



## Efficacy of Herbal Coccidiocidal and Coccidiostatic Blend (Biococcin) in Experimentally Induced Coccidiosis in Broiler Birds

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### Abstract

Present study was carried out on 350 broiler birds having seven different groups. Normal control (no infection), challenged control (only infection of *E. tenella*) and biococcin, monensin, amprolium, furazolidone, and sulphonamides group were challenged with *E. tenella* oocytes the different groups, each group was having 50 birds with 5 replicates. All the medicines were fed to the broiler birds from day 1. On day 14 of experiment *E. tenella* challenge was given to six groups and normal control was not given infection of *E. tenella*. Biococcin is the blend of rich phytochemicals consisted of *Holarrhena antidysenterica*, *Tinospora cordifolia*, *Allium sativum*, *Cinnamomum camphora*, which were able to acts as anticoccidial. The efficacy of biococcin was similar or little bit higher than the regular standard and most effective drugs like monensin and amprolium. Efficacy of biococcin was quite superior in respect to body weights, FCR, mortality pattern, gross and microscopic scoring of caecal lesions. OPG were significantly lower in Biococcin group as compared to all other challenged groups. The active ingredients present in Biococcin were able to destroy the infective stages and prevents the gametocyte formation of *E. tenella*.

**Keywords:** Biococcin; Herbal Coccidiocidal; *E. Tenella*; Broiler Birds

### Introduction

Coccidiosis is one of the most detrimental diseases in poultry due to primarily impaired feed conversion, depressed growth rate and, sometimes, increased mortality [13]. It leads to the extensive destruction of the intestinal epithelium which results in reduced food efficiency and body weight gain, as well as a temporary reduction in egg production [2,3]. This has a considerable economic loss at the level of poultry industry. The three most common species of *Eimeria* that affect the poultry industry are *Eimeria tenella*, *Eimeria maxima*, and *Eimeria acervulina* [4]. *Eimeria* infection is transmitted through hardy, thick walled sporulated oocysts which

are able to survive for lengthy periods in the poultry litter and soil particles.

Coccidiosis is mainly controlled by the hygienic measures and the use of chemotherapeutic agents or chemical anticoccidial agents [5]. However, the development of drug resistance to all the drugs introduced so far by the causative parasites and the escalating cost of drug development have greatly reduced the commercial incentive to develop new chemical anticoccidials [1,6-9]. Moreover, nowadays, consumers request poultry products that are free from residual drugs [12]. Consequently, the development of alternative,

safer and environmentally friendly anticoccidial agents have become priority in most parts of the world [13]. The use of natural products as an alternative to drugs may be the best solution to this consumer demand. Many studies have reported the *in vivo* efficiency of natural plant extracts in the treatment of coccidiosis [14-16]. Garlic (*Allium sativum*) and turmeric (*Curcuma longa*) are widely used to cure and prevent diseases in human beings as well as in animals [17,18,20,21].

This study is a part of a program aiming to find alternative strategies to synthetic coccidiostats for treatment and control of coccidiosis in chicken using medicinal plants (natural products). The objective of this study is to evaluate the efficacy of Biococcin product produced by Vinayak Ingredients Pvt. Ltd. in comparison with common antibiotics, against mixed *Eimeria* species through *in vivo* experimental designs.

Materials and Methods

The present investigation was conducted at the poultry unit of the Omega Labs farm at Lonand, District Satara, Maharashtra, INDIA. The farm is situated at 18°02'33.3"N North latitude and 74°10'52.6"E East longitude in the Western region of Maharashtra. It is about 518 metres above mean sea level (MSL). The climate is hot and humid with 500 to 1000 mm of average rainfall. The relative humidity (RH) ranges from 35% to 80%. The study was carried out in two steps.



Figure a

Experimentally induced coccidiosis

Experimental groups are given in table no 1. for experimental induction of coccidiosis. The oocytes of *Eimeria tenella*, were recovered from the field cases and incubated in potassium dichromate for 24hours. After 24hours the suspensions were made into standard of 25000 to 30000 oocytes per ml of inoculums. 1 ml of inoculums was fed to chicks on 14<sup>th</sup> day and clinical signs were observed for 15days. 50 experimental birds were kept in different pens and reared in standard managemental practices.

Experimental groups

Detail clinical signs were observed and detailed post-mortem examination of dead birds was carried out and visual score and findings were recorded and caeca were processed for microscopic examination. Oocytes per gram of faecal samples were evaluated at the end of experiment [10]. After completion of experiment on 42<sup>nd</sup> day 6 birds from each group were sacrificed and detailed examination of caeca was carried out and lesion score was undertaken [11]. Caeca from dead birds and sacrificed birds were preserved in 10% formalin for histopathological scoring [11]. Paraffin embedding technique was used for the tissue processing and H and E stained slides were observed for microscopic scoring. Microphotography of H and E stained sections were carried out by using Magnus Olympus camera. Weekly body weights and FCR was calculated and recorded. Statistical analysis was carried out by using Dunnett's Method/Dunnett's Multiple Comparison. [22].

Experimental groups	Numbers of birds in group
T0 (Negative control)	50 (10 birds in 1 pen X 5 pens)
T1 (Positive control)	50 (10 birds in 1 pen X 5 pens)
T2 Biococcin @ 500mg·kg <sup>-1</sup>	50 (10 birds in 1 pen X 5 pens)
T3 Monensin @ 150mg·kg <sup>-1</sup>	50 (10 birds in 1 pen X 5 pens)
T4 Sulphonamide @ 150mg·kg <sup>-1</sup>	50 (10 birds in 1 pen X 5 pens)
T5 Amprolium @ 300mg·kg <sup>-1</sup>	50 (10 birds in 1 pen X 5 pens)
T6 Furazolidone @ 300mg·kg <sup>-1</sup>	50 (10 birds in 1 pen X 5 pens)

Table 1

### Composition of Biococcin

Biococcin is the blend of rich phytochemicals consisted of *Holarrhena antidysentrica*, *Tinospora cordifolia*, *Allium sativum*, *Cinnamomum camphora*, which are actively acts as anticoccidial effects.

### Results and Discussion

In the present study average weekly body weights were significantly higher in the normal controlled group than the other treatment groups. Challenged group who received only infection showed significantly lower average body weights than the other groups. Group Biococcin did not showed significant differences in monensin, Sulphonamides, amprolium and Furazolidone groups for the body weights. Whereas FCR was significantly higher in challenged group as compared to all other groups. Biococcin group showed significantly lower FCR than the Sulphonamie, Amprolium and Furazolidone groups, but there was no any significant difference between monensin and Biococcin. Mortality percentages were significantly higher in group challenged control than the other groups. Biococcin group did not showed significant difference amongst the other treatment groups. Caecal gross pathological score was significantly higher in group challenged control than the other treatment groups. (Figure 1-4) Biococcin group showed significantly lower gross pathological score as compared to Amprolium, Furazolidone and Sulphonamide groups. Whereas there was no significant difference between Monensin and Biococcin. Biococcin group showed significantly lower OPG of fecal material as compared to all other treatment groups. Histopathological score was significantly lower in Biococcin group as compared to other treatment groups. (Figure 5-8) Similar types of observations were noticed by Giannenas, *et al.* 2003, in experimentally induced coccidia in broiler birds. Youn H.J., *et al.* 2001, studied the effectiveness of herbal anticoccidial herb extracts against *Eimeria tenella* and their findings were similar to the present experiment. In the present study it was observed that the phytochemicals presents in Biococcin viz. *Holarrhena antidysentrica*, *Tinospora cordifolia*, *Allium sativum* and *Cinnamomum camphora*, were able to destruct coccidial oocytes and other stages and prevent gamatogonia formations in the intestinal tract. Similar observations were observed by Sivropoulou, *et al.* 1996 [15], Kumar, *et al.* 2022 [20] and Fayed, *et al.* 2022 [21].



**Figure 1:** Challenged control group: Photograph showing severe hemorrhages, ballooning of caeca.



**Figure 2:** Amprolium group: Photograph showing moderate hemorrhages, ballooning of caeca.

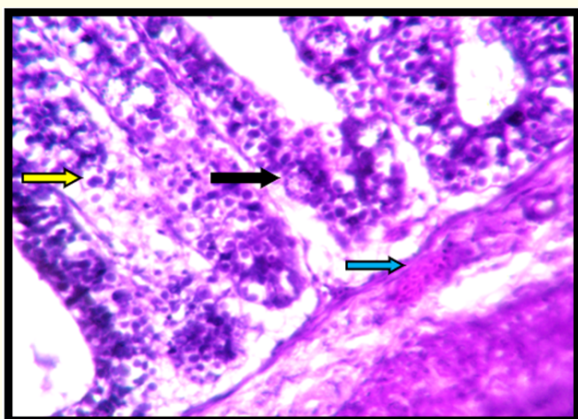


**Figure 3:** Biococcin 500 group: Photograph showing mild hemorrhages, ballooning of caeca.

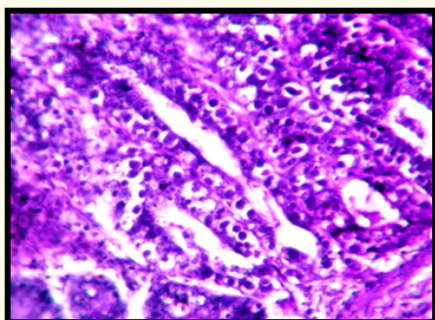




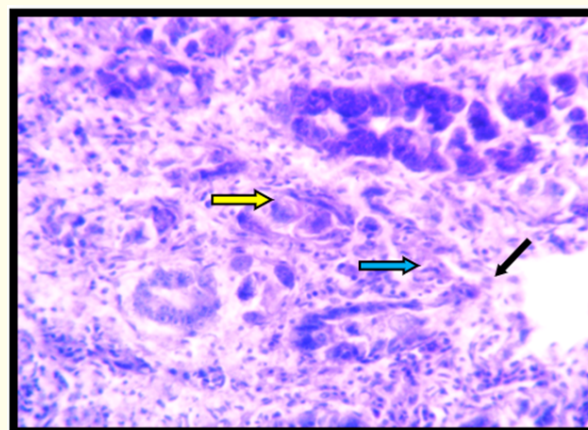
**Figure 4:** Monensin group: Photograph showing moderate hemorrhages, ballooning of caeca.



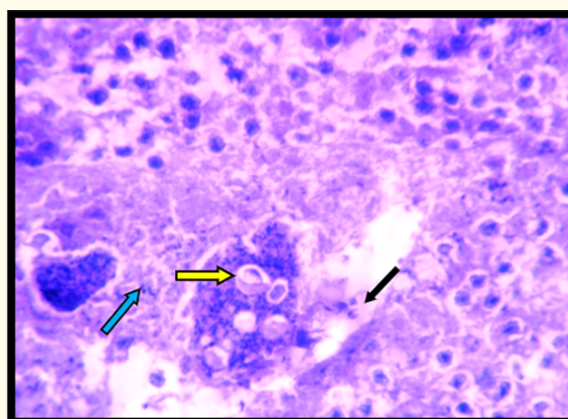
**Figure 5:** Challenged control group: Photograph showing necrosis of epithelial cells (black arrow), gametocytes of coccidian (yellow arrow), infiltration of leucocytes (blue arrow) H and E stain 400 X.



**Figure 6:** Amprolium group: Photograph showing necrosis of epithelial cells (black arrow), gametocytes of coccidian (yellow arrow), infiltration of leucocytes (blue arrow) H and E stain 400 X.



**Figure 7:** Biococcin 500 group: Photograph showing necrosis of epithelial cells (black arrow), gametocytes of coccidian (yellow arrow), infiltration of leucocytes (blue arrow) H and E stain 400 X.

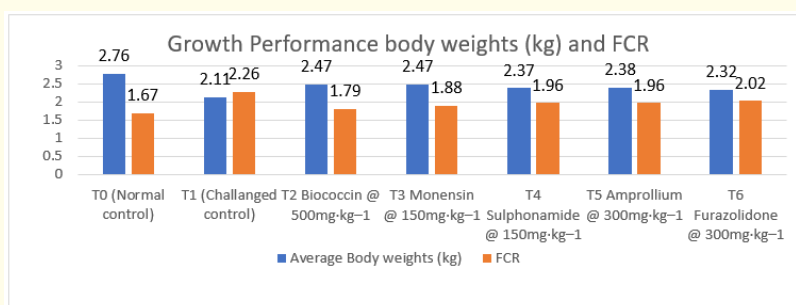
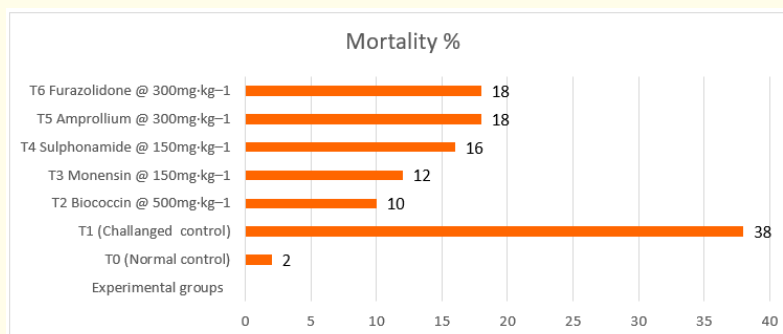


**Figure 8:** Monensin group: Photograph showing necrosis of epithelial cells (black arrow), gametocytes of coccidian (yellow arrow), infiltration of leucocytes (blue arrow) H and E stain 400 X.

## Conclusion

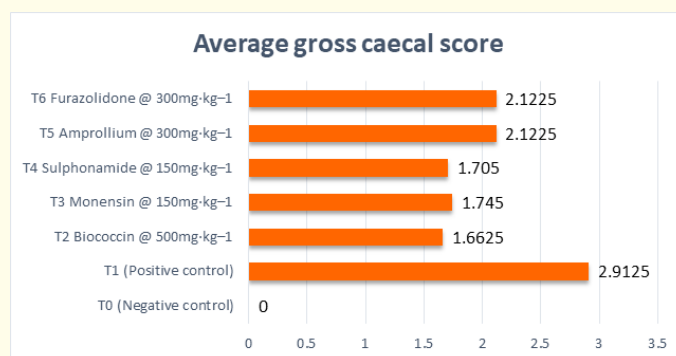
- BIOCOCCIN @ 500g/tonne of feed was the effective against treatment for the clinical coccidiosis caused by the infective species of *Eimeria* sp.
- Biococcin can be used in broiler feed throughout the period to prevent and cure Coccidiosis. It is economically viable and reduces the severity and losses due to the infection.
- It can replace the traditionally used anticoccidials without hampering the efficacy against coccidia for prevention and treatment of *Eimeria* spp.

Experimental groups	Average Body weights (kg)	FCR	Mortality %
T0 (Normal control)	2.76 <sup>a</sup>	1.67 <sup>a</sup>	2 <sup>c</sup>
T1 (Challenged control)	2.11 <sup>c</sup>	2.26 <sup>d</sup>	38 <sup>a</sup>
T2 Biococcin @ 500mg·kg-1	2.47 <sup>b</sup>	1.79 <sup>b</sup>	10 <sup>b</sup>
T3 Monensin @ 150mg·kg-1	2.47 <sup>b</sup>	1.88 <sup>b</sup>	12 <sup>b</sup>
T4 Sulphonamide @ 150mg·kg-1	2.37 <sup>b</sup>	1.96 <sup>c</sup>	16 <sup>b</sup>
T5 Amprolium @ 300mg·kg-1	2.38 <sup>b</sup>	1.96 <sup>c</sup>	18 <sup>b</sup>
T6 Furazolidone @ 300mg·kg-1	2.32 <sup>b</sup>	2.02 <sup>c</sup>	18 <sup>b</sup>

**Table 2:** Average body weights, FCR and mortality %.**Graph 1:** Growth performance.**Graph 2:** Mortality percentage.

Experimental groups	Haemorrhages	Ballooning	Core formation	Proliferative lesions	Average
T0 (Normal control)	0	0	0	0	0 <sup>d</sup>
T1 (Challenged control)	3.5	3.66	2.33	2.16	2.9125 <sup>a</sup>
T2 Biococcin @ 500mg·kg-1	2.33	1.83	1.33	1.16	1.6625 <sup>c</sup>
T3 Monensin @ 150mg·kg-1	2.33	1.83	1.66	1.16	1.745 <sup>c</sup>
T4 Sulphonamide @ 150mg·kg-1	2.33	1.83	1.33	1.33	1.705 <sup>c</sup>
T5 Amprolium @ 300mg·kg-1	2.5	2.66	1.83	1.5	2.1225 <sup>b</sup>
T6 Furazolidone @ 300mg·kg-1	2.5	2.66	1.83	1.5	2.1225 <sup>b</sup>

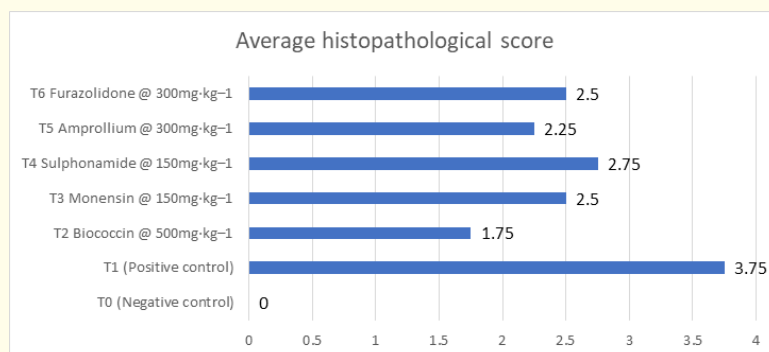
**Table 3:** Caecal gross score out of 4.



Graph 3: Average gross caecal score.

Experimental groups	Hemorrhages	Necrosis of epithelial cells	Gmatocytes	Cellular infiltration	Average
T0 (Normal control)	0	0	0	0	0
T1 (Challenged control)	4	4	4	3	3.75 <sup>a</sup>
T2 Biococcin @ 500mg·kg <sup>-1</sup>	2	2	1	2	1.75 <sup>c</sup>
T3 Monensin @ 150mg·kg <sup>-1</sup>	3	3	2	2	2.5 <sup>b</sup>
T4 Sulphonamide @ 150mg·kg <sup>-1</sup>	2	3	3	3	2.75 <sup>a</sup>
T5 Amprolium @ 300mg·kg <sup>-1</sup>	3	2	2	2	2.25 <sup>b</sup>
T6 Furazolidone @ 300mg·kg <sup>-1</sup>	3	3	2	2	2.5 <sup>b</sup>

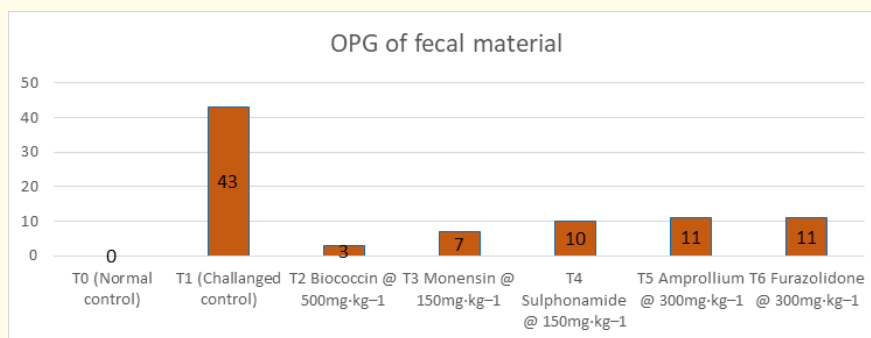
Table 4: Histopathological score of caeca out of 4.



Graph 4: Average histopathological score out of 4.

Experimental groups	OPG of fecal material
T0 (Normal control)	0 <sup>d</sup>
T1 (Challenged control)	43 <sup>a</sup>
T2 Biococcin @ 500mg·kg <sup>-1</sup>	3 <sup>d</sup>
T3 Monensin @ 150mg·kg <sup>-1</sup>	7 <sup>c</sup>
T4 Sulphonamide @ 150mg·kg <sup>-1</sup>	10 <sup>b</sup>
T5 Amprolium @ 300mg·kg <sup>-1</sup>	11 <sup>b</sup>
T6 Furazolidone @ 300mg·kg <sup>-1</sup>	11 <sup>b</sup>

Table 5: OPG counts.



Graph 5: Comparative OPG Counts.

### Conflict of Interest

Nil.

### Acknowledgements

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