

Efficacy of *Phyllanthus amarus* leaf on growth performance and carcass characteristics of broiler chickens challenged with *Eimeria* oocysts

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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 5 treatments with 4 replicates each. T1 received 5 mL of *P. amarus* extracts, T2 received 10 mL of *P. amarus* leaf extract, T3 received 15 mL of *P. amarus* leaf extract, T4 was positive control using Embazine for the and T5 negative 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, 10 and 15 mL) of *P. amarus* leaf extracts for 3 consecutive days with the exception of T4 and T5. Oocysts clearance showed a decrease in the amount of oocysts in the faeces 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: Inoculated, performance, *Phyllanthus amarus*, sporulation and treatments

Running title: Growth performance and carcass characteristics of broiler chickens challenged with *Eimeria* oocysts



Efficacité des feuilles de *Phyllanthus amarus* sur les performances de croissance et les caractéristiques de carcasse des poules de chair confrontées aux oocystes d'*Eimeria*

Résumé

Le développement de la résistance aux médicaments des *Eimeria* spp. chez les volailles a nécessité des alternatives de contrôle chimiothérapeutique à partir d'extraits de plantes comme solution potentielle. Cette étude a examiné l'évaluation in-vitro des propriétés anti-coccidiennes des extraits de feuilles de *P. amarus* et ses effets sur les performances de croissance et les caractéristiques de carcasse des poules de chair confrontées aux oocystes d'*Eimeria*. Le dépistage phytochimique des extraits de plantes a révélé la présence de flavonoïdes, de coumarine, de glycosides, de triterpènes et d'alcaloïdes, tandis que les saponines, les tanins, les phénoliques, les stéroïdes, les anthocyanines, les terpènes, les acides aminés et les phylobatanins étaient négatifs. Dans l'essai in-vitro, les effets de *P. amarus* sur l'inhibition de la sporulation des oocystes d'*Eimeria* spp. ont été étudiés. Le design expérimental était un design complètement aléatoire (DCA) comprenant 5 traitements avec 4 répliques chacun. T1 a reçu 5 mL d'extrait de *P. amarus*, T2 a reçu 10 mL

d'extraire de feuilles de *P. amarus*, T3 a reçu 15 mL d'extraire de feuilles de *P. amarus*, T4 était un contrôle positif utilisant Embazine, et T5 un contrôle négatif. Les poussins de tous les groupes ont été inoculés oralement avec des oocystes sporulés le jour 28 (4^e semaine), 7 jours après l'infection, les oiseaux ont été traités avec différents régimes (5, 10 et 15 mL) d'extraits de feuilles de *P. amarus* pendant 3 jours consécutifs, à l'exception de T4 et T5. L'élimination des oocystes a montré une diminution du nombre d'oocystes dans les fèces de tous les groupes traités avec l'extraire de feuilles de *P. amarus*. L'ingestion de nourriture, le gain de poids corporel et le ratio de conversion alimentaire se sont significativement améliorés dans tous les groupes de traitement par rapport aux poules de chair infectées mais non traitées. 5 mL d'extraire de *P. amarus* ont eu un pourcentage d'inhibition de la sporulation de 58 % à 72 heures, 10 mL d'extraire de *P. amarus* ont eu un taux d'inhibition de la sporulation de 73 %, tandis que 15 mL d'extraire de *P. amarus* ont montré une inhibition plus élevée de la sporulation de 84 % à 72 heures. En conclusion, l'étude a montré que 15 mL d'extraire éthanolique de *P. amarus* était efficace dans le contrôle de la coccidiose causée par l'infection par *Eimeria* chez les poules de chair.

Mots-clés : Inoculé, performance, *Phyllanthus amarus*, sporulation et traitements

Introduction

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. 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. 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.

This parasitic disease is an expensive disease and is estimated to cost the poultry industry by 3.2 billion USD annually (Dalloul and Lillehoj, 2006). In Nigeria where poultry farming is less developed, the disease is more serious and causes heavy economic losses which could be in the region of millions of Naira (Maikai *et al.*, 2007;

Fornace *et al.*, 2013). The major part of these costs, around 80%, is due to the losses in growth performance, and the rest is due to the costs of prophylaxis and treatment (Vermeulen *et al.*, 2001). Coccidiosis infection led to decreased body gain and deteriorate feed conversion ratio, as well as increased incidence of diarrhea and intestinal hemorrhage (Williams, 2005). All of these symptoms are economically significant to the poultry industry. Additionally, Williams, (2005) found other pathophysiological effects associated with coccidiosis which include, poor feed efficiency, reduced water intake, increased intestinal passage time, decreased digesta viscosity, intestinal mal-absorption, villus atrophy, intestinal leakage of plasma proteins, and increased intestinal activity owing to the widespread occurrence of drug resistant parasites. Coccidiosis is generally controlled by using anticoccidial drugs which are administered in feed of chickens (Blake and Tomley, 2014; Shivaramaiah *et al.*, 2014) and use of live vaccines, new approaches of controlling coccidiosis are urgently needed especially natural plant extract to treat and control *Eimeria* (Innes and Vermeulen, 2006; Williams, 2006; Morris *et al.*, 2007).

Management focuses on decreasing coccidial numbers to keep infection at a minimum until

immunity is established in young birds proper hygiene use of anti-coccidial drugs and vaccines play major roles in controlling coccidiosis in commercial broiler chicken production. Coccidiosis is traditionally treated by chemotherapy but the appearance of drug-resistant types of coccidia suggests the need of using alternative strategies such as the natural products from plant extract.

P. amarus is a small erect tropical herb that grows to a height of 10-60cm. It is an annual and widespread throughout the tropics and subtropics. It is found in sandy regions as a weed in cultivated and wastelands. The plant can be found along roads, valleys, on riverbanks and near lakes.

The broad objective of this study was to evaluate the efficacy of *P. amarus* leaf extract as alternative anticoccidial for broiler chicken production. While the specific objectives were to determine the effect of *P. amarus* leaf extract treatment on growth performance and carcass characteristics of broiler chickens infected with *Eimeria* oocysts.

Materials and Methods

Experimental site

The research is divided into two parts; the *in vitro* test was done at the Animal Production Laboratory Federal University of Technology Minna, and the *in vivo* was done at the Poultry Section of the skill Development Centre Niger State Agricultural and Mechanization Development Authority Minna. (Lat 9.63424° and Long 6.57875°) elevation 311, Bosso Local Government, Niger State, Nigeria.

Preparation of *P. amarus* extracts

Leaves of *P. amarus* were harvested from the NAMDA Demonstration Farm Minna, Niger 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-coccidial 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 potassium 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 (oocyst) 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 oocyst 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

To calculate the number of oocyst per gram of faeces, the formula is;

Number of oocyst counted x Total volume of salt solution used (12ml)

Weight of feces (2g) 2(0.15mL)

In-Vitro Experimental Design and Oocyst Sporulation Inhibition Percentage

The experimental design used in the present study was approved by Animal Production Laboratory of the Federal University of Minna. An *in vitro* sporulation inhibition trial was used to examine the effect of *P. amarus* on oocysts sporulation of different *Eimeria* species. In this trial, unsporulated oocysts were put in petri dishes and

exposed to three concentrations of *P. amarus* each (5,10 and 15mL) while Embazine forth were used as positive control. The sporulation of the oocysts was confirmed by examining sporocysts under inverted microscope at 40X every 12hrs to attain the sporulation inhibition percentage with time. The numbers of sporulated and unsporulated oocysts were counted and the percent sporulation was estimated by counting the number of sporulated oocyst over the total number of oocysts per concentration.

$$\text{Sporulation inhibition percentage} = \frac{\text{No. of sporulated oocyst}}{\text{Total no. of oocyst}} \times 100$$

Experimental birds and management

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* oocyst 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.

Data collection

Feed Intake

Daily feed offered to the animals and the corresponding remnants were measured and recorded during the experimental period. The daily remnant was subtracted from the offered feed to determine the intake. Feed offered-feed leftovers on daily bases

Body Weight Gain

The birds were weighed at the beginning of the challenging and subsequently on a weekly basis for the assessment of the growth rate using a spring balance scale. The average weight was recorded for each treatment group against its identification number on a weekly register book. The average daily weight gain (ADWG) was determined by dividing weight gain by number of birds and the total number of days of the experiment.

Weight gain = Final body weights – Initial body weights

Feed Conversion Ratio

The feed conversion efficiency of experimental animals was determined by dividing the average weekly body weight gain to the amount of feed consumed by the animal each week.

$$\text{feed conversion ratio} = \frac{\text{feed intake (g)}}{\text{weight gain (g)}}$$

$$\text{Average daily weight gain} = \frac{\text{average final weight gain} - \text{average initial weight}}{\text{Number of dayss}}$$

$$\text{Average daily feed intake (g)} = \frac{\text{quantity of feed served (g)} - \text{quantity of feed leftover (g)}}{\text{Number of birds}}$$

$$\text{Mortality rate} = \frac{\text{No of dead birds}}{\text{No of stocked birds}} \times 100$$

Oocyst clearance

Faecal samples were collected from the onset of successful infection (day zero) till the 5th day after treatment. Pooled daily oocyst counts per group were undertaken. Individual oocyst counts were not feasible because the birds were housed in group. A total of 10 g of feces was collected daily beginning from a day before treatment until the seventh day post treatment. The modified McMaster technique as described by Vassilev (2002) was used to estimate Oocyst per gram (OPG). This was done for the three replicates and the mean value was calculated.

Carcass characteristics

At the end of the experiments two birds per each replicate were weighed, slaughtered and dressed. The viscera and cut up parts were weighed and the weight where calculated as the percentage of the live weight.

Statistical Analysis

The data were analyzed using statistical package software (SPSS version 23). Differences in mean body weight were tested by one-way ANOVA.

Mean separation was done using Duncan multiple range test module of the same statistical software. $p < 0.05$ was considered as significant level.

Results and Discussion

Results

Growth performance of broiler chicken challenged with Eimeria oocysts

the initial body weight of the treatments shows no significant ($p < 0.05$) difference across the treatments samples, 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.

Table 2: Growth performance of broiler chicken challenged with Eimeria oocysts

ITEMS (g)	T1	T2	T3	T4	T5	SEM	LSD
Initial body weight	619.23 ^a	553.21 ^b	568.07 ^c	601.00 ^d	586.47 ^e	0.74	*
Final body weight	2205.20 ^a	2167.70 ^b	2083.4 ^c	2297.10 ^d	1950.30 ^e	2.21	*
Body weight gain	1585.90 ^a	1614.50 ^b	1515.40 ^c	1696.10 ^d	1363.80 ^e	2.78	*
Daily weight gain	26.43 ^a	26.90 ^b	25.25 ^c	28.26 ^d	22.73 ^e	0.05	*
Total feed intake	2746.10 ^a	2840.08 ^b	2660.07 ^c	2896.2 ^d	2489.2 ^e	0.61	*
Daily feed intake	45.77 ^a	47.34 ^b	44.34 ^c	48.27 ^d	41.48 ^e	0.01	*
Feed Conversion Ratio	1.75 ^a	1.76 ^b	1.76 ^c	1.71 ^d	1.83 ^e	0.00	*
Mortality (%)	5	4.5	4.5	4	9		NS

NS = Not significant

Means in the same row with different letters are significantly different at $P < 0.05$.

SEM = Standard error of mean

LSD: Least Significant Difference

Cut-up parts of broiler chickens challenged with EIMERIA oocysts

The weight of the various cut-up parts of broilers chicken as weighed showed that there was

significant ($p < 0.05$) difference between T1 for live weight when compared to the rest samples treatment while T3 observed where the lowest live weight of 1.82kg. The slaughter weight of the

various sample weight shows T1 and T4 to be not significantly ($p < 0.05$) different to each other but they are significantly different when compared with rest treatments. The dress weight on the other hand shows T1, T2, T3 and T4 to be non-significantly different to each other with values of 1.96, 1.82, 1.65 and 1.78kg respectively, while T5 appear to have the highest value of 2.57g making significantly different at ($p < 0.05$) when compared with rest samples. However, a different pattern was observed for eviscerated weight with T2 to the significantly different at ($p < 0.05$) when compared with other sample treatments which the remaining samples were not significantly different to each. The carcass weight also shows a significant difference of ($p < 0.05$) when compared to the rest of the sample treatments. The wings and breast meat all shows a significant difference ($p < 0.05$) when compared to the other

samples with highest value of (166.00g and 447.09g) while the lowest weight for both parameters was observed in T4 (191.37g and 380.63g). The thigh weight shows T5 to be significantly ($p < 0.05$) different when compared to the other samples and have a superior value of value of 247.21g while T3 had the lowest value of 195.56g. The drum stick of the various sample shows no significant ($p > 0.05$) difference between T1, T2, T3 and T4 when compared to each other but they vary significantly at ($p < 0.05$) when compared to T5. Shank weight of the sample treatment shows no significant difference across all the samples. However, the back weight of T5 shows a significant ($p < 0.05$) difference when compared to the rest sample treatments having a superior value of 284.91g which the least back weight was seen in T3 (217.19g).

Table 3: Cut-up parts of broiler chicken challenged with *Eimeria* oocysts

ITEMS (g)	T1	T2	T3	T4	T5	SEM	LSD
Live weight (kg)	2.10 ^a	1.98 ^b	1.82 ^c	1.97 ^b	2.00 ^b	0.06	*
Dress weight (kg)	1.96 ^b	1.82 ^b	1.65 ^b	1.78 ^b	1.97 ^a	1.14	*
Dress weight (%)	93.33 ^b	91.91 ^b	90.65 ^b	90.35 ^b	98.05 ^a	2.13	*
Eviscerated wt (kg)	1.75 ^{ab}	2.65 ^a	1.56 ^b	1.65 ^b	1.66 ^b	0.46	*
Carcass wt. (kg)	1.66 ^a	1.50 ^{ab}	1.44 ^b	1.52 ^{ab}	1.50 ^{ab}	0.08	*
Breast (%)	22.81 ^a	29.02 ^{ab}	27.61 ^b	25.04 ^b	25.88 ^b	23.25	*
Drumstick (%)	11.52 ^a	12.60 ^a	12.01 ^{ab}	11.93 ^{ab}	11.14 ^b	10.07	*
Thigh (%)	13.49 ^{ab}	14.66 ^b	13.58 ^c	15.44 ^{ab}	16.48 ^a	11.61	*
Wings (%)	10.00 ^a	11.25 ^a	12.17 ^a	12.59 ^{ab}	14.21 ^b	18.19	*
Back (%)	13.55 ^b	15.13 ^b	15.08 ^b	14.66 ^b	18.99 ^b	8.95	*

Internal organs of broiler chicken challenged with Eimeria oocysts

The internal organ of the various treatments was weighed where the liver, heart and the lungs all show no significant ($p > 0.05$) difference between all the samples when compared to each other. The gall bladder of the sample treatments shows T2 was significantly ($p < 0.05$) different compared with other sample Treatments with a higher value of 2.76g while T1 and T5 had the lowest values of 1.98g and 1.50g respectively. However, no

significant ($p > 0.05$) difference exist between T1, T2, T3 and T4 for spleen weights but a significant ($p < 0.05$) different exist between the sample Treatment with T5 which has the highest value of 8.90g. The abdominal fat content of the sample treatments shows no significant difference across the samples but for the full intestinal weight, a significant difference exist between T2 when compared to the rest samples with a superior value of 172.76g for full intestine, while T3 and T4 had the lowest values of 118.49g and 113.56g

respectively. The empty intestine, gizzard, proventriculus and pancreas all show no

significant ($p>0.05$) difference when compared to each other.

Table 4: Internal organs of broiler chicken challenged with *Eimeria* oocysts

PARAMETERS	T1	T2	T3	T4	T5	SEM	pValue
Live weight (kg)	2.10 ^a	1.98 ^b	1.82 ^c	1.97 ^b	1.95 ^b	0.06	*
Liver (%)	1.56	1.47	1.55	1.41	1.53	0.9	NS
Heart (%)	0.26	0.28	0.49	0.32	0.40	3.21	NS
Lung (%)	0.30	0.41	0.37	0.36	0.07	83.62	NS
Gall bladder (%)	0.07 ^b	0.13 ^a	0.10 ^{ab}	0.10 ^{ab}	0.10 ^{ab}	0.49	
Spleen (%)	0.07 ^b	0.08 ^{ab}	0.08 ^b	0.08 ^b	0.45 ^a	2.10	*
Abdominal fat (%)	1.68	2.07	1.98	1.77	1.68	4.99	NS
Full intestine (%)	6.19 ^b	8.72 ^a	7.84 ^b	6.01 ^c	5.82 ^c	3.77	*
Empty intest. (%)	6.05	6.65	6.94	7.01	7.36	24.04	NS
Gizzard (%)	12.06	12.60	12.36	11.09	11.24	68.64	NS
Proventriculus (%)	5.62	5.85	5.72	6.25	6.67	41.03	NS
Pancreas (%)	4.69	4.94	4.92	4.73	4.44	33.71	NS

Discussion

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. In agreement with the current findings, *in vitro* effects of *Aloe vera* and *Aloe spicata* on the inhibition of the sporulation of avian coccidian oocysts were reported by Mwale *et al.*, (2006). The authors found that *Aloe spicata* had higher sporulation inhibition efficacy at 30% concentration (65.42%) while *A. vera* had the least number of sporulated oocyst at 45% concentration with an inhibition percentage of 62.5%. The reduction in oocysts counts observed in this study is in agreement with the findings of

Elhkany *et al.*, (2011) who reported 80% sporulation inhibition effects of garlic powder. Silva *et al.*, (2005) evaluated the performance of broilers reared in environments with and without oocysts, the authors observed a lower body weight and a lower feed at 14 days of life conversion. Persia *et al.*, (2006) reported that the coccidiosis infection can lead to a drop in performance due to decreased dietary metabolisable energy, and digestibility of amino acids.

In the current study after challenging chickens with *Eimeria* species and then treated with *Phyllanthus almarus* ethanol leaf extracts, resulted in improved feed intake, body weight gain, and feed conversion ratio. There was a significant improvement in growth performance of chickens treated with synthetic drugs (*Embazine forte*) and *P. amarus* plant extracts. However, the performance of the infected not treated chickens reduced when compared with others. The body weight gains in the group treated with embazine forte had the highest weight gain when compared with others, while performance characteristics of treatment with 15mL of *P.*

amarus was found to be similar to those treated with embazine forte. The increase in weight gain can be attributed to the decrease in the number of *Eimeria* oocyst in the intestine. The improvement in BW of birds treated with *P. amarus* could also be attributed to the decrease in oocyst shed in the droppings, which translates to reduction of damage to the intestinal epithelium by the sporozoites of the parasite. The reduction in the damage to the intestinal lumen of the birds treated with the extract could be responsible to the decrease in blood/stained diarrhea observed in the treated groups. These findings are in agreement Du and Hu (2004), who observed significant higher body mass gains and mild lesion score in birds medicated with herbal complex for *E. tenella* infection in chicken in comparison with infected control group. Similarly, Chandrakesan *et al.* (2009) observed that the herbal complex can cause better body mass gain between the 4th and 5th weeks and superior feed conversion ratio after infection with *E. tenella*. Arczewska-Wlosek and Swiatkiewicz (2012) observations were also similar to the present findings that significant lower body weight gain in groups were infected with *Eimeria* oocysts due to adverse effect on the digestion, absorption and assimilation of nutrients.

Using the fecal oocysts output of chickens as the indicator of effect of plant extracts, the present findings showed significant reduction in oocysts output for *P. amarus* ethanol extract treatment in comparison with infected and not treated group. Jang *et al.* (2007) evaluated the anticoccidial effect of green tea-based diets against *E. maxima*, observed that the green tea-fed chickens produced significantly reduced fecal oocysts when compared to the *E. maxima* infected group

fed standard diet. Arczewska-Wlosek and Swiatkiewicz (2012) reported that a dietary herbal complex consisting of the blend of *Salvia officinalis*, *Allium sativum*, *Thymus vulgaris*, *Echinacea purpurea* and *Origanum vulgare* in broiler chickens was effective against many species of *Eimeria* in terms of reducing oocyst output. Kostadinovic *et al.*, (2012) induced coccidial infection in broiler chickens with *E. tenella* and the author showed that *Artemisia absinthium* extract reduced the severity of the coccidial infection and caused significant decrease in output number of oocysts per gram of faeces and also bloody diarrhea in the group treated with *A. absinthium* extract and was milder than in the other groups.

In overall, data from the current study suggest that 5, 10 and 15mL of *P. amarus* leaf extract can inhibit the growth of *Eimeria* in broiler chickens.

Conclusion and recommendation

This present study revealed that the use of *Phyllanthus amarus* leaf extract (PALE) to treat *Eimeria* infected or challenge broilers significantly inhibited the sporulation 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 *Phyllanthus 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.

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