International Journal of Veterinary Sciences and Animal Husbandry

ISSN: 2456-2912
VET 2020; 5(4): 156-159

www.veterinarypaper.com
Received: 18-05-2020
Accepted: 22-06-2020

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Controling coccidiosis in broilers using Tumeric paste

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Abstract
To reduce drug resistance and prevent outbreak of coccidiosis in broiler production, Tumeric paste (TP) was administered per os and used as an anticoccidial to raise broiler chickens. Coccidiosis is the bane of the poultry industry causing considerable economic loss. *Eimeria* species are protozoan parasites and the etiologic agent of coccidiosis causing morbidity and death in poultry. In addition to anticoccidial chemicals and vaccines, natural products are emerging as an alternative and complementary way to control avian coccidiosis. A study was carried out using 60 day old broilers which were randomly allocated into 3 groups: A, infected-treated with 0.5g/kg of turmeric paste, B, infected-treated with 1.0g/kg of turmeric paste and C, infected-untreated control). The groups received equal doses of 16000 sporulated coccidia oocysts orally at week three. Faecal coccidia oocysts counts were evaluated. There was significant difference ($p< 0.05$) in the means of faecal coccidia oocysts count in group B (0.00±0.00) compared to group A (1.66±0.33) and C (11.67±0.33), thus showing that turmeric paste has anticoccidial effects and works better at the dose of 1.0g/kg of body weight.

Keywords: Turmeric paste, coccidiosis, feacal coccidia oocysts count, efficacy dose

Introduction
The principle of poultry production is to achieve a high level of performance through efficient utilization of feed, keeping survivability as maximum as possible [1].
Poultry is one of the quickest and affordable means of meeting the protein (animal origin) requirement in human population. Adversely, poultry diseases are the main constraint in poultry industry in the world [2].
Among all the diseases which affect poultry is coccidiosis [3]. Coccidiosis is a protozoan disease of birds caused by virulent and pathogenic Eimeria species [4, 5].
Eimeria species have a complex life cycle, consisting of three developmental stages (sporogony, schizogony/merogony, and gametogony). Oocysts excreted in poultry feces sporulate in a favourable environment with high humidity at 25–30 °C. Once sporulating oocysts are ingested by birds, physical and chemical agents in their digestive tracts are released and mature infectious sporozoites form sporocysts. The sporozoites enter epithelial cells in the gut, depending on the specific *Eimeria* species, and form trophozoites and, later, schizonts during schizogony/merogony [6]. Merozoites, released from the schizont, can penetrate into the epithelia and continue this merogony stage 2 to 3 times in order to increase the cell number of merozoites at the asexual stage of reproduction. Alternatively, merozoites may enter the sexual stage of reproduction by forming male microgametes, the equivalent of sperm, and female macrogametes, equivalent of oocytes, in host cells. Following fertilization, the zygotes develop into oocysts and are excreted into poultry stool. *Eimeria* species may require 4–7 days to complete their entire life cycles [7], Thus, the asexual and sexual stages of reproduction in the *Eimeria* life cycle have been targeted by anticoccidial compounds [8].
There are various anticoccidial drugs ‘that are used in rearing birds worldwide but the common problem with the drugs is the emergence of resistant strains [9]. When this occurs, it renders drugs ineffective hence newer drugs or other solutions must be provided in order to keep pace with the evolution of the organisms and their disease. Presence of drug residues in eggs and milk is also a point of concern associated with the anti coccidials, so there is need for specified withdrawal periods before slaughter. In an effort to combat drug resistance, Growth promoters also known as feed additives were propounded and used in poultry feeds. These are edible products that can be added to ration to obtain some special effects [10].

Unfortunately, growth promoters have shown many disadvantages such as adverse effects on health of broilers, high cost and long residual properties and development of antibiotic resistance in microbes [11].

In order to abase drug resistance and outbreak of cocciodiosis in poultry industries, beneficial bioactive plant substances were used as an alternative to anticoccidial drugs and growth promoters. These bioactive plant substances yield useful effects in animal nutrition like stimulation of appetite and feed intake, improvement of endogenous digestive enzyme secretions, activation of immune responses, antibacterial, antiviral and antioxidant effects [12]. An example of such beneficial plant is Curcuma longa also known as turmeric. It is a tropical plant originated from southern and eastern Asia. The main yellow bioactive substances isolated from the rhizome of Curcuma longa are curcumin, demethoxy-curcumin, bisdemethoxy-curcumin and tetrahydro curcuminoids [13, 14], These plant extracts were found to have antifungal effects [13], and antioxidative value [14].

Turmeric (*C. longa*) has long been used as a spice and medicinal herb. One publication stated that *C. longa* showed anticoccidial activity [15, 16], Another reported that curcumin (diferuloylmethane), an active compound in *C. longa*, consistently destroyed sporozoites of *E. tenella* [17] Curcumin has also been studied extensively as a chemo-protective agent in several cancers [18], Another study reported that curcumin possesses hepatoprotective, antiviral and anticoccidial effects [19].

However, pharmacokinetic studies of curcuma longa in animals revealed that about 40-85% of an oral dose of turmeric powder (curcumin) passes through the gastrointestinal tract unchanged [20, 21],

Given the potential usefulness of curcumin but owing to its poor absorption in the gastrointestinal tract, this study was carried out to investigate the possibility of enhancing the absorption of turmeric thus increasing its anticoccidial effects in broilers by using Tumeric Paste. Turmeric paste is a mixture of turmeric powder (curcumin), black pepper (piperine) and oil in semi-solid form.

Black Pepper is a flowering vine which belongs to family: Piperaeceae, genus: piper and specie: nigrum. It is rich in glutathione peroxidase and glucose-6-phosphate dehydrogenase [22]. Its antioxidant property has been documented [23]. It is traditionally considered to be a hot, pungent herb that stimulates digestive tract, by supporting the secretion of fluids and circulation of blood in the gastrointestinal tract thereby increases the absorption of selenium, vitamin B complex, curcmin (Tumeric), Beta carotene and other nutrients [24], Black Pepper's most active constituent, Piperine, has been found to support the absorption of other herbs, specifically the Curcumins found in Turmeric. One study found that combining Turmeric and Black Pepper increased the bioavailability of Turmeric by 154% versus Turmeric alone [25], Piperine appears to support the inter-cellular permeability and uptake of Curcumins [26],

Materials and Methods

Sixty hundred (60) day old broiler chicks vaccinated against Newcastle and Gumboro disease but no anticoccidial given were randomly assigned into three groups, each of 20 chicks and reared in deep litter system. The groups (A, and B) received turmeric paste treatment per os at 0.5g/kg, and 1g/kg body weight respectively from day two till the 5th week of their lives while the other group (C) was not given turmeric paste. Also groups (A, B and C) were infected with 16000 sporulated coccidia oocysts per os at 3rd week of rearing them. Clinical examination of birds, feacal samples collection and faecal coccidia oocysts counts were carried out from day four (4) to day seven (7) post infection. The observations made were recorded.

The turmeric paste was prepared with freshly prepared turmeric powder, freshly ground black pepper, water and virgin olive oil at the ratio of 60g of turmeric: 2 cups of water: 1 to 2 teaspoon of black pepper:70mls of extra virgin olive oil [27], With this measurement, the mixture of turmeric and water were cooked for 10minutes. Olive oil and black pepper were added at the end of the cooking. The golden paste was allowed to cool and appropriate dosages were calculated and given to the birds. It was stored in the refrigerator throughout the period of experiment.

Eimeria strains used for the experiment were obtained as described by Ngongeh [2]. The ceca of birds clinically sick from coccidiosis were harvested at autopsy. The contents of ceca were emptied into sieves placed in bowls, crushed, washed out with water and allowed to sediment for 10 minutes. Later, the sediments were suspended in Teflons tubes containing 50mls of water. 20mls of the suspension was poured into five (5) petri dishes and about 2mls of potassium dichromate (sporulating agent) was added and the oocysts were left for 5 days to sporulate. After 5 days, the contents were washed and examined under the microscope to confirm that there was sporulation, identify specie of eimeria that is peculiar to sporulated oocysts and count the number of oocysts that sporulated. The sporulated oocysts were stored in the refrigerator until used for infection.

Means of feacal coccidia oocysts counts for the different infected groups were compared for statistical differences by One-way Analysis of Variance (ANOVA). The means were separated using Post-hock Statistical tool and results were compared with the control using SPSS. All through the experiment, the birds were humanely treated according to ethical standards laid down in the 1964 Declaration of Helsinki, as, operational in Nigeria.

Results

Eimeria strains identified was *Eimeria tenella*. Of all the groups infected with sporulated coccidia oocysts, only the Infected-untreated group showed the following signs: Diarrhea containing blood, listlessness, off feeding, weight loss, droopy wings and one death was recorded. The birds in the other two groups (infected-treated) did not show any clinical signs but coccidia oocysts were found in their faecal samples. The mean faecal coccidia oocysts count of the group treated with 1g/kg of turmeric paste 0.00±0.00 was significantly ($P< 0.01$) lower than the mean faecal coccidia oocysts counts 1.66±0.33, of the group treated with 0.5g/kg of turmeric paste and 11.67±0.33 of the infected-untreated group. Also, the percentage reduction in coccidia oocysts count post treatment(2 weeks) of the group treated with 1g/kg of TP was 100% and this is significantly (P,0.01) higher than that of the group treated with 0.5g/kg of TP but the infected-untreated group showed no reduction in faecal coccidia oocysts count instead it increased.

Table 1: Faecal coccidia oocysts count per gram of faeces of broilers treatment

Group	Faecal Coccidia Oocysts Count After infection	Faecal oocyst count during treatment	Percentage Reduction (week 1)	Feacal Oocyst count after treatment (wk 2)	Percentage Reduction (wk 2)
A	21.66^{a}±0.33	3.00^{b}±0.00	86.10	1.66^{b}±0.33	92.30
B	6.67^{b}±0.33	2.33^{c}±0.33	65.06	0.00^{c}±0.00	100.00
C	5.33^{c}±0.30	5.66^{a}±0.33	NO	11.67^{a}±0.33	NO

^{a,b,c,} are Means in the same raw with different superscripts are significantly different, A: Commercial feed+0.05% turmeric paste+ infection, B: Commercial feed+1g/kg turmeric paste + infection, C: Commercial feed+ infection, *Means significant at $P< 0.05$.

Discussion

Herbal remedies have been used since ancient times in medicine and have recently gained increasing popularity, especially because of the declining effectiveness of synthetic compounds and concerns of the general population about drug side effects and interactions [29]. In chicken coccidiosis, herbal extracts have been intensively studied in the recent years in the search for new alternatives to the traditional anticoccidial drugs [30]. The extensive use of anticoccidials in the poultry industry may lead to the occurrence of drug residues in meat and eggs [31]. As such, consumer interest in organic foods has been rapidly increasing in recent years. The organic requirements restrict the use of chemicals, so natural plant products may represent an effective solution for pathogen control in the organic poultry system [32].

There was statistical difference between the groups treated with TP and the control thus depicting that curcumin in TP has anticoccidial effects. This work agrees with the work done by Aranjo and Leon [33] and Kuchi *et al.* [34]. Also there was statistical difference between the two treated groups and this showed that increased dose of TP at 1g/kg gave better efficacy or effects than 0.5g/kg dose. The results showed that the group treated with 1g/kg of TP was able to combat coccidia infection and overcome it faster and completely within 2 weeks more than the group treated with 0.5g/kg of TP and this may be as a result of an increased dose of TP in that group. The mean faecal coccidia oocysts counts 2 weeks post infection and post treatment of the group treated with 1g/kg of TP was significantly lower than that of the other group treated with 0.5g/kg of TP. The group was completely cleared of the infection (coccidiosis) which is evidenced by zero faecal coccidia oocysts counts by 2 weeks after treatment while that of the infected-untreated group did not reduced but continued to increase showing that the group has no protection against coccidiosis. This effects agrees with the work of Allen *et al*., [35] that curcuma reduced oocyst shedding

The bioactive compounds in Tumeric paste are curcumin (turmeric), piperine (black pepper) and fats (extra virgin olive oil). These three compounds worked together to yield the anticoccidial effects we observed in this work. Curcumin in turmeric powder alone will not be able to give this effect owing to its inability to be absorbed through the gastrointestinal tract [21]. For this reason, piperine and fats were used to prepare the paste to enhance inter-cellular permeability and uptake of Curcumins [26].

Curcumins has a direct effect on the parasites, by altering the process of oocyst wall formation and inhibiting the formation of sporocysts [36], or by destroying the sporozoites [15]. Furthermore, there is a lower risk of developing resistance to these natural substances compared to anticoccidial drugs [37]. Furthermore, turmeric paste could improve recovery after coccidiosis [38].

Turmeric paste was effective in controlling coccidiosis in chickens more than the turmeric powder and can be used successfully as a natural anticoccidial and it gives better effects at 1g/kg body weight.

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