EFFECTS OF PHALLANTUS AMARUS IN THE TREATMENTS OF COCCIDIOSIS IN BROILER CHICHEN

BY

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Abstract

The development of drug resistance of Eimeria spp. in poultry necessitated chemotherapeutic control alternatives from plant extracts as potential solution. This study investigated in-vitro evaluation was studied on the anti-coccidial properties from P. amarus leaf extracts and its effects on growth performance and carcass characteristics of broiler chicken challenged with Eimeria oocysts. Phytochemical screening of the plant extracts revealed the presence of flavonoids, cummarin, glycoside, triterpenes and alkaloids while saponin, tannin, phenolics, steroid, anthocyanins, terpenoid, amino acids and phylobatanins were negative. In the In-vitro trial, the effects of P. amarus on the inhibition of the sporulation of oocysts of Eimeria spp. were undertaken. The experimental design was complete randomized design (CRD) consisting of 2 treatments with 3 replicates each with control. T1 received 5 mL of P. amarus extracts, T2 received 10 mL of P. amarus leaf extract, and T0 is control. The chicks of all the groups were inoculated orally with sporulated oocysts on day 28 (4th week), 7 days post-infection, the birds were treated with different regimens, (5 & 10mL) of P. amarus leaf extracts for 3 consecutive days with the exception of T0. Oocysts clearance showed a decrease in the amount of oocysts in the feaces of all the groups treated with P. amarus leaf extract. The feed intake, body weight gain and feed conversion ratio were significantly improved in all treatment groups in comparison with the infected but not treated broiler chickens. 5 mL of P. amarus extract had sporulation inhibition percentage of 58% at 72 hours, 10 mls of P. amarus had sporulation inhibition rate of 73% while 15 mL of P. amarus has higher sporulation inhibition of 84% at 72 hours. In conclusion, the study showed that 15mls of ethanoic extract of P. amarus was effective in control of coccidiosis caused by Eimeria infection in broiler chicken.

Keywords: Coccidiosis, Coccidiostat, Eimeria oocysts, Phyllantus amari and chemotherapeutic

Introduction

It is common with poultry birds that they contact diseases when the farmer did not keep all or part of the hygiene practice, when poultry birds contacted disease and became sick, it is a loss to the farmer when the cost of treatment or even mortality is considered Lawal,et, al., (2022). Treating poultry birds with antibiotics is common but it comes with some challenges and this include drug resistance, side effect and residual effect to birds and final consumers. The FAO and WHO has advised strongly against the use of antibiotics for poultry birds because of these challenges and therefore both Veterinarian and Animal scientist has faced the challenges to find an alternative for the use of antibiotics, especially usage of plant part as a antibiotics comes with nom side effect and gives good result and this is why *Phalantus amari* comes to mind because of its Phytochemical constituents. Coccidiosis is an infective disease of many species of mammals and birds caused by protozoa of the Phylum Apicomplexa which causes diarrhoea, retarded growth, a slower feed conversion and increased mortality especially in chickens a (Maikai et al., 2007; Fornace et al., 2013)

The disease is caused by parasites of the genus Eimeria,Isospora and Cryptospora with a complex life cycle, affecting mainly the intestinal tract, especially in chicken it is one of the most serious diseases in chicken production as economic losses are possible even before the manifestation of clinical signs of the disease (Dalloul and Lillehoj, 2006). These parasites cause damage in chicken production, chickens reared in large numbers and high densities

(Peek, 2010). Management has always been important to control coccidiosis in poultry, however, it is very difficult to keep chickens coccidia free as oocytes are omnipresent and spread widely in the poultry house, and requires the administration of various drugs through feed and water. Attempt has recently been taken to reduce the cost of feed, including the incorporation of agro-industrial by products in broiler diets as an energy source (Sugiharto and Rajitkar, 2019).

However, some limitations may exist when using the agro-industrial by-products as the ingredients in broiler rations. The high and low contents of fibre and protein in the by products may limit the digestibility and thus inclusion level of such by-products (Sugiharto et al., 2018). It is known that some foliage contain a number of bioactive compounds that are beneficial for the health of chickens (Rama-Rao et al., 2019). These compounds include vitamins, phenolic acids, flavonoids, isothiocyanates, tannins as well as saponins (Vergara Jimenez et al., 2017). The use of medicinal plants in animal production has increased research interests as a potential substitute for antibiotics (Lillehoj et al., 2018). In this regard, the use of leaf meal in rations may not only reduce the cost of feeds, but also elicit the health-promoting effect on broiler chickens (Sugiharto et al., 2019). Phyllanthus amarus is widely distributed in all tropical and subtropical regions of the planet (Edeoga et al., 2006). In Nigeria, it is called "Oyomokeisoamank edem" in Efik, "Iyin Olobe" in Yoruba and "Ebebenizo" in Bini (Etta, 2008). P. amarus has various groups of compounds such as alkaloids, flavanoids, hydrolysable tannins, major lignans and polyphenols (Peters et al., 2015). Lignans like phyllanthin and hypophyllanthin, flavonoids like quercetin were isolated from the leaves of P. amarus (Meena et al., 2018). Phyllanthus amarus is a plant of the family Euphorbiaceae and has approximately 800 species which are found in tropical and subtropical countries of the world (Tahseen and Mishra, 2013). The name 'Phyllanthus' means "leaf and flower" and named so because of its appearance where flower, fruit and leaf appears fused (Kumar et al., 2011). Its effect in excretory system is due to its antiurolithic property and is used in the treatment of kidney/gallstones, other kidney related problems, appendix inflammation and prostate problems (Sen and Batra, 2013; Ushie et al., 2013). P. amarus like other tropical tree leaves contains some bioactive compounds which may affect nutrient utilization, haematological and serum biochemical parameters in animals (Ogbuewu et al., 2010).

The anti-nutritional compounds contained in P. amarus include oxalate, phytate, hydrogen cyanide, nitrate and tannin (Gafar et al., 2012). Herbal medicines are believed to be safer than synthetic medicine because phytochemicals in the plant extract target its biochemical pathways (Sandigawad, 2015). Evaluation of haematological parameters are not only used to determine the extent of deleterious effect of herbal extracts, but also to explain functions of plant extracts or their products on the blood of animals (Bin Jaliah et al., 2014). Therefore, the present study examined the effect of haematology and serum biochemistry of broiler chickens inoculated with Salmonella Enteritidis and treated with Phyllanthus amarus leaf extract rinsed with distilled water and air dried under shade until they were crispy to touch, while still retained their green colour. The leaves were ground with domestic electric grinding machine (SonikR Model SB-464) to produce P. amarus leaf meal (PALM). The PALM was subjected to 80% methanol extract maceration technique by putting one hundred grams (100 g) of the PALM in 500 mL of 80% methanol at room temperature for 72 h while shaking intermittently wit rotary shaker. The extract was filtered through a muslin cloth and Whatman No. 1 filter paper and funnel. The extract was placed in a beaker and evaporated by placing it inside the water bath at 45 °C for 3 days to obtain a thick concentration.

Justification

Herbal plants are available all year round since it is sources locally, they are rich in antioxidant ability, with reasonable quantity of vitamin and Minerals, has no side effect and the problem of drug resistance has been removed and it is cost effective, so it is better than the use of antibiotic.

Objectives of the study

i-Prepare the extract of Phyllantus amari
ii-Prepare the extract of Eimeria oocysts

iii-Feed the extract to the birds
iv- Monitor the response of the infected birds treated with *Phyllantus amari* on Feed intake, wight gain and feed to weight gain

Material and methods

The experiment was carried out in the Agricultural garden of Kwara State Polytechnic, Ilorin. Leaves of P. amarus were harvested from the Polytechnic campus in Kwara State. The plant materials were dried under shade for one week. The dried leaves were milled to fine powder using a milling machine. The fine powder was placed in a thimble made from thick filter paper which was loaded unto the main chamber of the soxhlet extractor. The solvent (250ml of ethanol) was added to a round bottom flask which is attached to the soxhlet extractor and condenser. The solvent was heated and as it boils its vapors rises up and are condensed by the condenser. The condensate then drips into the reservoir containing the thimble. Once the level of solvent reaches the siphon it pours back into the flask and the cycle begins again. Embazine forth was the conventional anti-coccidual drug used. Extraction of Eimeria oocyst About 20g of dry infected droppings was homogenised in distilled water (100mL) and then soaked with 2.5% (W/V) of passions dichromate and allowed to stay for 24hrs. The mixture was filtered using muslin cloth, the residue was discarded and the filtrate was further treated with saturated sodium chloride for precipitation, the mixture was centrifuged at 1500 rpm for 30min. The tubes were removed and the supernatant was discarded. The sediment (occyst) transferred into distilled water for floatation to take place. After floatation, it was then centralized and the supernatant discarded while the residues which are the occyst collected and kept in a plastic material until ready for use.

The solution was pipetted to fill both sides of the Mc Master slide and was examined under the microscope at a magnification of 40X. A total of 200 day old Ross 308 strain chicks were purchased from Agrited farm Kaduna and used for the experiment and Eimeria infection. A week to the arrival of the animals on the experimental site, the pen with the cages was thoroughly washed and cleaned using disinfectant (lysol) and allowed to dry. Heat was provided for the birds day and night for the first two weeks of their age. The birds were fed ad libitum on a proprietary broiler ration without coccidiostat additives and were also given access to water ad libitum. They were routinely vaccinated against Gumboro and Newcastle diseases. At 27day old, the birds were inoculated orally with sporulated oocysts, they were starved with water the night before infection to enhance quick intake of the water. The prepared sporulated Eimeria ooycst was added into their drinking water. After a successful infection the birds were transferred to their respective cages and were assigned randomly to treatment groups. Seven days post infection when the birds started to show clinical signs of coccidiosis, the P. amarus treatment was administered to the birds. The data were analyzed using statistical package software (SPSS version 23). Differences in mean body weight were tested by one-way ANOVA.

Result and Discussion

The initial body weight of the treatments shows no significant ($P<0.05$) difference across the treatment's samples, while for the final weight of the sample treatments shows T1 was significantly ($P<0.05$) different across all the sampled treatments. while for the final weight of the sample treatments shows T1 was significantly ($P<0.05$) different when compared with rest sample treatments with a superior weight of 2205.20g while the lowest final weight was observed in T5 (1950.30g). A similar trend was observed in the body weight gain, daily body weight, feed intake, daily feed intake and feed conversion ration with T1 observed to be significantly ($p<0.05$) different when compared to the rest samples. However, there was no significant ($p>0.05$) different across all the sampled treatments, the bcontrol of the experiment is the least in all the values obtained in the experiment.

The findings of this study indicate that different concentrations of P. amarus ethanol leaf extract have anticoccidial activities as evidenced by their ability to decrease ($p < 0:05$) significantly the sporulation of unsporulated. Eimeria oocysts compared with the infected control incubation. This implies that extract reduced the growth and

development of oocysts to become the infective stage of Eimeria (sporozoites). P. amarus had sporulation inhibition percentage of 58% at 72hrs, 10mL of P. amarus had sporulation inhibition of 73% while 15mL of P. amarus had higher sporulation inhibition of 84% at 72hrs.

Table 1: Growth performance of broiler chicken challenged with Eimeria oocysts and treated with Phyllantus amari.

Items (g)	T0	T1	T2	SEM
Initial body weight	590.50[b]	620.00[a]	555.00[c]	0.44
Final body weight	1960.40[c]	2210.22[a]	2170.80b	0.21
Body weight gain	1400.70c	1590.89b	1640.60[a]	0.75
Daily weight gain	20.70[c]	27.50[a]	26.99[b]	0.01
Total feed intake	2590.20[c]	2700.00[b]	2840.08[a]	0.16
Daily feed intake	40.50$_c$	45.77[b]	47.34[a]	0.01
Feed conversion ration	1.83	1.22	1.30	
Mortality rate (%)	9	5	4.5	

Conclusion and recommendation

This present study revealed that the use of Phyllathus amarus leaf extract (PALE) to treat Eimeria infected or challenge broilers significantly inhibited the sporulution of Eimeria oocysts. The feed intake, body weight gain and feed conversion ratio were also significantly improved in all treatment groups in comparison with the infected but not treated broiler chickens. The use of 15mL P. amarus had the best final weight and growth statistics as such it is recommended. Further research should be carried out to determine the active compound in Phyllathus amarus leaf that is useful in treatment of other diseases. The use of high dosage of PALE in the treatment of Eimeria oocysts can be further investigated.

References

Agbaje, EO. Adeneye, AA. & Daramola, AO (2009). Biochemical and toxicological studies of aqueous extract of Syzigium aromaticum (L.) Merr. & Perry (Myrtaceae) in rodents. Afr. J. Tradit. Complement. Altern. Med. 6(3): 241-54.

Ajit, S, Doley, P, Neeraj, P & Prasad, J (2014). Effect of dietary supplementation of tulsi (Ocimum sanctum) leaf powder on haematology and serum biochemistry of broiler chicks. Int. J. Biol. Pharm. Res. 5(1): 89 - 92.

Akande, T.O; Odunsi, A.A; Rafiu, T.A; Olaniyi, C.O; & Binuomote, R.T (2013). Growth and serological assessment of broiler chickens fed differently processed castor (Ricinus communis Linn.) kernel cake based diets. Afr. J. Agric. Res. 8 (41): 5161 5165.

Akande, TO; Odunsi, AA; Rafiu, TA; Olaniyi, C.O, & Binuomote, RT (2013). Growth and serological assessment of broiler chickens fed differently processed castor (Ricinus communis Linn.) kernel cake based diets. Afr. J. Agric. Res. 8(41): 5161 5165.

Arunsi, UO; Chinyere, GC; Ngwogu, KO; Ngwogu, AC; Atasie, OC; Oti, UA; Akojuobi, JK; Udeogu, C; & Ibe, C (2020). Evaluation of the biochemical, haematological and histopathological parameters of female Wistar rats fed with aqueous and ethanol extracts of Aspilia africana leaves. J. Herbmed Pharmacol. 9(3): 257-267.

Babatunde, SK; Abubakare, AA; Abdulraheem, YJ; & Ajiboye, EA (2014). Antimicrobial activity of Phyllanthus amarus on some human intestinal facultatively anaerobic flora. Inter. J. Med. Biomed. Res. 3(1): 52-57.

Bin-Jaliah, I; Dallak, MA; Al-Hashem, FH; Nwoye, LO; Sakr, HF; & Jamil. AM (2014). Derangement of hemopoiesis and hematological indices in Khat (Catha edulis)-treated rats. Afr. J. Biotechnol. 13(2): 349-55.

Bowes, V.A; Julian, R.J; & Stirtzinger, T (1989). Comparison of serum biochemical profiles of male broilers with female broilers and White Leghorn chickens. Canad. J. Vet. Res. 53(1): 7 11.

Chanda, S; Parekh, J; Vaghasiya, Y; Dave, R; Baravalia, Y; & Nair, R (2015). Medicinal plants - from traditional use to toxicity assessment: a review. Inter. J. Pharmaceut. Sci. Res. 6(7): 2652 70.

Chechik, BE; Sengupta, S; & Fernandes, B (1986). Novel heterophile chicken antigen: Immunohistochemical localization using antisera to Mycobacterium smegmatis and possible association with lymphocyte Histochem. J. 18: 36-40.

Dalloul, R. A., & Lillehoj, H. S. (2006). Recent advances in immunomodulation and vaccination strategies against coccidiosis. Avian Dis. 49:1-8.

Fornace, K. M., Clark, E. L., Macdonald, S. E., Namangala, B, Karimuribo, E., Awuni, J. A, Thieme, O., Blake, D. P. & Rushton, J. (2013). Occurrence of Eimeria species parasites on small- scale commercial chicken farms in Africa and indication of economic profitability. PLoS ONE, 8(12): 1-8.

Maikai, V. A., Makeri, H. K., Adeiza, A. A., & Maikai, B. V. O (2007). Preliminary studies of anticoccidial effect of Mahogany (Khaya senegalensis) and African Locust Bean Tree (Parkia biglobosa) aqueous bark extracts on chicken infected with coccidia. Savannah Journal of Agriculture, 2: 43-45.

Peek, H. W & Landman, W. J. M. (2010). Resistance to anticoccidial drugs of Dutch avian Eimeria spp. field isolates originating from 1996, 1999 and 2001. Avian Pathol, 32: 391–401.

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