Kafrelsheikh Vet. Med. J. Vol. 4 No. 2 (2006) (59-72)

THE ANTICOCCIDIAL EFFECTS OF TURMERIC AND GINGER AGAINST *EIMERIA TENELLA*

Mahmoud M. Ismail[*] *and Abdel-Razek Y. Desouky*[**]

[*]Department of Poultry and Fish and [**]Department of Parasitology

Faculty of Veterinary Medicine, Kafr El-Sheikh University, Kafr El-Sheikh ,Egypt.

ABSTRACT

Chicken coccidiosis is caused by intracellular protozoan parasites belonging to seven species of Eimeria, their development in the lining of the intestine and ceci causes diarrhea, morbidity and mortality, which lead into serious economic consequences in poultry industry. Due to the emergence of drug-resistant coccidial strains and the bad impact of drug-residues in poultry products on health of consumers, we have sought safe alternative anticoccidial dietary spice materials (turmeric and ginger) for the control of chicken coccidiosis.

Two hundred and twenty five, 28-days-old chicks were used in the present study, two trials were conducted, 5 groups of birds in each trial, four groups were infected orally with 10,000 sporulated Eimeria tenella oocysts and group 5 were kept as non infected control. Group 1 received ration containing 5% turmeric, group 2 received ration containing 2% ginger, group 3 received plain non medicated ration but amprolium was added to drinking water, Groups 4 and 5 were fed on plain ration without additives to serve as infected unmedicated control and non infected unmedicated control groups respectively. Ration used in all groups was formulated without anticoccidals. Bloody diarrhea, oocysts excretion, lesion scores, mortalities and the mean body weights were investigated throughout the experimental period.

Results showed that bloody diarrhea was observed from day 4 to day 8 post infection in infected unmedicated control group and in group which received ginger in ration. The turmeric-treated group and the non infected control group showed no bloody diarrhea. There was minimal bloody diarrhea in amprolium treated group at 5 days post infection, excreted oocysts in the groups treated with ration supplemented with 5% turmeric powder were markedly lower than that of the infected unmedicated control group. Lesion scores in groups treated with turmeric and amprolium were markedly lower than that of the infected non medicated control groups and groups treated with ginger, by the end of the experiment, the highest mortalities were recorded in the infected unmedicated control groups, followed by groups treated with ginger, then in groups treated with amprolium and there were no mortalities in groups treated with turmeric. The mean body weights in groups treated with turmeric and amprolium were significantly higher than that of the infected unmedicated control groups.

On the basis of the overall results of the present investigation,turmeric is a promising new natural herbal coccidiostat. Further research to indicate the active principle, mechanism of action, the optimal anticoccidial dose and the possible tanning effect of turmeric on the meat of poultry will be carried out by the authors.This paper documents the use of turmeric as anticoccidial in chickens for the first time.

INTRODUCTION

Avian coccidiosis is a serious widespread enteric protozoal disease of poultry caused by species of genus Eimeria (Apicomplexa: Eimeriidae) and costs the world commercial chicken producers at least 800 million USD every year ***(Williams, 1998)***. This sum comprises the cost of prophylactic in-feed medication in broilers and breeding stock, plus the cost of therapy to control the disease outbreaks if prophylaxis fails and the financial losses due to mortality and poor performance of surviving birds ***(Hilali, 2003)*.**

Eimeria tenella is one of the highly pathogenic species and is primarily responsible for cecal coccidiosis, which occurs principally in chickens of 3-7 weeks of age. The first stage schizonts of this species develop deep in the glands. The second stage schizonts are also unusual in that the epithelial cells in which they develop leave the mucosa and migrate into the lamina propria and submucosa, when this schizonts mature and rupture, about 72 hours after ingestion of sporulated oocysts, hemorrhage occurs, and the mucosal surface is largely detached and clinical signs become apparent. Clinical disease occurs when large numbers of sporulated oocysts are ingested over a short period and is characterized by the presence of soft feces often containing blood. The chicks are dull and listless with droopy wings. At postmortem examination of chickens which had blood in their feces, the ceci are found to be dilated and contain mixture of clotted and unclotted blood. In longer-standing infections the cecal contents become caseous and adherent to the mucosa ***(Urquhart et al., 1988)*.**

Thus, many anticoccidial drugs have been developed and introduced in the poultry industry all over the world since ***Levine (1939)*** discovered sulfanilamide would cure coccidiosis in chickens, various anticoccidial feed additives, predominantly polyether ionophorous antibiotics, have been developed and used ***(Matsuda et al., 1989)*.** While effective for avian coccidiosis, the continuous uncontrolled use of anticoccidial drugs have led to the emergence of drug-resistant coccidial strains ***(Long, 1982)*.**

To prevent the emergence of drug resistant strains, new drugs have been developed and administered on rotational basis with existing drugs. However, this has resulted in the increased cost of poultry products. Furthermore, drug-or antibiotic-residue in the poultry product is potentially annoyance to consumer. Therefore, it is sought that the regulations for anticoccidial drugs should be strengthened gradually ***(Youn and Noh, 2001)*.**

Prohibition of antibiotics, as well as the risk of resistance to anticoccidials and their residues in the poultry products, indicates that a search for alternative ways to maintain the health of poultry stock and the consumers is highly encouraged. Extracts of herbs were known to be effective against parasites such as Plasmodium, amoeba, trichomonads, arthropods and helminthes ***(Foure and Bennejean, 1974; Klayman et al., 1984; Akhtar and Rifaat, 1985; Jiang et al., 1985; Klayman, 1985; Cooke et al., 1987; Lin et al., 1987; Dutta et al., 1989, 1990; He and Zhang, 1989; Matsuda et al., 1989, 1991; Shuhua and Catto, 1989; Ou-Yang et al., 1990; Quan, 1990).*** Testing the efficacy of some herbal extracts against coccidia was tried by ***Akhtar and Rifaat (1987), Oh et al., (1995 a&b) and Youn and Noh (2001).*** The aim of the present study is to investigate the anticoccidial effect of two common dietary spices (Trumeric and Ginger). Turmeric (*Curcuma longa*), is a common Indian dietary pigment and spice, and has been used therapeutically in traditional folk medicine. It has been shown to exhibit a variety of biological activities including antioxidative activity ***(Cheng et al., 2005; Chen et al., 2006),*** anti-proliferative, anticarcinogenic, antioxidative and hypocholesterolemic activities ***(Indap et al., 2006),*** antidepressant, anti-inflammatory activities ***(Tapsell et al., 2006),*** antibacterial activities ***(Park et al., 2005),*** and finally, turmeric has gastro-protective and antiulcerogenic effects ***(Kim et al., 2005).*** Ginger has been shown, also to have anti-inflammatory, antibacterial and anti-tumor activities ***(Park et al., 2006).*** The anticoccidial activities of turmeric and ginger were tested for the first time in the present investigation.

MATERIALS &METHODS

Two hundred and twenty five, 1-day-old commercial broiler chicks were used in all trials in this experiment. Chicks were free from detectable chicken pathogens. All the birds used in the different experimental groups were kept in high-security isolation rooms and

provided with feed (free from anticoccidials) and water *ad libitum*. Chicks were vaccinated against Newcastle disease by Hitchiner B1 at 7 days old and at 25 days by La Sota vaccine via drinking water. Birds were kept together in the same room until one day before challenge after that, different experimental groups were kept in separate rooms. Challenge with sporulated coccidial oocysts was done at 28 days old.

Coccidial Oocysts: Strain of *Eimeria tenella* used in this study was obtained from field cases from chicken farms, and was cloned by single oocyst infection. The oocysts were preserved in 2.5% potassium dichromate solution to assist sporulation and kept in refrigerator (4C) till used. Each bird was orally inoculated with 10,000 sporulated oocysts at 28 days of age. Oocysts count was done according to method described by Hodgson (1970) using McMaster counting chamber.

Experimental inoculation of chickens:

Trial 1: One hundred, 28-days old birds were allotted into five groups of 20 birds each. Groups 1, 2, 3, and 4 each bird was orally inoculated with sporulated oocysts as described previously. Group 1 received ration containing 5% Turmeric acid, Group 2 received ration with 2% Ginger. Group 3 received regular ration but amprolium was added to drinking water. Group 4 was fed on ration free of anticoccidials to serve as infected unmedicated control. Group 5 was kept as non infected unmedicated controls.

Trial 2: Hundred and twenty five, 28-days old birds were allotted into five groups of 25 birds each. Groups 1, 2, 3, and 4 each bird was orally inoculated with sporulated oocysts as described previously. Group 1 received ration containing 5% Turmeric acid, Group 2 received ration with 2% Ginger. Group 3 received regular ration but amprolium was added to drinking water. Group 4 was fed on ration free of anticoccidials to serve as infected unmedicated controls. Group 5 was kept as non infected unmedicated controls.

In both trials, chickens were observed twice daily for clinical signs. Bloody diarrhea was investigated from day 0 - 15 post infection. The degree of bloody diarrhea was recorded as score from 0 - 4 where 0 (-) is normal and 4 (++++) is the maximum degree of bloody diarrhea. Cecal lesions were also recorded as score from 0-4 where 0 (-) is normal and 4 (++++) is the most severe lesion (Johnson and Reid, 1970). Excreted oocysts were counted from day 0 to 10, after infection with *E. tenella*. Mortalities were recorded and chicken in each group were weighed at the end of the experiment.

Statistical analysis: Statistical comparison of body weights between different groups and control birds was performed using one way analysis of variance (ANOVA). ***(Snedecor and Cochran, 1980).***

RESULTS

Bloody diarrhea was observed from day 4 to day 8 post infection in infected control group and in group which received ginger in ration. The turmeric-treated group and the non infected control group showed no bloody diarrhea. There was minimal bloody diarrhea in amprolium-treated group at 5 days post infection (Table 1).

Excreted oocysts in the groups treated with ration supplemented with 5% turmeric powder were markedly lower than that of the infected control group. In the groups treated with ration supplemented with ginger powder and amprolium, the peak excretion of oocysts was delayed 2 days relative to the control infected group (Table 2).

Lesion scores in ceci of chickens were recorded for all groups from day 4 to day 8 post infection. The lesion scores in groups treated with turmeric and amprolium were markedly lower than that of the infected control groups and groups treated with ginger (Table 3).

At the end of the experiment, the highest mortalities were seen in the infected control groups, followed by groups treated with ginger, then in groups treated with amprolium and there were no mortalities in groups treated with turmeric (Table 4).

The mean body weight in groups treated with turmeric and amprolium was significantly higher than that in the infected control groups. While in ginger-treated group and infected unmedicated group body weight was significantly lower than non infected control group (Table 4). The overall results indicated that turmeric is a promising new coccidiostat. It is slightly better than amprolium on the basis of the results of the present investigation, while ginger did not have anticoccidial effect at the used dose.

Table (1): Score of bloody diarrhea in chickens treated with turmeric, ginger, amprolium and challenged with *Eimeria tenella*.

Group	Bloody diarrhea at days post inoculation									
	4		5		6		7		8	
	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
Turmeric 5%	-	-	-	-	-	-	-	-	-	-
Ginger 2%	+	+	++	+++	++	++	+	+	-	-
Amprolium	-	-	+	+	-	-	-	-	-	-
Infected, unmedicated	+	+++	+++	+++	++	++	+	+	+	-
Non infected, unmedicated	-	-	-	-	-	-	-	-	-	-

Table (2): Oocyst shedding from chickens treated with turmeric, ginger and amprolium and challenged with *Eimeria tenella* (x1000):

Group	Oocyst count / gram of feces at days post inoculation									
	6		7		8		9		10	
	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
Turmeric 5%	25	20	16	15	4	4	1	0	0	0
Ginger 2%	32	30	40	48	6	8	3	4	1	1
Amprolium	14	12	20	25	8	10	2	1	1	0
Infected, unmedicated	105	110	70	60	30	35	6	5	1	2
Non infected, unmedicated	0	0	0	0	0	0	0	0	0	0

Table (3): Lesion scores in ceci of chickens treated with turmeric, ginger, amprolium and challenged with *Eimeria tenella*.

Group	Lesion score at days post inoculation									
	4		5		6		7		8	
	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
Turmeric 5%	-	-	-	-	-	-	+	-	-	-
Ginger 2%	+	+	++	+++	+++	++	++	++	+	+
Amprolium	-	-	+	+	-	-	-	-	-	-
Infected, unmedicated	+	+++	++++	++++	+++	+++	++	++	++	++
Non infected, unmedicated	-	-	-	-	-	-	-	-	-	-

Table (4): Mortalities and mean body weight in (grams ± SD) in chickens treated with turmeric, ginger, amprolium and challenged with *Eimeria tenella* at the end of the experiment.

Group	Turmeric 5%		Ginger 2%		Amprolium		Infected Unmed. control		Non infected Unmed. control	
	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
Mortality	-	-	3/20	2/25	1/20	-	4/20	5/25	-	-
Mean body weight	1987.1[a] ± 46.11	1888.5[a] ±41.1	1719.1[b] ±74.2	1730.5[b] ±67.2	1810.2[a] ±82.2	1875[a] ±56.2	1689.3[b] ±68.7	1707[b] ±60.8	1910 ±71.18	1961.2 ±43.9

a Values were significantly higher than positive controls ($p< 0.05$).

b Values were significantly lower than negative controls ($p< 0.05$).

DISCUSSION

After challenge with *E. tenella* and treatment with 5% turmeric, 2% ginger and amprolium, bloody diarrhea, oocysts excretion, lesion scores, mortalities and the mean body weights were investigated throughout the experimental period. Bloody diarrhea was observed from day 4 to day 8 post infection in infected control group and in group which received ginger in ration. The turmeric-treated group and the non infected control group showed no bloody diarrhea. There was minimal bloody diarrhea in

amprolium treated group at 5 days post infection. Excreted oocysts in the groups treated with ration supplemented with 5% turmeric powder were markedly lower than that of the infected control group. Lesion scores in groups treated with turmeric and amprolium were markedly lower than that of the infected control groups and groups treated with ginger. By the end of the experiment, the highest mortalities were recorded in the infected unmedicated control groups, followed by group treated with ginger, then in group treated with amprolium and there were no mortalities in groups treated with turmeric, the mean body weights in groups treated with turmeric and amprolium were significantly higher than that in the infected unmedicated control groups. The anticoccidial activities of herbal extracts have only been reported by a few scientists ***(Akhtar and Rifaat, 1987; Oh et al., 1995 a&b and Youn and Noh, 2001).*** Interestingly, we found that the groups treated with turmeric were much better than infected non medicated control groups and slightly better than amprolium treated groups. On the basis of the overall results of the present investigation, turmeric is a promising new natural herbal coccidiostat which has the advantage of improving the body weight gain, minimizing lesions and reducing number of excreted oocysts and without risk of developing drug resistance nor residues. The anticoccidial activities of turmeric may be attributed to the facts that it has anti-proliferative,antioxidative,antidepressant,anti-inflammatory,antibacterial, gastro-protective and antiulcerogenic activities as mentioned by ***Kim et al., (2005) ; Park et al., (2005); Indap et al., (2006) and Tapsell et al., (2006).*** Further research to delineate the mechanism of action, the active principle, the optimal anticoccidial dose and the possible tanning effect of turmeric on the meat of poultry will be carried out by the authors.

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اختبار فاعلية الكركم و الزنجبيل كمضادات للكوكسيديا في الدجاج

محمود موسى إسماعيل [*] ، ***عبد الرازق يوسف دسوقي*** [**]

[*] **قسم الدواجن و الأسماك** و [**] **قسم الطفيليات**

كلية الطب البيطري– جامعة كفر الشيخ

يعتبر مرض الكوكسيديا في الدجاج من أهم الأمراض التي تسبب خسائر اقتصادية فادحة لصناعة الدواجن على مستوى العالم و تقدر تلك الخسائر بما يزيد عن 800 مليون دولار أمريكي سنويا نتيجة للاستخدام المستمر لمضادات الكوكسيديا الكيميائية كمضيفات أعلاف لمنع الإصابة في مزارع البياض و التسمين بالإضافة إلى تكاليف العلاج و الخسائر الناتجة عن نفوق أعداد كبيرة من الطيور المصابة و ضعف إنتاجية الطيور الحية.و لقد أدى الاستخدام المستمر و العشوائي لمضادات الكوكسيديا الكيميائية لظهور عترات من الكوكسيديا مقاومة لتلك المضادات كما أن معظم تلك المضادات لها متبقيات في أنسجة الطيور مما يشكل خطرا كبيرا على صحة المستهلكين. لذا كان من الضروري البحث عن طرق بديلة و امنة لعلاج الكوكسيديا و تتلافى فى نفس الوقت عيوب المضادات الكيميائية، لهذا قام الباحثان بتجربة إستخدام كل من الكركم و الزنجبيل الخام كمضاد للكوكسيديا فى الدجاج، و اجريت الدراسة على عدد 225 دجاجة عمر 28 يوم خالية من الأمراض ظاهريا ومحصنة ضد الفيروسات المعروفة و تم تربيتها خصيصا على عليقة خالية من مضادات الكوكسيديا و تحت ظروف صحية مناسبة، تم عمل تجربتين حيث قسم الدجاج فى كل تجربة الى خمس مجوعات و تم عدوى الدجاج بعدد 10.000/ لكل طائر من الحويصلات القادرة على احداث العدوى عن طريق الفم،

مجموعة 1 تم تغذيتها على عليقة بها 5% كركم ، مجموعة 2 تم تغذيتها على عليقة بها 2% زنجبيل، مجموعة 3 تم تغذيتها على عليقة عادية ولكن تم وضع الامبروليوم كمضاد للكوكسيديا فى الماء، مجموعة 4 مجموعة معدية غير معالجة ضابطة، المجموعة5 مجموعة غير معدية ضابطة ، و أظهرت النتائج أن المجموعات التى تم تغذيتها على عليقة مضاف اليها 5% كركم كانت الأفضل من حيث منع الاعراض المرضية المتمثلة فى الاسهال المدمم و تقليل عدد الحويصلات المفرزة مع الاخراج و كذلك تقليل الصفة التشريحية المميزة للكوكسيديا فى الاعورين و منع النفوق و كذلك المساعدة على زيادة الوزن و من هذا يتضح أن الكركم من مضادات الكوكسيديا الواعدة و التى بالاضافة الى المزايا السابق ذكرها فانه ليس له اثار جانبية كظهور عترات مقاومه له أو اثار متبقية فى الانسجة ، و يرى الباحثان اجراء دراسات مستقبلية لتحديد المادة الفعالة المضادة للكزكسيديا فى الكركم و كيفية عملها و تحديد الجرعة المناسبة، و يعتبر هذا البحث أول إشارة لاستخدام الكركم فى علاج الإصابة بالكوكسيديا فى الدجاج.