and Khan 2014 [37,38] result who reported *lactobacilli* increased to some extent, but enterobacteriaceae, including the detection rate of *Proteus*, showed a significant decrease. It seems that the activities of the intestinal flora may lead to lower pH and a decrease in caecal putrefactive bacteria such as *Proteus*. The present study revealed that *Viola odorata* extract has inhibitory effect on most gram negative bacteria such as *Salmonella specie*, *Klebsiella spp*, *Escherichia coli*, *Staphylococcus saprophyticus*.

Table 2. Performance of broiler chickens offered *Viola odorata* extract (0-4 weeks)

Parameters(g)	T1	T2	T3	T4	SEM
Initial weight	44.52	44.37	44.50	44.41	0.04
Final weight	811.31	883.33	825.00	870.24	28.56
Weight gained	766.79	838.97	780.50	825.83	28.55
Daily feed intake	51.07^{c}	52.21^{b}	50.73^{c}	55.52^{a}	0.58
Total feed intake	1436.75^{c}	1465.80^{b}	1416.44^{d}	1562.12^{a}	16.84
FCR (%)	2.29^{a}	1.94^{b}	2.01^{b}	2.30^{a}	0.05

a, b, c, d= Means in the same row having different superscript are significantly different (P<0.05)
T1 (control) group without antibiotics, T2 with antibiotics (Amoxycol® wsp containing Amoxycillin 200 mg + sulphate 1,000,000 iu) at 1g/litre of drinking water, T3 contain 10% Viola odorata extracts offered at 1 ml extract/1litre of drinking water, while T4 contain 20% Viola odorata extracts offered at 1ml extract/1litre of drinking water

Table 3. Performance of broiler chickens offered *Viola odorata* extract (4-8 weeks)

Parameters(g)	T1	T2	T3	T4	SEM
Initial weight	811.31	883.33	825.00	870.24	28.36
Final weight	2132.10	2289.30	2258.30	2304.80	38.60
Weight gained	1320.83^{b}	1405.95^{ab}	1433.33^{a}	1434.52^{a}	18.03
Daily feed intake	131.28	132.41	130.96	136.93	1.22
Total feed intake	3675.89	3707.50	3668.07	3833.92	34.27
FCR (%)	2.79	2.64	2.56	2.67	0.04

a, b= Means in the same row having different superscript are significantly different (P<0.05)
T1 (control) group without antibiotics, T2 with antibiotics (Amoxycol® wsp containing Amoxycillin 200 mg + sulphate 1,000,000 iu) at 1 g/litre of drinking water, T3 contain 10% Viola odorata extracts offered at 1 ml extract/1 litre of drinking water, while T4 contain 20% Viola odorata extracts offered at 1ml extract/1litre of drinking water

Table 4. Carcass yield of broiler chickens offered *Viola odorata* extract

Parameters(g)	T1	T2	T3	T4	SEM
Live weight	2150.00	2250.00	2258.33	2300.00	26.06
Plucked weight	2000.00	2083.33	2116.67	2085.00	30.26
Dressed weight	1483.33^{b}	1591.67^{ab}	1600.00^{ab}	1663.33^{a}	28.06
Dressing %	68.81	70.77	70.79	72.32	0.64
Drumstick	11.13	10.65	11.45	10.69	0.19
Back	16.00^{b}	15.40^{b}	17.20^{ab}	18.31^{a}	0.41
Thigh	10.62^{a}	12.66^{a}	12.14^{a}	11.39^{ab}	0.25
Wing	9.66	9.89	10.14	9.09	0.21
Breast	20.66	21.53	18.37	20.27	0.56

a,b,c,d= Means in the same row having different superscript are significantly different (P<0.05)
T1 (control) group without antibiotics, T2 with antibiotics (Amoxycol® wsp containing Amoxycillin 200 mg + sulphate 1,000,000 iu) at 1 g/litre of drinking water, T3 contain 10% Viola odorata extracts offered at 1 ml extract/1 litre of drinking water, while T4 contain 20% Viola odorata extracts offered at 1 ml extract/1 litre of drinking water

Table 5. Gut micro flora of broiler chickens offered *Viola odorata* extract

Parameters (log_{10} of the organism)	Control	Antibiotics	10% extract	20% extract	SEM
Total bacteria count	2.93	3.10	3.28	3.23	0.19
Total anaerobic count	2.54^{b}	2.85^{ab}	3.06^{ab}	3.19^{a}	0.18
E. coli,	2.30	2.74	2.95	2.90	0.12
Pseudomonas fluorescence,	2.00	2.30	2.30	2.30	0.06
Salmonella specie,	1.70	0.00	2.30	2.30	0.04
Klebsiella spp.,	2.70	2.30	2.40	2.40	0.00
Clostridium	2.30^{c}	2.30^{c}	2.60^{b}	2.98^{a}	0.15
Lactobacillus spp	2.00	2.48	2.00	2.30	0.04
Staphylococcus saprophyticus	1.70	2.18	1.70	1.70	0.04
Bacillus subtilis	0.00	2.00	2.48	2.30	0.06
Proteus mirabilis,	0.00b	2.00^{a}	2.40^{a}	2.00^{a}	0.04
Citrobacter spp.,	2.00	0.00	1.70	1.70	0.02
Streptococcus spp.,	1.70	2.40	1.70	2.00	0.04
Staphylococcus aureus.	1.70b	0.00^{c}	1.70^{b}	2.00^{a}	0.04

a,b,c,d= Means in the same row having different superscript are significantly different (P<0.05)

4. CONCLUSION

The result of this study shows that phytogenic plant has the potential of improving the growth performance and the intestinal health of broiler chickens. And also, the inclusion of the leaf extracts at either 10% or 20% concentration has no detrimental effect to broiler chickens.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Botsoglou NA, Florou-Paneri E, Christaki E, Fletouris DJ, Spais BA. Effect of dietary oregano essential oil on performance of chickens and on iron-induced lipid oxidation of breast, thigh and abdominal fat tissues. Brit. Poult. Sci. 2002;43:223-230.
2. Borazjanizadeh M, Eslami M, Bojarpour M, Chaji M, Fayazi J. The effect of clove and oregano on economic value of broiler chickens diet under hot weather of Khuzesta. J. of Anim. Vet. Adv. 2011; 10(2):169-173.
3. Windisch W, Schedle K, Plitzner C, Kroismayr A. Use of phytogenic products as feed additives for swine and poultry. J. of Anim. Sci. 2008;86:140-8.
4. Barrow PA. Probiotics for chicken, in: R. Fuller (ED). Probiotics: "The scientific basis". 1992;255- 257 (Chapman and Hall, London).
5. Droleskey RE, Oyofo BA, Hargis BM, Corrier DE, Deloach JR. Effect of mannose on *Salmonella typhimurium*-mediated loss of mucosal epithelial integrity in cultured chick intestinal segments. Avian Diseases. 1994;38:275-281.
6. Neish AS. The gut micro flora and intestinal epithelial cells: A continuing dialogue. Micro. and Infect. 2002;4:309-317.
7. Schone F, Vetter A, Hartung H. Effects of essential oils from fennel *Foenicul aetheroleum* and caraway *Carvi aetheroleum* in pigs. J. of Anim. Physio. and Anim. Nutr. 2006;90:500-10.
8. Dibner JJ, Richards JD. Antibiotic growth promoters in agriculture: History and mode of action. Poult. Sci. 2005;84:634-643.
9. Lan Y, Verstegen MW, Tamminga S, Williams BA. The role of the commensal gut microbial community in broiler chickens. W. Poult. Sci. J. 2005;61:95-104.
10. Ravindran V. Broiler nutrition in New Zealand- challenges and strategies; 2006. Available:www.feedinfo.com
11. Jayaweera DM. Medicinal plants, Part 3. "Nat. Sci. Cou" of Sri Lanka publication Colombo. 1981;100.
12. Khan RU, Rahman ZU, Javed I, Muhammad F. Effect of vitamins, protein level and probiotics immune response of moulted male broiler breeders. J. of Anim. Physil and Anim. Nutrition; 2014. DOI: 10.1111/jpn.12087

13. Usmanghani K, Saeed A, Alam MT. Indusyuni medicine. Karachi University press, *Viola odorata* linn. 1997;440-441.
14. Kapoor LD. Handbook of "Ayurvedic Medicinal Plants". Boca Raton. CRC Press *Viola odorata.* 1990;335.
15. Maryam A, Farahnaz KS, Mohammad MC, Faraz MV, Mozaffariane HZ. Introduction of medicinal plants species with the most traditional usage in Alamut Region. Iran. J. of Pharma. Res. 2012;11(1):185-194.
16. Daljit SA, Gurinder JK. Antibacterial activity of some Indian medicinal plants. J. of Nat. Med. 2007;61:313-317.
17. Ebrahimzadeh MA, Nabavi SM, Nabavi SF, Bahramian F, Bekhradnia AR. Antioxidant and free radical scavenging activity of *H. officinalis* L. var. angustifolius, *V. odorata*, *B. hyrcana* and *C. speciosum*. Pak. J. of Pharmacol. Sci. 2010;23:29–34.
18. Vishal A, Parveen K, Pooja S, Kannappan N, Kumar S. Diuretic, laxative and toxicity studies of *Viola odorata* aerial parts. Pharma. Online. 2009;1:739–748.
19. Mollah MR, Rahman MM, Akter F, Mostofa M. Effects of nishyinda, lack pepper and Cinamon extract as growth promoter in broiler. T. Bangl. Vet. 2012;29(2):69-77.
20. NRC, Nutrient requirement of domestic animals, "Nutrient requirement of poultry" 9th edition. National Academy press, Washington; 1994.
21. Jansen WM, Carre B. Influence of fibre on digestibility of poultry feed. In recent Advances in Animal Nutrition. Ed. Haresign, W and Cole, D.J.A. Butterworth, London. 1985;50.
22. Oso AO, Sobayo RA, Jegede AV, Fafiolu AO, Iyasere OS, Dele P, Bamgbose AM, Cecilia A. Effect of dietary inclusion of sorghum milling waste on growth response, nutrient utilization, gut characteristics and caecal microflora of weaner rabbits. Anim. Sc. J. 2011;82:468-474.
23. SAS, Statistical Analysis Systems user's guide: Version 8.02 Edition. SAS Institute, Inc., Cary. N.C., USA; 2001.
24. Duncan DB. Multiple range and multiple F tests. Biometrics. 1955;11:1-42.
25. Windisch W, Kroismayr A. The effects of phytobiotics on performance and gut function in monogastrics; 2006.
Available:www.feedinfo.com
26. Gauthier R. (XVIII Congreso Lantioamericano de Avicultura) Poultry therapeutics: New alternatives; 2002.
Available:http://www.jefo.ca/pdf/ALA2003_en.pdf
27. Abbas RJ. Effect of using fenugreek, parsley and sweet basil seeds as feed additives on the performance of broiler chickens. Int. J. of Poult. Sci. 2010;9(3): 278-282.
28. Olobatoke RY, Oloniruha JA. Haematological assessment of bitter leaf *Vergonia amygdalina* efficacy in reducing infections in cockerels. Proc. of World cong. on Med. and Aro. Plt. Cape Town, South Africa; 2008.
29. Oleforuh-Okoleh VU, Olorunleke SO, Obi IC. Evaluation of broiler Chicks fed bitter leaf *Vernonia amygdalina* aqueous extracts. Proc. of 40th Ann. Conf. of Nig. Soc. for Anim. Prod. (NSAP). NAPRI/ABU, Zaria; 2015.
30. Prasannabalaji N, Muralitharan G, Sivanandan RN, Kumaran S, Pugazhvendan SR. Antibacterial activities of some Indian traditional plant extracts. Asian Pac. J. of Trop. Dis. 2012;2:S291-S295.
31. Zanu HK, Kagya-Agyemang JK, Kwenin WKJ, Bonsu FRK, Antwi E, Ateni S. Physiological response of broiler chickens to NOeem and akakapenpen decoctions: Performance and carcass characteristics. Int. J. Poult. Sci. 2011;10(9):730-733.
32. Jamroz D, Orda J, Kamel C, Wiliczkiewicz A, Wertelecki T, Skorupinska J. The influence of phytogenic extracts on performance, nutrient digestibility, carcass characteristics, and gut microbial status in broiler chickens. J. of Anim. Feed Sci. 2003;12:583-596.
33. Alcicek A, Bozkurt M, Çabuk M. The effect of a mixture of herbal essential oils, an organic acid or a probiotic on broiler performance. S. Afri. Soc. for Anim. Sc. 2004;34(4):217-222.
34. Elgayyar M, Draughon F, Golden D, Mount J. Antimicrobial activity of essential oils from plants against selected pathogenic and saprophytic microorganisms. J. of Food Prot. 2001,64:1019-1024.
35. Guo F, Williams B, Kwakkel R, Li H, Li X, Luo J, Li W, Verstegen M. Effects of mushroom and herb polysaccharides, as alternatives for an antibiotic, on the cecal microbial ecosystem in broiler chickens. Poult. Sci. 2004;83:175-182.

36. Willis W, Isikhuemhen O, Ibrahim S. Performance assessment of broiler chickens given mushroom extract alone or in combination with probiotics. Poult. Sci. 2007;86:1856-1860.
37. Mountzouris KC, Paraskevas V, Tsirtsikos P, Palamidi I, Steiner T, Schatzmayr G, Fegeros K. Assessment of phytogenic additives on performance, morphology, nutrient digestibility and ceacal microflora composition. Anim. Feed Sci. Tech. 2011; 168(3):223-231.
38. Khan SH. 'The use of green tea (*Camellia sinensis*) as a phytogenic substance in poultry diets' Onde. J. of Vet. Res. 2014,81(1):706-8. Available:http://dx.doi.org/10.4102/ojvr.v81i2.706

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