Effects of Papaya Leaf (Carica papaya) and Black Cumin (Nigella sativa) as an Alternative to Antibiotics on Production Index and Hematological Indicators of Broiler Chicken

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ABSTRACT

The purpose of the study was to determine the effectiveness of dietary supplements of papaya leaf meal (Carica papaya) and black cumin (Nigella sativa) seeds on the production index and hematological status of commercial broiler chicken. T1 (Control), T2 (antibiotic), T3 (2% PLM: Papaya Leaf Meal), T4 (1% BCS: Black Cumin Seed) and T5 (1% of each PLM & BCS) were the five treatment groups that the 150day-old Cobb 500 straight run chicks were randomly assigned. Ten chicks were used in each of the three replications of each treatment. The findings demonstrated that feeding broilers supplements of PLM and BCS did not significantly (p>0.05) affect feed consumption (FC) (g), final live weight (g) and feed conversion ratio (FCR) when compared to the control and antibiotic group. FC was found to be numerically (p>0.05) higher in the control group than in the other groups. However, the T3 group of birds given 2% PLM had superior ultimate live weight and an improved FCR value compared to antibiotic, control, and the values of the other groups, which were statistically insignificant (p>0.05). The relative weight of the spleen in various groups was unaffected (p>0.05) by dietary supplementation with PLM and BCS. But the T3 (2% PLM) group had significantly (p<0.05) increased bursa weight (g). The concentrations of glucose and cholesterol were similar across all groups (p>0.05), while the group receiving 2% PLM supplements had relatively lower cholesterol levels (mg/dl). Moreover, the supplemented groups with PLM and BCS revealed a significant (p<0.05) increase in the hematological parameters red blood cell (RBC), white blood cell (WBC), lymphocyte, and packed cell volume (PCV) as compared to the antibiotic and control groups. The final live weight, FCR, immune organ weight, and blood indices were all improved in birds fed a meal supplemented with 2% PLM, leading to superior results.

Keywords: Production performance, Hematological parameters, papaya leaf meal, black cumin

INTRODUCTION

In Bangladesh, poultry production stands as one of the most significant sub-sector of the livestock industry. Despite earlier constraints, the industry managed to supply 36% of the nation's meat needs in 2019, according to the USDA. Around 1.5 to 1.6 percent of Bangladesh's GDP is currently produced by the poultry industry, according to the Bangladesh Poultry Industry Central Council (BPICC). Furthermore, the industry directly and indirectly supports almost 6 million new employment opportunities nationwide (Karmoker, 2022). Throughout the past few decades, the majority of nations have used a variety of antimicrobials in the production of chickens (Boamah et al., 2016). According to scientific evidence, the uncontrolled widespread use of antibiotics has contributed to an increase in the problem of antibiotic resistance (Furtula et al., 2010; Forgetta et al., 2012), which leads to the spread of resistant microbes and the presence of antibiotic residues in feed and the environment (Carvalho and Santos, 2016; Ronquillo and Hernandez, 2017). Because of this, using herbs and medicinal

plants as a potential substitute for antibiotic growth promoters is possible (AGPs). Herbs include a complex blend of organic compounds with immune-stimulating, antibacterial, antipyretic, and anti-inflammatory activities (Guo et al., 2003). In the current investigation, papaya leaves and black cumin seeds were utilized in place of antibiotics. Papaya is a medicinal plant contains a number of phytochemicals, including lycopene and polyphenols (Kale et al., 2003). The proteolytic enzymes papain and chymopapain, which have the ability to degrade protein, are abundant in papaya leaves and helpful in preventing digestive issues and intestinal worms (Burkill, 1985). Moreover, papaya leaves contain carotene, a provitamin A that can provide around 18 – 50 IU and is a promising natural source of xanthophyl. Iron, calcium, phosphorus, vitamin C, and vitamin E are all found in papaya leaves. In addition, the leaves have a crude protein content of 20.88%, 0.99% calcium, 0.47% phosphorus, and 2912 kcal/kg gross energy. Malaria and dengue illness are treated using papaya leaves. Papaya leaves have been shown to greatly reduce the risk of coccidiosis (AL-Fifi, 2007; Nghonjuyi, 2015). It has been discovered that papaya leaf contributed to the blood's color, which in the supplemented study had the maximum quantity of RBC, hemoglobin, platelets, and PCV (Agboola et al., 2018). Several researches demonstrate that broilers treated with papaya leaf could obtain higher growth performance and immunological response (Sorwar et al., 2016; Oloruntola et al., 2018). Black cumin often referred to as "Kalojeera" locally, and is being recognized as a miracle plant. Black cumin holds the top spot among herbal medications as a result of its remarkable therapeutic abilities. Black cumin has a wide range of medicinal potentials, including antibacterial, antihypertensive, anticancer (Padhye et al., 2008), immune-modulatory, analgesic, antimicrobial, antihelminthic, anti-inflammatory, gastro protective, and renal protective effects (Zaoui et al., 2000). According to several research, black cumin seed was effective at promoting weight gain, FCR (Mansoori et al., 2006; Khan et al., 2012), feed intake, dressing percentage, and the weight of various internal organs (Durrani et al., 2007). On the basis of this background, the trial was designed to investigate the effects of papaya leaf meal and black cumin seed as potential replacements for antibiotic growth promoters on the growth performance, immunological condition, and blood indices of broiler chickens.

MATERIALS AND METHODS

Statement of the Experiment

To determine the viability of using papaya leaf and black cumin in commercial broiler diet on growth performance, immune status, and blood indices of broiler chickens, research was conducted in the experimental trial shed at Sher-e-Bangla Agricultural University Poultry Farm, Dhaka, using 150 day-old Cobb 500 straight run commercial broiler chicks from a single hatch over a 28-day period from February 19 to March 19, 2019.

Experimental Materials and Design

From a reputable hatchery, 150-day-old Cobb 500 straight run commercial broiler chicks were collected. Following standard brooding practices, they were kept in an electric brooder for seven days. Only the most basic nutrients in farm made starter (3027 ME Kcal/Kg & 21.86% CP) and grower ration (3152 ME Kcal/Kg & 21.12% CP) was offered. One hundred and fifty (150) DOCs were divided into five treatment groups at random after four days, with three replicates in each group (10 chicks). Good-quality papaya leaves and black cumin were gathered from papaya

gardens and nearby markets, respectively. Following collection, papaya leaves were properly cleaned, allowed to air dry beneath a shed for seven days, and then crushed in a wooden mortar and pestle. Then, using a sieve, the leaf veins were separated. Moreover, fresh drinking water was used to wash and adequately dry the black cumin. Both components were suspended in air. Until utilized, both ingredients were kept separate in an airtight container.

The following experimental diets were created T1 = Basal diets (control) T2 = Basal diets + Antibiotics ((Doxivet ® - 1g/2 litre of drinking water) T3 = 2% PLM (2 kg of PLM/100 kg of the feeds) T4 = 1% BCS (1 kg of BCS/100 kg of the feeds) T5 = 1% PLM & 1% BCS (1 kg PLM & 1 kg BCS/100 kg of the feeds)

Management Procedures

The experimental shed was cleaned, sanitized, and disinfected. The open-sided, south-facing shelter allows cross ventilation. Over the duration of the experiment, averages for room temperature (290C) and relative humidity (73%) were noted. Rice husk was used as the litter material, and a depth of 6 cm of litter was formed. Litter was stirred at the end of each day to minimize the buildup of noxious substances and to lessen parasite infestation. The birds were given rations and free access to clean water. Water was provided twice day, while food was provided three times daily. The shed was equipped with lighting at night. For the first two days, there was 24 hour light. Ranikhet Disease, Infectious Bronchitis and Infectious Bursal Disease (IBD) vaccines were given to chicks in time.

Recorded Parameters

Information was gathered on the weekly live weight, weekly feed consumption, and chick deaths to calculate survivability rate. The FCR was determined using the ultimate live weight and total feed consumed by each bird in each replication. At the end of the experiment, In order to assess the weight of the broiler chicken's spleen and bursa, three birds from each replicate that had been fasting for 12 hours were randomly chosen and slaughtered. The wing vein was used to collect blood samples (about 3 mL each) that were used to evaluate the hemato-biochemical characteristics of the birds before slaughter. Ethylene diethyltetraacetic acid (EDTA) was added to the blood sample tubes; however a portion of each sample was collected without EDTA to obtain serum. Within an hour of being collected, samples were brought to the lab for analysis. Analysis from each replication to measure glucose and cholesterol level, hemoglobin, RBC, WBC, PCV level and lymphocyte percentage.

Analytical Statistics

The data was statistically analyzed using the statistical package for social sciences (SPSS) version 16 by applying one way ANOVA. Duncan's multiple comparison test was used to evaluate differences between means, with a significance level of p<0.05.

RESULTS AND DISCUSSION

Production Index *Final Live Weight*

The effects of treatments on final live weight (g/bird) were not significant (p>0.05), according to the data reported in Table 1. However, the 2% PLM treated group was shown to have an insignificantly (p>0.05) higher Final live weight (1559.57g) of birds than the other groups. These findings agreed with those reported by Unigwe et al. (2014), who discovered that dietary PLM supplementation had no noticeable (p>0.05) effect on the average final body weight of broiler chicken. Moreover, these results were at conflict with Ebenebe et al. (2011) who found that adding PLM to the diet of broiler chickens resulted in greater weight gain than in the control group. According to Jahan et al. (2015), adding BCS Meal to the diet increased the live weight of broiler chickens by a significant (p<0.05) amount when compared to the control group.

Total Feed Consumption

No significant (p>0.05) differences in the feed intake of broiler chickens different treatment groups were revealed by the data in Table 1. Insignificantly (p>0.05), the control group consumed more feed per bird (2212.60g), while the 1% BCS (T4) treatment group received less feed per bird (2175.23g). These findings concur with those of Bolu et al. (2009) who noted that adding papaya leaf to broiler diets had no significant impact (p>0.05) on feed intake and Guler et al. (2006) who also noted that adding black cumin seed to broiler diets had no significant impact (p>0.05) on feed consumption of broiler chicken. However, other researchers (Rumokoy et al., 2016) came to the opposite conclusion, finding that dietary papaya extract supplementation had a greater significant (p<0.01) interaction on feed intake.

Feed Conversion Ratio

According to the data in Table 1, the treatments' effects on the feed conversion ratio (FCR) in broiler chickens were not statistically significant (p>0.05). Numerically (p>0.05) the control group FCR was higher (1.46), while the birds supplemented with 2% PLM had a lower FCR (1.41). These outcomes were in line with those of Unigwe et al. (2014), who came to the conclusion that FCR in broilers supplemented with dried PLM at 5%, 10%, and 15% inclusion levels were not significantly (p>0.05) different between all treatment groups and compared to that of the control group. Moreover, results from other studies (Majeed et al., 2010; Saeid et al., 2013) revealed that the dietary supplementation of Black cumin seed did not significantly affect FCR in broiler chicken.

Survivability

According to the data in Table 1, adding PLM and BCS to the diet had no negative effects on the broiler chicken's survival rate (100%). During the study period, No mortality was found.

Immune Organs

Table 2 showed that adding papaya leaf meal and black cumin seed to broiler ration did not significantly (p>0.05) affect spleen weight (g), but did significantly (p<0.05) affect bursa weight. Insignificantly (p>0.05) the lowest value was in the control group (1.78g), whereas the greatest spleen weight (2.28g) was recorded in the T4 group. In this case, significantly (p<0.05) the highest bursa weight (2.50g) was found in the 2% PLM supplemented group and lowest (1.67g)

in the control group. In relation to the findings, a number of researchers reported that the addition of papaya leaf (Battaa et al., 2015; Haruna and Odunsi, 2018) and black cumin (Toghyani et al., 2010) seed supplements increased spleen and bursa weight in comparison to the control group.

Serum Biochemical Parameters

Moreover, Table 3 showed that there were no significant differences (p>0.05) in the concentrations of glucose and cholesterol (mg/dl) throughout the various dietary groups Mean value of glucose and cholesterol was 196.89mg/dl and 172.22mg/dl respectively. The 2% PLM supplementation group (T3) had lower cholesterol levels (169.11 mg/dl) than the antibiotic and control groups, but there was no statistically significant (p>0.05) difference among the groups. The results of the current study contradicted those of other researchers (Akhtar et al., 2003; Khadr and Abdel-Fattah, 2006), who claimed that dietary papaya leaf and black cumin supplementation effectively decreased serum glucose level. These findings are in line with those of other studies (Zetina-Esquivel et al., 2015), which found that supplementing with PLM reduced serum cholesterol levels in hypercholesterolemic rats.

Table 1 Production index of broiler chicken supplemented with Papaya leaf meal, Black cumin seed and antibiotic.

Treatments	Final Live Weight	Total Feed Consumption	FCR	Survivability (%)
	(g)/bird	(g)/bird		
T_1	1511.90 ± 10.11	2212.60 ± 11.32	$1.46 \pm .01$	100.00 ± 00
T_2	1541.33 ± 22.25	2193.70 ± 17.50	$1.42 \pm .02$	100.00 ± 00
T_3	1559.57 ± 26.404	2198.43 ± 10.45	$1.41 \pm .03$	100.00 ± 00
T_4	1532.77 ± 27.16	2175.23 ± 7.04	$1.42 \pm .02$	100.00 ± 00
T_5	1520.27 ± 10.58	2184.03 ± 10.52	$1.44 \pm .02$	100.00 ± 00
$Mean \pm SE$	$1533.17^{\text{NS}} \pm 9.00$	$2192.80^{NS} \pm 5.63$	$1.43^{NS} \pm .01$	100.00 ± 0.00

Here, T_1 = (Control); T_2 = (Antibiotic), T_3 = (2% PLM supplementation); T_4 = (1% BCS supplementation) and T_5 = (1% PLM & 1% BCS supplementation); NS= Non-significant (p>0.05); SE= Standard Error

Table 2 Effects of Papaya leaf meal, Black cumin and antibiotic supplementation to broiler diet on some immune organs.

Treatments	Spleen weight (g)	Bursa weight (g)	
T ₁	1.78 ± 0.12	$1.67\pm0.17^{\mathrm{b}}$	
T_2	1.83 ± 0.14	2.22 ± 0.25^{ab}	
T ₃	1.940 ± 0.29	$2.50\pm0.41^{\rm a}$	
T_4	2.28 ± 0.17	$1.72\pm0.22^{\mathrm{ab}}$	
T ₅	1.94 ± 0.16	$2.22\pm0.12^{\mathrm{ab}}$	
Mean ±SE	$1.96^{\rm NS}\pm0.08$	$2.07 \pm 0.12*$	

Here, T_1 = (Control); T2= (Antibiotic); T3= (2% PLM supplementation); T₄= (1% BCS supplementation) and T₅= (1% PLM & 1% BCS supplementation); * = Significant (p<0.05); NS= Non-significant (p>0.05) ^{a, b} Mean values with different superscripts within the same column differ significantly; SE= Standard Error

Table 3 Effects of Papaya leaf meal, Black cumin seed and antibiotic supplementation on serum biochemical concentration of broiler chickens.

Treatments	Glucose (mg/dl)	Cholesterol (mg/dl)
T_1	199.11 ± 3.65	176.00 ± 3.85
T_2	202.11 ± 5.18	172.11 ± 4.72
T_3	196.11 ± 2.12	169.11 ± 5.31
T_4	192.11 ± 1.47	170.67 ± 5.21
T_5	195.00 ± 2.06	173.22 ± 5.39
Mean \pm SE	$196.89^{\rm NS} \pm 1.46$	$172.22^{NS} \pm 2.13$

Here, T_1 = (Control); T2= (Antibiotic); T3= (2% PLM supplementation); T₄= (1% BCS supplementation) and T₅= (1% PLM & 1% BCS supplementation); NS= Non-significant (p>0.05); SE= Standard Error

Table 4 Effects of Papaya leaf meal, Black cumin seed and antibiotic on blood parameters of broiler chickens.

Treatment group	Hb (g/dl)	RBC (million/cumm)	PCV %	WBC (×10 ³ /cumm)	Lymphocytes%
T_1	10.63 ± 0.30	$3.61 \pm 0.10^{\circ}$	$36.99\pm0.32^{\rm c}$	$7.48\pm0.44^{\rm c}$	23.56 ± 0.63^{b}
T_2	10.97 ± 0.36	3.98 ± 0.08^{b}	$35.60\pm0.45^{\rm c}$	7.77 ± 0.50^{bc}	24.00 ± 0.82^{b}
T_3	11.93 ± 0.57	$4.51\pm0.16^{\rm a}$	42.00 ± 1.18^{a}	$9.53\pm0.08^{\rm a}$	35.22 ± 2.60^a
T_4	11.53 ± 0.44	$4.39\pm0.10^{\rm a}$	38.41 ± 1.11^{bc}	8.03 ± 0.34^{bc}	31.11 ± 2.14^{a}
T_5	11.32 ± 0.40	4.28 ± 0.09^{ab}	41.00 ± 1.94^{ab}	8.86 ± 0.41^{ab}	32.67 ± 1.85^a
Mean± SE	$11.28^{\text{NS}}\pm0.19$	$4.15\pm0.07*$	$38.80\pm0.61*$	$8.33\pm0.20^*$	$29.31 \pm 1.04*$

Here, T_1 = (Control); T2= (Antibiotic); T3= (2% PLM supplementation); T_4 = (1% BCS supplementation) and T_5 = (1% PLM & 1% BCS supplementation); * = Significant (p<0.05); NS= Non-significant (p>0.05); ^{a, b, c} Mean values with different superscript letters within the same column differ significantly; SE= Standard Error

Hematological Parameters

Table 4 showed that, with the exception of Hb (g/dl), papaya leaf meal and black cumin seed in the feed had a significant (p<0.05) effect on the RBC (million/cumm), PCV (%), WBC (×103/cumm) and lymphocyte (%) concentrations of broiler chicks in the treatment group. RBC, WBC, lymphocyte and PCV values were greater (p<0.05) in the T3 (2% PLM), T4 (1% BCS), and T5 (1% of both PLM and BCS) groups than in the antibiotic (T2) and control (T1) groups. RBC (4.51 million/cumm), PCV (42.00%), WBC (9.53×103 /cumm) and lymphocyte (35.22%) values were highest in the T3 group. Agboola et al. (2018) found that broiler chicken diets incorporating papaya leaf meal showed the greatest values of packed cell volume, red blood cells, and hemoglobin in all the papaya leaf included treatments, were in agreement with these findings. On the other hand, these results were at conflicts with those of earlier studies, which found that dietary supplements of Carica papaya leaf and black cumin did not significantly differ (p>0.05) from control groups in terms of the general blood parameters (Jamroz and Kamel, 2002; Bolu et al., 2009).

CONCLUSION

According to the results of this study indicated that birds fed 2% PLM had numerically higher body weight, improved FCR, and lower serum cholesterol levels. Moreover, hematological indicators improved and the weight of the immune organ (bursa) increased significantly as compared to the control, antibiotic, and other supplemented groups receiving Black cumin seed and Papaya leaf meal diet.

CONFLICT OF INTEREST

The authors declared there is no conflict of interest.

REFERENCES

- Agboolal BE, AD Ologhobo, IO Adejumo and GO Adeyemo (2018): Response of Broiler Chickens to Carica papaya and Taliniumtriangulare Leaf Meal under Normal and Subnormal Diets. Annual Research & Review in Biology, 23: 1-7.
- Akhtar MS, Z Nasir and AA Rehman (2003): Effect of feeding powdered Nigella sativa seeds on poultry egg production and their suitability for human consumption.Veterinarskiarhiv, 73: 181-190.
- AL-Fifi Z (2007): Effect of leaves extract of Carica papaya, Vernoniaamigdalina and Azadiratchaindica on the coccidiosis in free-range chickens. Asian Journal of Animal Sciences, 1: 26–32.
- Battaa, AM El-Neney, NB Awadien and TA Ebeid (2015): The productive performance and immunological traits of local chicken strain by using natural enzymes (plant papain) and remnants of plant papaya. Effect of papaya latex on laying period. Egypt Poultry Science, 35: 1-24.
- Boamah VE, C Agyare, H Odoi, A Dalsgaard (2016): Practices and factors influencing the use of antibiotics in selected poultry farms in Ghana. Journal of antimicrobial Agents, 2: 2-8.
- Bolu SAO, FE Sola-Ojo, OA Olorunsanya and K Idris (2009): Effect of Graded Levels of Dried Pawpaw (Carica papaya) Seed on the Performance, Hematology, Serum Biochemistry and Carcass Evaluation of Chicken Broilers. International Journal of Poultry Science, 8: 905-909.
- Burkill HM (1985): The useful plants of West Tropical Africa. Royal Botanic Garden, Kew, UK. Volume 3.
- Carvalho LT and L Santos (2016): Antibiotics in the aquatic environments: a review of the European scenario. Environment International, 94: 736-757.
- Durrani FR, N Chand, K Zaka, A Sultan, FM Khattak and Z Durrani (2007): Effect of different levels of feed added black seed (Nigella sativa L) on the performance of broiler chicks. Pakistan Journal of Biological Sciences, 10: 4164-4167.
- Ebenebe CI, O Itefue, TC Ebere-Ohameje and JC Okonkwo (2011): Fortification of the nutritive value of Mushroom (Termitomycesmicrocarpus) with paw-paw leaf meal for broiler chicks diet. Pakistan Journal of Nutrition, 10: 155-158.
- Forgetta V, H Rempel, F Malouin, R Vaillancourt Jr, E Topp and K Dewar (2012): Pathogenic and multidrug-resistant Escherichia fergusonii from broiler chicken. Poultry Science, 91: 512-525.
- Furtula V, EG Farrell, F Diarrassouba, H Rempel, J Pritchard and MS Diarra (2010): Veterinary pharmaceuticals and antibiotic resistance of Escherichia Coli isolates in poultry litter from commercial farms and controlled feeding trials. Poultry Science, 89: 180-188.

- Guler T, B Dalkilic, ON Ertas and M Ciftici (2006): The effect of black cumin seeds on the performance of broilers. Asian-Australasian Journal of Animal Sciences, 19: 425-430.
- Guo FC, BA Williams, RP Kwakkel and MWA Verstegen (2003): In Vitro fermentation characteristics of two mushroom species, an herb and their polysaccharide fractions, using chicken cecal contents as inoculum. Poultry Science, 82: 1608-1615.
- Haruna MA and AA Odunsi (2018): Growth Performance and Carcass Quality of Broiler Chickens Fed Dried Pawpaw (Carica Papaya Linn) Latex. Journal of World's Poultry Research, 8: 31-36.
- Jahan MS, M Khairunnesa, S Afrin and MS Ali (2015): Dietary Black cumin (Nigella sativa) seed meal on growth and meat yield performance of broiler. SAARC Journal of Agriculture, 13: 151-160.
- Jamroz D and C Kamel (2002): Plant extracts enhance broiler performance. In non- ruminant nutrition: Antimicrobial agents and plant extracts on immunity, health and performance. Journal of Animal Science, 80:41-41.
- Kale BP, MA Kothekar, HP Tayade, JB Jaju and M Mateeddin (2003): Effect of aqueous extract of Azadirachtaindica leaves on hepatotoxicity induced by ant tubercular drugs in rats. Indian Journal of Pharmacology, 35:177.
- Karmoker Y (2022): Self Sufficiency in Protein: Poultry Industry in Bangladesh.https://businessinspection.com.bd/poultry-industry-in-bangladesh.
- Khadr NA and FAI Abdel-Fattah (2006): Response of Broiler Chickens to Diet Containing Black Seed (Nigella sativa L.) as Medical Plant. Benha Veterinary Medical Journa,17: 323-343.
- Khan SH, J Ansari, UH Ahsan and A Ghulam (2012): Black cumin seeds as phytogenic product in broiler diets and its effects on performance, blood constituents, immunity and caecal microbial population. Italian Journal of Animal Science, 11: 438 – 444.
- Majeed LHA, KA Abdelati, NM El-Bagir, A Alhaidary, HE MOHAMED and AB Beynen (2010): Performance of broiler chickens fed diets containing low inclusion levels of black cumin seeds. Journal of Animal Veterinary Advances, 9: 2725-2728.
- Mansoori B, M Modirsanei and MK Saeid Mohammad (2006): Cumin seed meal with enzyme and polyethylene glycol as an alternative to wheat bran in broiler diets. Journal of the Science of Food and Agriculture, 86: 2621-2627.
- Nghonjuyi NW (2015): Efficacy of ethanolic extract of Carica papaya leaves as a substitute of sulphanomide for the control of coccidiosis in KABIR chickens in Cameroon. Journal of Animal Health and Production, 3: 21–27.
- Oloruntola OD, SO Ayodele and DA Oloruntola (2018): Effect of pawpaw (Carica papaya) leaf meal and dietary enzymes on broiler performance, digestibility, carcass and blood composition. Revue d'élevageet de médecinevétérinaire des pays tropicaux, 71: 121-129.
- Padhye S, S Banerjee, A Ahmad, R Mohammad and FH Sarkar (2008): From here to eternity the secret of Pharaoh: Therapeutic potential of N. sativa seeds and beyond. Cancer Therapy, 6: 459-510.

- Ronquillo MG and JC Hernandez (2017): Antibiotic and synthetic growth promoters in animal diets: review of impact and analytical methods. Food control, 72:255-67.
- Rumokoy L, E Pudjihastuti, IM Untu and WL Toar (2016). The Effects of Papain Crude Extract Addition in Diets on Broilers Production Performances. Animal Production, 18: 30-35.
- Saeid JM, Arkan, B Mohamed and MA Al-Baddy (2013): Effect of adding Garlic Powder (Allium sativum) and Black Seed (Nigella sativa) in Feed on Broiler Growth Performance and Intestinal Wall Structure. Journal of Natural Sciences Research, 3: 35-41
- Sorwar MG, M Mostofa, MN Hasan, M Billah and MT Rahman (2016): Effect of kalojeera seed and papaya leaf supplementation on the performance of broiler. Bangladesh Journal of Veterinary Medicine, 14: 37-42
- Toghyani M, A Gheisari, G Ghalamkari and MM Rezaei (2010). Growth performance, serum biochemistry and blood hematology of broiler chicks fed different levels of black seed (Nigella sativa) and peppermint (Menthapiperita). Livestock Science, 129: 173-178.
- Unigwe CR, UP Okorafor, UM Ogbu1 and C Nwufoh (2014): The Nutritive Profile of Sun-Dried Paw-Paw (Carica Papaya) Leaf Meal and its Effect on the Growth Performance of Broiler Chickens. International Journal of Pure and Applied Sciences and Technology, 20: 72-78.
- Zaoui A, Y Cherrah, MA Lacaille-Dubois, A Settaf, H Amarouch and M Hassar (2000): Diuretic and hypotensive effects of Nigella sativa in the spontaneously hypertensive rat. Therapies, 55: 379-382.
- Zetina-Esquivel AM (2015). Effect of Carica papaya Leaf Extract on Serum Lipids and Liver Metabolic Parameters of Rats Fed a High Cholesterol Diet. Health, 7: 1196-1205.