

Comparative efficacy of commonly used anti-coccidial drugs in broiler chickens experimentally infected with *Eimeria* species

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Abstract

Poultry meat is a rich source of proteins, vitamins and minerals. Poultry production faces several challenges, one of which is the occurrence of poultry diseases such as coccidiosis. This present study investigated the efficacy of the most commonly used anti-coccidial drugs in broilers experimentally infected with *Eimeria* species. Forty two broiler chickens were used for the study. They were randomly assigned to six groups (A, B, C, D, E and F) of seven birds each. Chickens in Groups A, B, C, D and E were experimentally infected with *Eimeria* species on day 21 of age (100,000 sporulated *Eimeria* oocysts per/ml per chick), while Group F chickens served as the uninfected control. Three days post-infection, the infected groups were treated as follows: Group A – Sulphaquinoxaline + diaveridine combination; Group B – Diclazuril; Group C – Tortrazuril; Group D – Diclazuril + Tylosin tartrate combination; Group E – Infected untreated group. The treatment was done for the number of days specified by each drug's manufacturer. Observed clinical signs of coccidiosis, mortality, weight gain, mean faecal oocyst count, feed conversion efficiency, packed cell volume (PCV) and red blood cell (RBC) counts were determined and recorded across 18 days period post-infection. Results showed that treatment with all the anti-coccidial drugs used minimized coccidiosis related mortality, oocyst count per gramme of faeces, improved weight gain, PCV, RBC counts and feed conversion ratio, when compared to infected-untreated control. Broiler chickens in Groups A, B and C had the lowest mortality. Group B chickens (treated with Diclazuril) had the lowest faecal oocysts count among the infected groups and also the highest weight gain and better feed conversion than those of other infected groups, which suggests that diclazuril exhibited better efficacy in the treatment of experimental *Eimeria* infection in the broiler chickens.

Keywords: Broiler chickens; Coccidiosis; *Eimeria* species; Treatment; Anti-coccidial drugs.

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Introduction

Poultry meat, besides being rich in protein, is a good source of minerals and vitamins. It has been reported to contain less fat than beef and pork (FAO, 2017). Coccidiosis, a protozoan disease complex, is among the most frequently documented diseases of poultry in Nigeria (Corria *et al.*, 2021), with multiple *Eimeria* species reportedly co-infecting chickens (Agishi *et al.*, 2016). Everywhere chickens are raised, coccidiosis is common, and manifests as enteritis, bloody diarrhoea, and mortality [Ezema *et al.*, 2024]. Coccidiosis is reported to cost the Nigerian poultry industry £58.67 million annually, while the disease causes economic losses annually of almost £10 billion worldwide (Eze *et al.*, 2019).

The aetiology of coccidiosis involves multiple *Eimeria* species. Nine *Eimeria* spp are reportedly implicated in coccidiosis in broiler chickens, with *Eimeria tenella* and *Eimeria necatrix* as the most virulent (Eze *et al.*, 2019). Depending on the species of *Eimeria*, infective dose and site of infection, coccidiosis can result in limited enteritis with fluid loss and malabsorption of nutrients (*E. acervulina* and *E. mitis*), inflammation of the intestinal wall with pinpoint hemorrhages and sloughing of epithelia (*E. bruneti*, *E. maxima* and *E. mivati*) or complete villi destruction resulting in extensive haemorrhage and death (*E. necatrix* and *E. tenella*). Yet, the degree to which the different *Eimeria* species affect broiler performance and infection outcome are currently poorly understood (Kipper *et al.*, 2013, Gilbert *et al.*, 2020).

The prevalence of coccidiosis in poultry varies, depending on factors such as geography, management practices and age of the birds. The control options commonly adopted against coccidiosis include the use of vaccines, anti-coccidial drugs and stringent management practices (Blake *et al.*, 2020). The use of anti-coccidial drugs in combination with sanitation and hygiene is the most reliable and

widely used method of coccidiosis prevention and control (Noack *et al.*, 2019). The treatment, management, and control of avian coccidiosis is principally based on the use of synthetic (orthodox) anti-coccidial drugs. However, the use of synthetic drugs is accompanied by numerous challenges including resistance, high cost, treatment failures, withdrawal periods prior to slaughter, and residues in meat, eggs and the environment (Noack *et al.*, 2019; Ahmad *et al.*, 2024). Some of the most commonly used anti-coccidial drugs in Nigeria are sulphaquinoxaline and diaveridine combination, Toltrazuril, Diclazuril, and Amprolium.

Sulphaquinoxaline and diaveridine combination work by disrupting folate metabolism, effectively hindering DNA, RNA and protein synthesis in *Eimeria*, leading to cessation of the parasite life cycle (Chapman and Shirley, 1997). The drug can be administered through drinking water or through poultry feed. Diclazuril is a synthetic compound with a potent anti-coccidial properties. It belongs to a class of compounds known as triazine. Diclazuril works by interfering with development of *Eimeria* parasites within the host bird's intestine. It is highly effective in both preventing and treating coccidiosis in birds. Diclazuril has been reported to exhibit a strong anti-coccidial efficacy against a broad spectrum of *Eimeria* spp, making it a valuable tool for controlling coccidiosis in poultry (Lillehoj *et al.*, 2002); it is administered through drinking water. Toltrazuril is another triazine-based drug, which is widely used to combat coccidiosis in birds. Toltrazuril is known for its ability to disrupt the life-cycle of the *Eimeria* parasite, and it works as a coccidiostat in the control of avian coccidiosis, with reported efficacy against a variety of *Eimeria* spp (Chapman, 1999); it is administered through drinking water. Diclazuril + Tylosin tartrate combination is another anti-coccidial agent

that works by interfering with development of *Eimeria* parasites within the host bird's intestine; it is highly effective in both preventing and treating coccidiosis in birds, and it is administered through drinking water (Lillehoj *et al.*, 2002).

There is paucity of information in available literature on the comparative efficacy of these known and available anti-coccidial drugs in broiler chickens experimentally infected with *Eimeria* species. This present study investigated the comparative efficacy of the most commonly used anti-coccidial drugs in broiler chickens experimentally infected with *Eimeria* species.

Materials and Methods

Experimental birds and management: A total of forty two (42) day-old broiler chicks were used for the experiment. The birds were purchased from Agrited commercial hatchery, Ibadan, Oyo State, Nigeria. Brooding was carried out for a period of two weeks. They were fed with commercial poultry feed (Hybrid feed; broiler starter Mash, Premier Feed Mills Co. Ltd, Ibadan, Oyo State, Nigeria), and clean drinking water was provided *ad libitum* throughout the study. The broilers chicks were routinely vaccinated against Newcastle and Infectious bursal disease at 2 weeks of age. The experimental protocol was approved by the Institutional Animal Care and Use Committee of the Faculty of Veterinary Medicine, University of Nigeria, Nsukka (IACUC FVM UNN); with approval reference number: FVM-UNN-IACUC-2024-01/183. Guidelines for the humane handling of animals were followed all through the study. After the brooding, the broilers were weighed and assigned to separate cages (7 birds per cage) during the third week, by a restricted randomization procedure that approximately equalized initial weights.

Experimental Drugs: Sulphaquinoxaline and diaveridine combination (Embazine Fort®

Turner Right Ltd, Lagos, Nigeria) – this was administered at the dose of 0.6g/L in drinking water as follows: medicated water was administered for three days, followed by plain water for two days, and then medicated water for another three days (3-2-3), as prescribed by the manufacturer.

Diclazuril (Diclazor®, Hebei Kexing Pharmaceutical Co., Ltd, China); this was administered in drinking water at 1ml/2L for a period of 24 hours as prescribed by the manufacturer.

Toltrazuril (Kepro B. V. Holland) – this was administered in drinking water at a concentration of 25 mg/L for a period of 2 days as prescribed by the manufacturer.

Diclazuril + Tylosin tartrate combination (Bac-Cox® 7 Veterinary Tech. Co. Ltd, Enugu Nigeria), this was administered in drinking water for a period of 5 days as prescribed by the manufacturer.

Experimental Design: The 42 broiler chicks used for the study were randomly assigned to six groups (A, B, C, D, E, F) of seven birds each after week 2 of age. Group A were infected and medicated with Sulphaquinoxaline and diaveridine combination at 0.6mg/L; Group B were infected and medicated with Diclazuril at 1mg/L; Group C were infected and medicated with Tortrazuril at 1ml/L; Group D were infected and treated with Diclazuril + Tylosin tartrate combination at 1ml/L, Group E were then infected non-medicated (untreated) group. While Group F was the non-infected non-medicated control group. Broilers in groups A, B, C, D and E were infected with 100,000 sporulated *Eimeria* oocysts/ml per bird at day 21 of age. As from day 3 post infection, each broiler chicken in group A, B, C and D was treated with their group specific medications according to the manufacturer's specifications earlier stated above. Body weights, mean oocyst count per gramme of faeces, feed conversion ratio, packed cell volume (PCV), red blood cell (RBC) count and

mortality were recorded. Feed intake and body weight of the animals were recorded on daily basis. Faecal samples were also collected and observed every other day to enumerate the number of oocysts/g of faeces, for a period of 18 days. Blood samples (0.2 ml) were collected from the wing vein of each bird every three days for the determination of the packed cell volume (PCV) and for red blood cell (RBC) counting.

Oocyst output determination: The salt flotation technique (Levine 1973) was employed to detect and enumerate oocysts in the faeces of each bird. Oocyst per gramme of faeces (OPG) were quantified daily from day 3 post infection until the end of the experiment using the modified McMaster method.

Measurement of Body weights: Live body weights were recorded daily using standard weighing balance; the measurements were done every morning between 8.00 and 9.00 am before feeding. Body weight gain was calculated by subtracting the initial weight at the beginning of the experiment from the final weight at the end of the experiment.

Calculation of the feed conversion ratio: The daily feed consumption was obtained by subtracting the leftover feed after 24 hours from the total feed provided each day. The feed conversion ratio (FCR) for each group was calculated using the formula: $FCR = \text{Total feed intake} / \text{Total weight gain}$.

Packed cell volume (PCV) determination and Red blood cell (RBC) counting: Packed cell volume of the blood samples was determined by the microhaematocrit method (Coles, 1986), while the RBC counts of the blood samples was done by the haemocytometer method (Schalm *et al*, 1995).

Data Analysis: Data generated from the study were analyzed using a one way analysis of variance (ANOVA), and variant means were separated post-hoc using least significant difference (LSD) method. Significance was accepted at 0.05 probability level. Summary

results were presented in tables, line graphs and bar charts.

Results

The birds of infected groups A, B, C, D, and E showed clinical signs typical of coccidiosis such as inappetence, brownish diarrhoea, droopiness, ruffled feathers and death. Mortality was recorded in all the infected Groups (Table 1). In Groups A, B and C, only one broiler died in each of the groups on days 5, 11 and 6, respectively (Table 1), whereas two broilers died in Group D on day 5 (Table 1). In Group E (infected/untreated) all the seven broilers died on the following specified days post infection: two broilers on day 5, one broiler on day 7, two broilers on day 10 and two broilers on day 11 (Table 1). At necropsy of the dead birds, gross lesions typical of coccidiosis (ballooning and haemorrhages of the caeca and intestines and anaemia) were seen.

Following the infection of the birds, oocysts were first recorded in the faeces of the infected groups on day 3, but by day 4 post infection all the infected groups were shedding oocysts in their faeces. Oocysts numbers rose rapidly in the infected untreated group (E) and attained a peak level on day 7 post-infection (Figure 1). A similar rise was noted in all the other infected groups, but treatment with the anti-coccidial agents used for the study, suppressed the rise in oocysts output which became completely eliminated (no more seen in faeces) on days 12, 14 and 16 respectively post-infection in Groups B, C and A, respectively (Figure 1). Oocysts counts were highest in the Group E (infected untreated group) followed in descending order by those in Groups D, A, C and B (Figure 1).

The mean body weight gain was significantly higher ($p < 0.05$) in the Group F birds (uninfected untreated control) (Figure 2). Among the groups treated with anti-coccidial drugs, the mean body weight gain was highest

in group B (1.30kg), followed by Group D, A and C (1.20, 1.10 and 1.00kg, respectively). The Group E birds (infected untreated control) showed weight loss (Figure 2).

The mean feed conversion ratio was significantly lower ($p < 0.05$) in the Group F birds (uninfected untreated control group) (Figure 3). Among the groups treated with anti-coccidial drugs, the feed conversion ratio was significantly lower ($p < 0.05$) in Group B birds (2.00) which was treated with diclazuril followed respectively in ascending order by Group A, C and D (2.50, 2.60, and 2.70, respectively) [Figure 3]. However, the group E (infected untreated group) had a negative feed conversion ratio, (-2.00) [Figure 3].

The mean PCV of the Group F birds (uninfected untreated group) remained within

the pre-infection level throughout the study period (Figure 4). The infected untreated group showed a sharp decline in the mean PCV from day 4 PI, which continued till the death of all the birds in that group (Figure 4). Treatment with the anti-coccidial drugs reduced the rate and level of decline of PCV (Figure 4), and thereafter, the mean PCV improved significantly ($p < 0.05$) in all treated groups (Figure 4).

The mean RBC counts of the infected untreated group showed a significant ($p < 0.05$) decline in number and the lowest value was attained on day 8 post infection (Figure 5). Treatment with anti coccidial drugs led to a significant ($p < 0.05$) improvement in the mean RBC counts of the infected treated groups (Figure 5).

Table 1. Mortality pattern of the chicks infected with *Eimeria spp* and treated with Sulphaquinoxaline + diaveridine combination, Diclazuril, Tortrazuril or Diclazuril + Tylosin tartrate combination, compared with untreated and uninfected controls.

Groups (Treatments in brackets)	No. of birds that died/No. in the group (% mortality in brackets)	Date of death (days post-infection)
Group A (Infected and medicated with Sulphaquinoxaline + diaveridine combination).	1/7 (14.3%)	Day 5
Group B (Infected and medicated with Diclazuril).	1/7 (14.3%)	Day 11
Group C (Infected and medicated with Tortrazuril).	1/7 (14.3%)	Day 6
Group D (infected and treated with Diclazuril + Tylosin tartrate combination)	2/7 (28.6%)	Day 5
Group E (Infected non-medicated group)	7/7 (100%)	Days 5, 7 10 and 11
Group F (Uninfected non-medicated control group)	0/7 (0%)	-

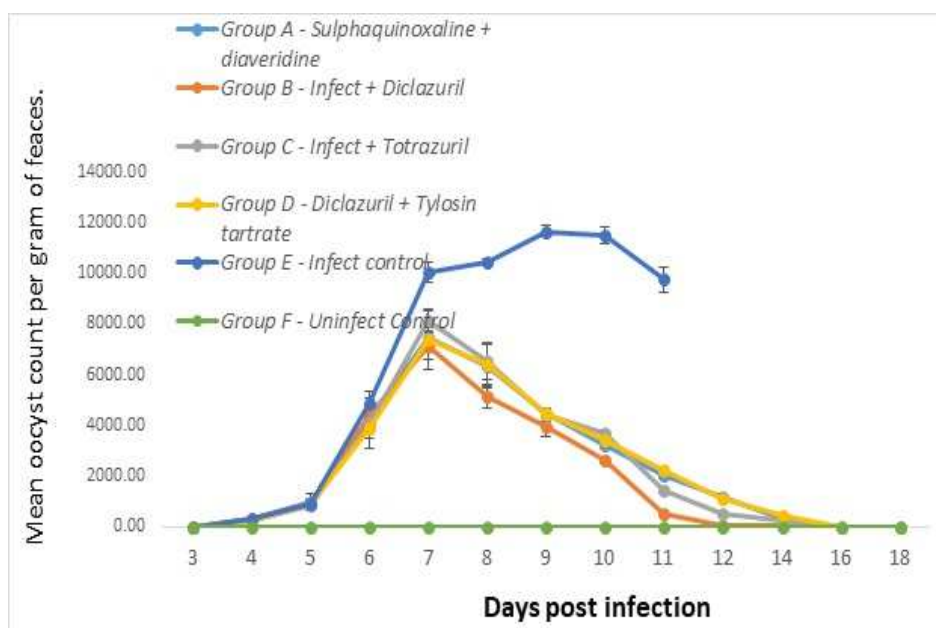


Figure 1. A line graph showing the mean oocyst count per gramme of faeces of groups of broiler chickens infected with *Eimeria* spp. and treated with Sulphaquinoxaline + diaveridine combination, Diclazuril, Tortrazuril or Diclazuril + Tylosin tartrate combination, compared with an untreated control.

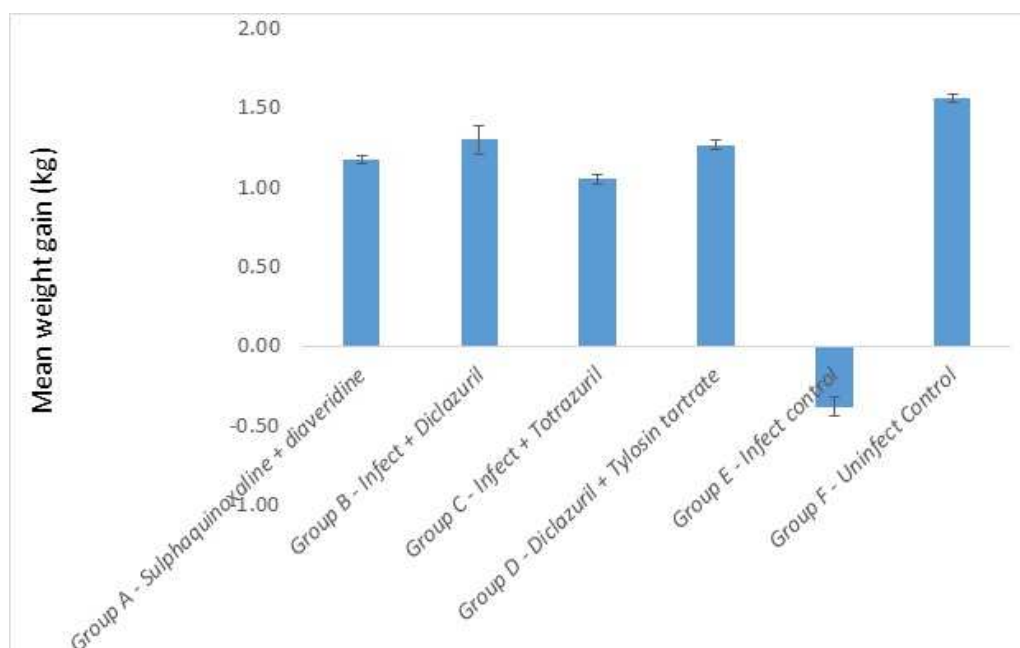


Figure 2. Mean body weight gains or loss groups of broiler chickens infected with *Eimeria* spp. and treated with Sulphaquinoxaline + diaveridine combination, Diclazuril, Tortrazuril or Diclazuril + Tylosin tartrate combination, compared with untreated and uninfected controls.

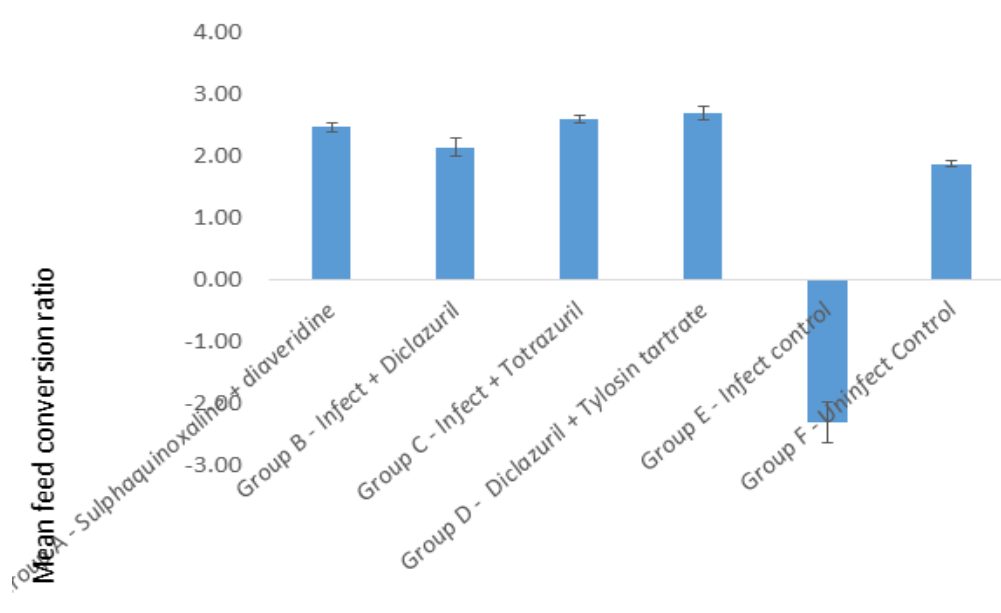


Figure 3. Feed conversion ratio of groups of broiler chickens infected with *Eimeria* spp. and treated with Sulphaquinoxaline + diaveridine combination, Diclazuril, Tortrazuril or Diclazuril + Tylosin tartrate combination, compared with untreated and uninfected controls.

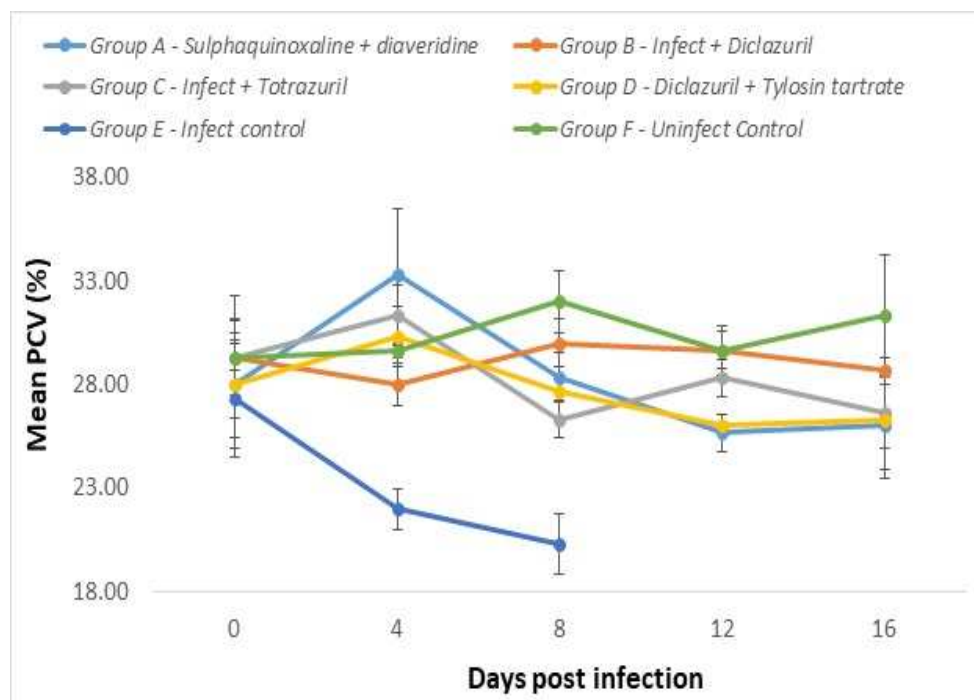


Figure 4. Mean packed cell volume of groups of broiler chickens infected with *Eimeria* spp. and treated with Sulphaquinoxaline + diaveridine combination, Diclazuril, Tortrazuril or Diclazuril + Tylosin tartrate combination, compared with untreated and uninfected controls.

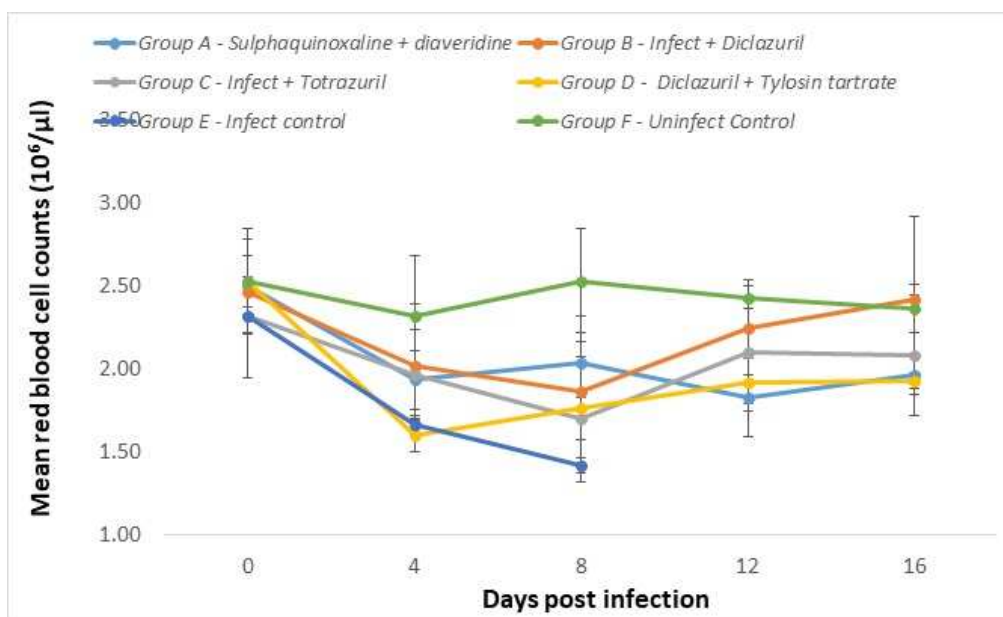


Figure 5. Mean red blood cell count of groups of broiler chickens infected with *Eimeria* spp. and treated with Sulphaquinoxaline + diaveridine combination, Diclazuril, Tortrazuril or Diclazuril + Tylosin tartrate combination, compared with untreated and uninfected controls.

Discussion

The clinical signs and gross lesions recorded in the infected groups of broilers were consistent with the earlier findings in chickens experimentally infected with *Eimeria* spp. (Guo et al, 2007). The occurrence of mortality is also a consistent sign of coccidiosis more especially during infection with the highly pathogenic species like *E. tenella*, *E. necatrix* and *E. acervulina* (Bedmik, 1983). This is also in agreement with reports of Tipu et al , (2002), who also observed highest mortality in birds that were infected with *Eimeria* spp and were not treated.

The reduction in oocyst count per gramme of faeces (OPG) in the birds treated with anti-coccidial agents is in agreement with the reports by Anosa et al. (2017), which showed reduction in oocyst count per gramme of faeces in birds treated with Sulphaquinoxaline and diaveridine combination and/or Tortrazuril. Conway et al (2001) demonstrated that, the use of 1 ppm diclazuril in shuttle programme was highly efficacious against a

mixed inoculum of *Eimeria* spp in comparison with nicarbazine, narasine + nicarbazine and zoalene in starter diets and salinomycin, monesin and lasalocid in grower diet.

The results of the present study indicate that diclazuril was more effective in improving mean weight gain when compared to other anti-coccidial drugs used in this study. Weight loss recorded in the infected untreated group might be due to anorexia and malabsorption. Bedmik (1969) demonstrated that a diminished effectiveness of an anti-coccidial drugs adversely impacts weight gain while the improvement in weight gain of the treated groups can be attributed to the decline in the oocyst count of the birds as well as reduction in the inflammation of intestinal mucosa leading to enhance nutrient absorption.

The relatively better feed conversion ratio in the infected birds treated with anti-coccidial agents is an indication of the efficacy of the anti-coccidial drugs used. The finding in the present study that Group B birds had the lowest feed conversion ratio among the

groups treated with anti-coccidial agents implies that group B broilers which were treated with diclazuril required a little quantity of feed to gain higher body weight, unlike the other treated groups that required relatively larger quantity of feed to gain weight. Also, in addition to anti-coccidial activity exhibited by the drugs, diclazuril treatment may have also stimulated appetite leading to increase in feed consumption and better weight gain (Aiemojoy and Tsion, 2020).

The reduction in the PCV and RBC of the various groups of the *Eimeria*-infected broilers was an indication of anaemia due to caecal coccidiosis, which could be as a result of haemorrhage or blood loss in the intestine and caecum (Aiemojoy and Tsion, 2020). Anaemia due to blood loss is a common finding in *Eimeria* species infection in chicken (McDougald and Reid, 1997). Improvement of these erythrocytic parameters in *Eimeria* infected birds after administering anti-coccidial drugs such as diclazuril and salimimucin has been reported earlier by Hirani et al (2018).

It is thought that the variation in the activity of the various anti-coccidial drugs could be due to the fact that Sulphaquinoxaline and diaveridine combination and Tortrazuril have been in use in Nigeria for a long time and resistance to them has been claimed. Conversely, Diclazuril is a more recently introduced drug to Nigeria; therefore; resistance to it is less likely to have developed. Also, diclazuril may also be more efficacious than others, because it is only administered for a period of 24 hours, unlike the other two drugs that are administered for five days. There is thus a higher chance of under dosing and subsequent development of resistance for the other anti-coccidial drugs that are given over five days (Anosa et al., 2011). Earlier studies by Obi et al. (2025) reported improved weight gain, feed conversion ratio, lower faecal oocyst counts and better PCV in chickens medicated with diclazuril.

Conclusion: All the four anti-coccidial drugs used in this study were effective in the treatment of coccidiosis, but diclazuril demonstrated a better anti-coccidial effect by reducing mortality and faecal oocyst counts while improving PCV, weight gain and feed conversion efficiency more than the other anti-coccidials drugs. The use of diclazuril is recommended.

Conflict of interest

No conflict of interest exist

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