Growth performance, carcass yield and bacteria load of broiler chickens on oral administration of Nigella sativa oil

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Abstract
One hundred and forty-four, one day – old broiler (arbor acre strain) chicks were used in a 49-day trial to assess the influence of oral administration of black seed oil on growth performance, carcass traits and bacteria load in broiler chickens. The birds were divided into four groups (36 per group) and randomly assigned to treatments with varying levels of black seed oil (1.5 ml/L, 3.0 ml/L and 4.5 ml/L of water) and control (antibiotics). Data obtained were subjected to one-way Analysis of variance at α = 0.05. Oral administration of varying levels of Nigella sativa oil did not have any significant (P>0.05) effect on the growth performance indices (final weight includes 1899.49, 1940.28, 1888.51 and 1795.83g across the groups) and carcass traits of broiler chickens. Black seed oil had no significant (p>0.05) effect on the faecal bacteria count, only numerically reduced total blood coliform count with increasing dosage of Nigella sativa oil (0.50, 0.40, 0.33 and 0.40 × 10^6 cfu/ml) in water while caecal coliform reduced (p <0.05) markedly at 4.5ml/L dosage (2.03 vs 2.53, 2.37, 2.43). It was concluded that a minimum dosage of 4.5ml/L of black seed oil be adopted for a significant reduction in bacterial load in chickens.

Keywords: Performance, carcass trait, bacterial load, antibiotics and Nigella sativa

Introduction
Growth promoters are chemical and biological substances that are added to livestock feed to improve the growth of broiler chickens. Antibiotics used to be the principal growth – promoting substances in poultry production. The results of using antibiotics were magnificent in increasing the performance traits, but they contributed to the development of antibiotics – resistant bacteria (Kim et al., 2011; Hassanpou et al., 2013). Inclusion of antibiotics as a principal growth promoter in poultry feed often resulted in the incidence of cross resistance among pathogens and also a source of residues in animal body tissues (Schwarz et al., 2001). Consequently, the European Union banned the use of antibiotics as a growth promoter in animal feeds in January 2006 (Toghyani et al., 2010) thus leading to researches on alternative natural growth promoting substances such as essential oils and medicinal plants, which are proving to be more beneficial because of their antimicrobial effects (Elgayyar et al., 2001; Salmeron, 2003). Nigella sativa seed (Kalanji or black cumin) family Ranunculaceae is utilized as a medicinal plant, herb and spice worldwide (Akhtar et al., 2003). Its oil is a multipurpose feedbase growth promoter and very promising in improving broiler performance (Al Beitawi et al., 2009). The use of black cumin (1%) on broiler chickens resulted in increased carcass yield, liver, abdominal fat, breast, thigh, wing and neck weight
Growth performance, carcass yield and bacteria load of broiler chickens on oral administration

(Guller et al., 2007; Toghyani et al., 2010). In addition, Hermes et al. (2009) reported that Black seed oil also known as Nigella seed oil (NSO) at 0.5 and 1% increased growth and relative weight of caecum, liver, spleen and thymus gland while decreasing the mortality rate in broiler chickens. Arslan et al. (2005) also found that black seed oil supplementation in broiler chickens increased their growth rate and improved their feed conversion ratio while decreasing their feed intake. The seeds of Nigella sativa were shown to be effective against total coliform count in the intestine of broilers (Erener et al., 2010). Oil extracted from black seed was shown to effectively inhibit L. monocytogenes (Nair et al., 2005; Ali et al., 2007). Erener et al. (2010), reported that Nigella sativa Linn was also able to increase the broilers resistance to the propagation of Enterobacteriaceae, Also Nigella sativa Linn was found to have an antimicrobial activity on Escherichia coli, Bacillus subtilis, Streptococcus faecalis, Staphylococcus aureus, Pseudomonas aeruginosa and Candida albicans (Kooti et al., 2016 and Chahal et al., 2017).

Previous studies have only considered the use of Nigella seed oil in feed, this study therefore investigated the effects of oral administration of Nigella sativa oil in drinking water on growth performance indices, carcass traits and bacteria load of broiler chickens.

Materials and methods

Experimental site
The study was carried out at the Poultry Unit of the Teaching and Research Farms of the Federal University of Agriculture, Abeokuta (Latitude 7°N and Longitude 3° E) in Nigeria (Google Earth, 2018). The farm site is located in the rain forest zone of South - Western Nigeria with altitude 76mm above the sea level. The annual mean rainfall, mean temperature and humidity are 1037mm, 34.7°C and 82%, respectively.

Source of test ingredient
Black seed oil extracted and prepared from natural black seeds using latest cold pressing technique was purchased from a reputable store in Abeokuta.

Experimental animals and brooding
One hundred and forty-four (144), one day-old arbor acre broiler chicks were obtained from a reputable hatchery. Prior to their arrival, biosecurity measures were taken. The brooding pen was washed and fumigated. Drinkers and feeders were thoroughly washed. Wood shavings was provided and spread as litter materials. On arrival, the day-old chicks were placed in the brooding pen with a source of heat such as charcoal coal pot and bulb to light up the brooding pen. The temperature was well monitored using LCD digital thermometer kept at the level of the birds. Feed and water were provided ad-libitum.

Experimental design and management
The experiment consists of four treatments viz: the dosages of black seed oil (1.5 ml/L, 3.0 ml/L and 4.5 ml/L) and control. The birds were divided into four groups (36 per group) and randomly assigned in a Completely Randomized Design to the four treatments at day old. Each treatment was replicated thrice with12 birds per replicate. Birds were fed commercial starter diet and finisher diet at the starter (0 - 4 weeks) and finisher (4 - 7 weeks) phase, respectively, ad libitum. Fresh clean water was given daily. Birds were vaccinated against Gumboro disease (7th and 21st day) and Newcastle disease (28th day). The control groups were administered antibiotics (Enrofloxacin) while other groups were exempted. Black seed oil was administered at the varying levels to the respective groups in water for three consecutive days in a week throughout the experimental period.
Data collection
Body weight, weight gain, feed intake, water intake and mortality rate of the birds were recorded on a weekly basis. Feed Conversion Ratio was also calculated.

Carcass yield
On the 49th day of the study, two (2) birds with weight equal to average of the birds in each replicate were selected and sacrificed via severing of the carotid arteries to bleed for 2 minutes followed by scalding at 60°C to evaluate carcass traits. The heads and shanks were removed and weighed after defeathering. The viscerales were removed and the dressed weight was determined. The weight of the cut parts (thigh, breast, back, wings and drumstick), organs (heart, liver, and gizzards) and the abdominal fat were determined and expressed as a percentage of the live weight.

Bacteria load estimation

Faecal collection and analysis
Faecal samples were collected at the 49th day in order to determine the bacteria count in their faeces. The excreta microbial count was estimated using procedures of estimation of bacterial total count according to Hedge (2002).

Bacteria count and coliform count
Five (5ml) of blood samples was collected from the jugular vein at the neck of one bird per replicate at the 49th day of the experiment in a sterile bottle. The blood samples were taken to the laboratory to determine the coliform count as well as identify the coliform bacteria present according to Kinley (2009).

Statistical analysis
The data obtained were subjected to One–way Analysis of Variance (ANOVA) in a Completely Randomized Design using SAS (2000) while significant means was separated using Duncan's Multiple Range Test at 5% level of significance.

Results and discussion
Growth performance of broiler chickens

Effect of oral administration of *Nigella sativa* oil on the growth performance of broiler chickens is presented in Table 1. All parameters measured for the effect of oral administration of *Nigella sativa* oil were not significantly (P>0.05) different. Similar performance among the various groups (antibiotics and black seed oil) could be adduced to high level of hygiene adhered to during the study. Lee et al. (2003) reported that well-nourished healthy chickens do not respond to antibiotics supplement (as well as phytobiotics) provided they are housed under clean and disinfected conditions. This present study showed no significant effect on the feed intake. Similarly, Denli et al. (2004) reported that supplementation with black cumin seed extract did not significantly affect feed intake of quail. In contrast, Abdul - Karim and Mohammed (2013) results on birds given 2.5 % and 3.5 % black cumin seed in diets revealed significant increase in average daily feed intake when compared with the control groups or birds getting 1.5% black cumin seed. Sogut et al. (2012) also reported that supplementation of black seeds to the broilers chicks diet resulted in a significant (P < 0.05) decrease in feed intake. Birds in this study received black seed oil through water and not feed which suggest the reason it neither impaired or improved feed intake of the birds. Similar effect of *Nigella sativa* on mortality observed in this study is in consonance with the findings of Ismail (2011) who observed no effect of inclusion of black cumin in broiler chickens diet on mortality. However, Hermes et al. (2009) reported that Nigella seed oil (NSO) at 0.5 and 1% significantly decreased mortality rate in broiler chickens.

Carcass characteristics of broiler chickens
Effect of oral administration of *Nigella sativa* oil on the carcass characteristics of broiler chickens is presented in Table 2. All parameters measured for the effect of *Nigella sativa* oil were not significantly
Table 1: Effect of Black Seed (Nigella sativa) oil on the growth performance of broiler chickens

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Antibiotics</th>
<th>1.5ml/l</th>
<th>3.0ml/l</th>
<th>4.5ml/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial weight (g/bird)</td>
<td>44.45±4.81</td>
<td>43.06±2.40</td>
<td>43.06±2.40</td>
<td>43.06±2.40</td>
</tr>
<tr>
<td>Final weight (g/bird)</td>
<td>1943.94±103.28</td>
<td>1983.33±192.21</td>
<td>1931.56±23.18</td>
<td>1838.89±91.79</td>
</tr>
<tr>
<td>Total weight gain (g/bird)</td>
<td>1899.49±98.84</td>
<td>1940.28±193.84</td>
<td>1888.51±24.47</td>
<td>1795.83±90.81</td>
</tr>
<tr>
<td>Weight gain/day (g/bd/day)</td>
<td>38.76±2.02</td>
<td>39.60±3.96</td>
<td>38.54±0.50</td>
<td>36.65±1.86</td>
</tr>
<tr>
<td>Total feed intake (g/bird)</td>
<td>5123.00±162.52</td>
<td>5061.00±112.97</td>
<td>4781.33±826.04</td>
<td>4977.67±57.29</td>
</tr>
<tr>
<td>Feed intake/day (g/bird/day)</td>
<td>104.55±3.32</td>
<td>103.29±2.31</td>
<td>97.58±16.86</td>
<td>101.59±1.17</td>
</tr>
<tr>
<td>Total water intake (ml/bird)</td>
<td>9154.60±371.51</td>
<td>9171.02±502.84</td>
<td>8780.93±856.39</td>
<td>8794.02±131.55</td>
</tr>
<tr>
<td>Water intake/day (ml/bird/day)</td>
<td>213.66±10.06</td>
<td>218.35±8.97</td>
<td>218.92±5.50</td>
<td>206.16±5.33</td>
</tr>
<tr>
<td>FCR</td>
<td>2.70±0.06</td>
<td>2.62±0.19</td>
<td>2.53±0.43</td>
<td>2.78±0.18</td>
</tr>
<tr>
<td>Mortality (%)</td>
<td>5.55±4.81</td>
<td>0.00±0.00</td>
<td>5.55±4.81</td>
<td>0.00±0.00</td>
</tr>
</tbody>
</table>
Table 2: Effect of black seed (*nigella sativa*) oil on the carcass characteristics of broiler chickens

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Antibiotics</th>
<th>1.5ml/l</th>
<th>3.0ml/l</th>
<th>4.5ml/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>Live weight (g)</td>
<td>1766.67±57.74</td>
<td>1733.33±321.4</td>
<td>1700±50.00</td>
<td>1633.33±160.73</td>
</tr>
<tr>
<td>Eviscerated weight (%)</td>
<td>77.80±1.10</td>
<td>76.73±4.78</td>
<td>71.81±3.80</td>
<td>69.77±4.84</td>
</tr>
<tr>
<td>Dressed weight (%)</td>
<td>66.39±1.49</td>
<td>62.83±0.67</td>
<td>61.75±2.97</td>
<td>59.05±4.22</td>
</tr>
<tr>
<td>Cut parts (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thigh</td>
<td>10.58±1.01</td>
<td>9.64±0.22</td>
<td>10.05±0.85</td>
<td>9.37±0.85</td>
</tr>
<tr>
<td>Drumstick</td>
<td>10.68±0.42</td>
<td>10.26±1.07</td>
<td>10.02±1.21</td>
<td>9.40±0.51</td>
</tr>
<tr>
<td>Back</td>
<td>14.64±0.32</td>
<td>14.72±1.36</td>
<td>13.56±1.64</td>
<td>13.17±1.28</td>
</tr>
<tr>
<td>Wings</td>
<td>8.36±0.80</td>
<td>8.69±0.45</td>
<td>8.13±0.64</td>
<td>7.58±0.37</td>
</tr>
<tr>
<td>Breast</td>
<td>20.91±1.69</td>
<td>21.55±2.79</td>
<td>19.19±1.82</td>
<td>18.68±3.11</td>
</tr>
<tr>
<td>Internal organs (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Liver</td>
<td>1.72±0.16</td>
<td>1.52±0.15</td>
<td>1.87±0.44</td>
<td>1.85±0.18</td>
</tr>
<tr>
<td>Empty gizzard</td>
<td>1.72±0.22</td>
<td>1.91±0.38</td>
<td>1.88±0.21</td>
<td>2.00±0.11</td>
</tr>
<tr>
<td>Heart</td>
<td>0.41±0.03</td>
<td>0.47±0.09</td>
<td>0.35±0.06</td>
<td>0.34±0.01</td>
</tr>
<tr>
<td>Abdominal fat</td>
<td>0.52±0.30</td>
<td>0.75±0.70</td>
<td>0.34±0.60</td>
<td>0.16±0.28</td>
</tr>
</tbody>
</table>

Table 3: Bacteria load of broiler chickens administered varying dosages of black seed oil in water

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Antibiotics</th>
<th>1.5ml/l</th>
<th>3.0ml/l</th>
<th>4.5ml/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>FBC ((× 10^6 cfu/ml))</td>
<td>0.70±0.26</td>
<td>0.70±0.10</td>
<td>0.63±0.15</td>
<td>0.50±0.36</td>
</tr>
<tr>
<td>CC ((× 10^6 cfu/ml))</td>
<td>2.53±0.12(^a)</td>
<td>2.37±0.35(^a)</td>
<td>2.43±0.38(^a)</td>
<td>2.03±0.06(^b)</td>
</tr>
<tr>
<td>TBCC ((× 10^6 cfu/ml))</td>
<td>0.50±0.87</td>
<td>0.40±0.61</td>
<td>0.33±0.58</td>
<td>0.40±0.69</td>
</tr>
</tbody>
</table>

\(^a\)\(^b\) means within row with different superscript differs significantly (P<0.05)

Ayoola, Ekunsetan, Olatunbosun, Muhammad, Oguntoye and Adejola (P>0.05) different. Similar carcass traits observed in this study is in consonance with the findings of Cetin et al. (2008) and Toghyani et al. (2010) on carcass characteristics in chickens fed *Nigella sativa* oil. In a related study, Shewita and Taha (2011) reported no significant effects of supplemental *Nigella sativa* (1% and 0.4%) on dressing percentage or relative weight of inner edible organs in broiler chickens. Contrarily, increased carcass yield, liver, abdominal fat, breast, thigh, wing and neck weights was reported by Guler et al. (2007) and Toghyani et al. (2010) in broiler chickens fed diet containing 1% black cumin. According to Durrani et al. (2007) and Nasir and Grashorn (2010) supplementation of broiler chicken feed with varying levels of the *Nigella sativa* seed, alone or in combination with other medicinal plants, improved dressing percentage and breast and thigh weights of the carcass, while at the same time reducing abdominal fat weight, compared to the control diet lacking any supplementation. Frequency of use, dosage as well as in-water application adopted in the study could be responsible for the differences.

Bacteria load of broiler chickens administered varying dosages of black seed oil in water

Table 3 shows the bacteria load of broiler chickens administered varying dosages of black seed oil orally. The black seed oil had no significant (p>0.05) effect on the faecal total bacteria count of the broiler chickens. Al-Jabre et al. (2003) found that volatile oils in black seed contain constituents capable of inducing beneficial and pharmacological effects against bacteria such as Staphylococcus and *Escherichia coli*.
coli. However, the result of this experiment revealed that the administration of black seed oil at various levels in water for broiler chickens did not significantly influence the total bacteria count. This observation could be that the level of inclusion of black seed oil was not enough to effect a change or the frequency of administration was rather low (3 days in a week). This study also agreed with the study of Bölükbaşı et al. (2009) that coliform count in the faeces did not differ with changes in inclusion levels of the black seed oil in feed (1 ml/kg-3 ml/kg) of laying hens. Similarity in faecal bacteria count of the birds on antibiotics and those administered black seed oil is an indication that the latter can be an effective alternative to the former. Reduced coliform count in birds administered black seed oil especially at the highest dosage (4.5ml/L) is in consonance with the result reported by Arici et al. (2005) in a study investigating the effect of varying levels of administration of black seed extracts on total coliform bacteria count of broiler chickens. This supports the assertion made by several authors (El-fatatry, 1975; Hameed et al. 2008; Jabeen et al. 2008).

The antibacterial activity of black seed oil was due to thymoquinone (TQ) and melanin activity, in the (El-fatatry, 1975: Arici et al., 2005; Bakathir and Abbas, 2011). In a similar study, Nair et al. (2005) reported that *N. sativa* seed oil has a strong antibacterial activity against all the strains of *L. monocytogenes*, yielding a significantly greater inhibition zone than that of the antibiotic gentamicin.

Total blood coliform count was also not significantly (p>0.05) different. Although, the numerical low blood coliform count in birds administered *Nigella sativa* observed in this study corroborated the findings of Erener et al. (2010), on a decline in the total blood coliform count in broiler chickens. Also, Ismail (2011) reported a numerically beneficial effect of Black seed on the total coliform bacteria counts in the caecal intestine of broilers. This may be due to the fact that black cumin bears an excellent potential as alternative to antibiotics and vaccines to improve immunity and to reduce mortality in poultry as reported by Tamoor et al. (2014).

**Conclusion**

It can be concluded that various levels of *Nigella sativa oil* adopted in this study were able to induce similar responses with those administered antibiotics in terms of growth performance, carcass trait and bacterial load except caecal coliforms that was greatly reduced in birds administered 4.5 ml of black seed oil in water.

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