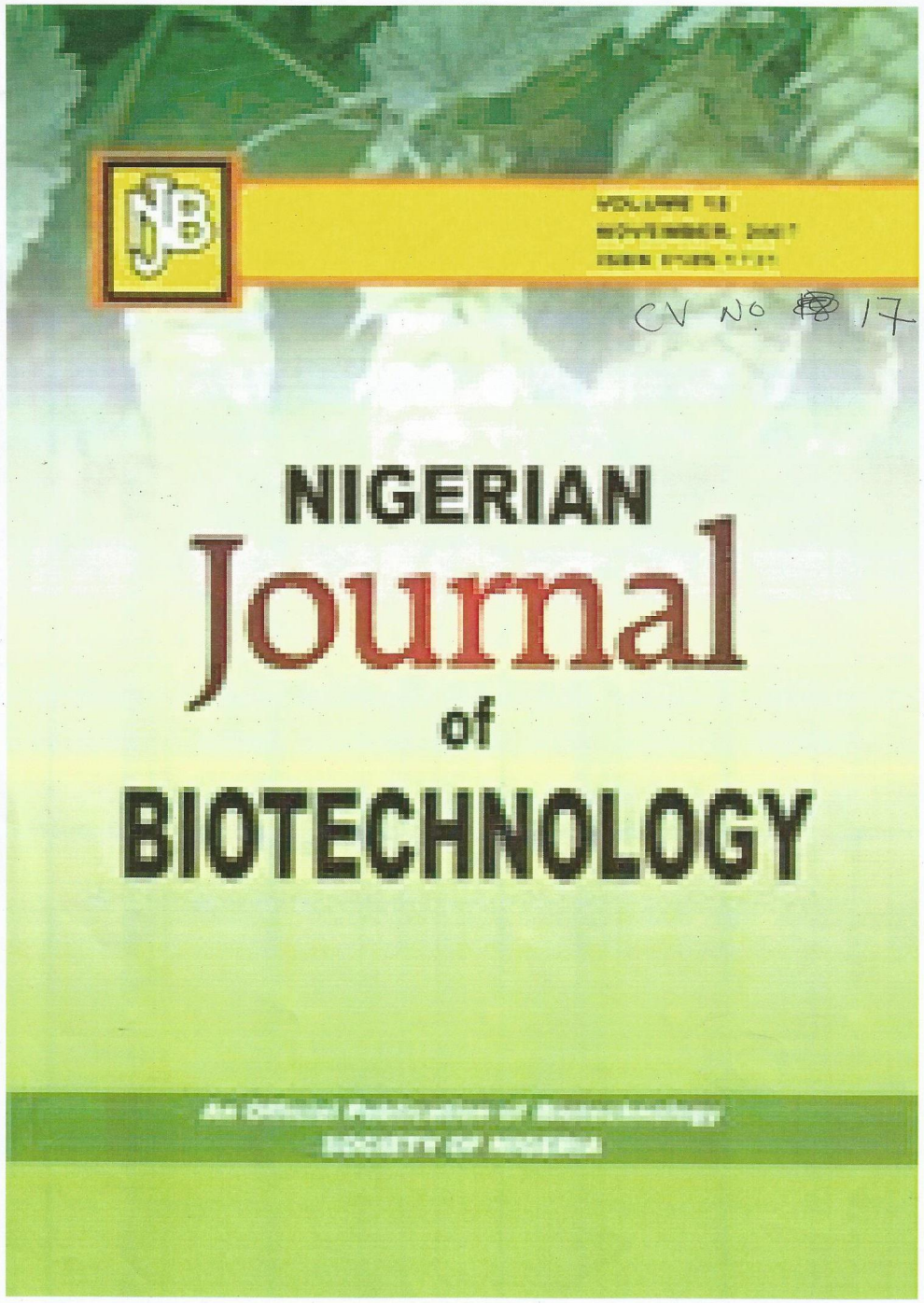
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The anticoccidial property of the aqueous extract of garlic, Allium sativum, in broiler birds.

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Abstracts

Thirty five-week old broilers, reared from day one under controlled conditions, were studied for their response to treatment with aqueous garlic extract when infected experimentally with coccidiosis. The birds were grouped into five. Groups I to III are the controls (uninfected, infected subclinically and clinically, respectively) while groups IV and V are the tests (infected subclinically and clinically respectively, and treated with 200 mg/ml/100 gm body weight aqueous garlic extract). Faecal analysis showed a progressive increase in oocysts from day one for groups II and III with mortalities recorded in group in from day 6. There was a sharp increase (P0.05) in the number of oocysts in groups IV and V and a gradual decrease from days 9 and 7, respectively. Coccidial infection inhibited serum lipase activity while treatment with aqueous garlic extract activated the enzyme activity (P0.05). It is concluded that aqueous garlic extract actually has anticoccidial property and possibly acts by inducing the maturation and release of oocysts.

THE ANTICOCCIDIAL PROPERTY OF THE AQUEOUS EXTRACT OF GARLIC, ALLIUM SATIVUM, IN BROILER BIRDS

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ABSTRACT

Thirty five-week old broilers, reared from day one under controlled conditions, were studied for their response to treatment with aqueous garlic extract when infected experimentally with coccidiosis. The birds were grouped into five. Groups I to III are the controls (uninfected, infected subclinically and clinically, respectively) while groups IV and V are the tests (infected subclinically and clinically, respectively, and treated with 200mg/ml/100gm body weight aqueous garlic extract). Faecal analysis showed a progressive increase in oocysts from day one for groups II and III with mortalities recorded in group in from day 6. There was a sharp increase ($P<0.05$) in the number of oocysts in groups IV and V and a gradual decrease from days 9 and 7, respectively. Coccidial infection inhibited serum lipase activity while treatment with aqueous garlic extract activated the enzyme activity ($P<0.05$). It is concluded that aqueous garlic extract actually has anticoccidial property and possibly acts by inducing the maturation and release of oocysts.

KEY WORDS: **Coccidiosis, Oocysts, *Allium sativuum*, Serum lipase activity.**

INTRODUCTION

The global poultry industry has to contend with the threat of many diseases. The most important one attributable to parasites is coccidiosis (Jenkins *et al.*, 1991). Coccidiosis is an infectious protozoan disease caused by any combinations of seven species of *Eimeria spp* (Adewuyi, 1986; Liliehoj and Trout, 1993). Sporulated oocysts when ingested by susceptible hosts initiate the infective cycle. The disease is cosmopolitan in distribution and occurs in all kinds of animals.

The development of *Eimeria spp* is almost exclusively intestinal (Ball *et al.*, 1989), though the parasites have been detected in such tissues as lymph nodes (Lindsay et al., 1990), blood, lung and liver (Ball *et al.*, 1990). The clinical signs vary somewhat depending on the specie of the causative organism. Usually infected birds become depressed, tend to huddle, consume less feed and water and consequently have decreased weight gain and feed efficiency. Diarrhoea is a common feature. Bloody droppings may be observed with *E. necatrix* and *E. tenella* infections (Ajayi and Umoren, 1984).

Approximately 30 billion chickens are reportedly reared worldwide annually (Anon, 2001). Since every flock of chickens is at risk from coccidiosis, they must be protected. The most current prophylactic method is the use of "Paracox", a coccidiosis vaccine developed in 1989 (Rose, 1989). However, in the event of outbreak of the disease, there are now many coccidiostats and anticoccidial preparations like amprollium. However, the increasing emergence of drug-resistant strains of coccidia has forced attention towards the search for alternative control strategies that are locally available, reproducible, environmentally-friendly, cheap and effective. This work is aimed at verifying the claims by local farmers that aqueous garlic extract is effective in controlling avian coccidiosis.

MATERIALS AND METHODS

Plant Material

Garlic, *Allium sativuum*, was purchased from Jos. The outer covering was removed manually with a sharp knife and the garlic dried in a hot air oven (AE 166) at 50°C. The dried bulbs were pounded in a mortar with pestle, sieved and the dried powder used for the study.

Preparation of Aqueous Garlic Extract

20gm of the dried powdered garlic was stepped in 100ml distilled water for 12 hours, filtered and the filtrate dried using a rotary evaporator. The percentage, yield was 15%.

Management of Experimental Birds

30 day-old broiler chicks purchased from ECWA Farms, Bukuru and reared in deep litter system for five weeks were used for the study. They were given feed (ECWA Farms Bukuru) and tap water ad libitum and vaccines at appropriate times. No coccidiostats were given.

Experimental Infection

The birds were divided into five groups. Group I is the control (uninfected, untreated). Group n is the subclinical control (infected, untreated), Group m is the clinical control (infected,'untreated). Group IV is the subclinical test (infected and treated) while Group V is the clinical test (infected and treated). Oocysts isolated from faecal samples, collected from Farms in Bukuru and Vom, according to the method described by Malcolm (1976), were used in infecting the birds. The oocysts were mixed with the poultry feed and administered to the birds ad libitum for two days according to Ajayi (1995; Personal Communication). The subclinical groups (n and IV) were given proper hygiene and adequate feed to maintain low and natural level of infection. For the clinical groups (in and V), the litter was left unchanged and the birds poorly fed for the infections proceed to a clinical stage. Each group was kept in a different cage.

Administration of Aqueous Garlic Extract

The subclinical test group started receiving treatment on completion of infection while the clinical test group was allowed to manifest all the symptoms of infection before treatment started. The aqueous garlic extract was administered orally with syringe without needle in a dose of 200mg/ml in a total volume of 5ml per bird per day for 5 days.

Collection of Faecal Samples and Oocyst Count

Faecal samples were collected randomly from each cage on alternate days and the coccidial oocysts recovered by salt floatation method (Malcolm, 1976). The oocysts were counted microscopically.

Blood Collection and Serum Preparation

Blood samples were collected from the birds through the wing vein with sterile syringe and needle. These were stored at 12°C for one hour in the refrigerator before centrifuging at 2,500 rpm for 5 minutes. The supernatant was recovered and stored at 0°C, if not used immediately for enzyme assay.

Enzyme Assay and Protein Determination

Serum lipase activity was measured according to the titrimetric method described by Tietz (1987). The principle is based on the hydrolysis of olive oil with serum enzyme and titrating the liberated fatty acid to thymolphthalein endpoint with sodium hydroxide. Enzyme activity was expressed in units per mg protein. The serum protein was determined using biuret reagent according to the method described by Plummer (1987).

Statistics

The result are presented as mean ±, standard deviation. The results were compared using Students t-test (Snedecor and Cochran, 1987).

RESULTS

Table 1: Effect of Aqueous Garlic Extract on Coccidiosis in Broiler Birds.

Day	No. of Oocysts/gm Faecal Sample				
	Group I	Group II	Group III	Group IV	Group V
1	5.00+0.50	400.50+20.10	800.65+45.53	1900:801240.5"	3400.4+420t:
3	7.15+0.25	410.20±15.10	900.10±70.7-	2340.70+200.01'	5420.51400'
5	10.75+1.20	440.75+25.3	650.25±75-2	2620.1+400[b]	3300.20+300
7	8.15±2.05	460.50±30.1	-	1340.5+4201'	242.75+300
9	5.17+1.05	480.25+22.5	-	233.65+15.2	173.1 ±20.5
11	5.25+0.50	540.50+35.4.	-	40.05+5.0	27.70±5-05

Values are mean + SD for three independent determinations.
[a] Significantly higher as compared lo the corresponding group II (P<0.05)
[b] Significantly higher as compared to the corresponding group II (P<0-05)
[c] Significantly higher as compared to the corresponding group III (P<0.05)

Table 1 summaries the effect of aqueous garlic treatment on birds infected with subclinical and clinical coccidiosis. Though there were increases in oocyst count as the subclinical disease condition progressed (Group II), it was not statistically significant. The oocyst count for group III was significantly higher (P <0.05) when compared against group n but the birds started dying by the fourth day and attained 100% mortality by the seventh day. Treatment with aqueous garlic extract (groups TV and V) showed very sharp rise (P<0.05) in oocyst count when compared against the corresponding infected but untreated birds (groups II and III) but which gradually dropped as the treatment progressed.

Table 2: Effect of Aqueous Garlic Extract on the Serum Lipase activity

Day	Enzyme Activity (Units/mg Protein)				
	Group I	Group II	Group III	Group IV	Group V
1	0.119+0.001	0.027±0.003[a]	0.125±0.002	0.205±0.0051:	0.078±0.006
3	0.121±0.010	0-017^0-002[a]	0.010±0.001[b]	0.217±0.001(:	0.110±0.003d
5	0.119+0.005	0.015±0.002[a,c]	-	0.225±0.002[c]	0.120±0-005

Values are mean j. SD for three independent determinations.
[a] Significantly lower as compared to Group I, P<0.05
[b] Significantly lower as compared to Group I, P<0.05
[c] Significantly higher as compared to the corresponding group III (P<0.05)
[d] Significantly higher as compared to the corresponding group III (P<0.05)
[e] Significantly lower as compared to Day I, P<0.05

Table 2 summarizes the effect of aqueous garlic extract on the serum lipase activity. of birds experimentally infected with coccidiosis. Birds in group n showed a statistically significant decreased enzyme activity (P<0.05) when compared against the healthy birds (Group I) and a statistically significant decrease as the disease progressed (P<0.05). Birds in group HI also showed a decrease in enzyme activity when compared against Group I (P<0.05). On treatment with aqueous garlic extract (Groups IV and V), the enzyme activity was enhanced (P<0.05).

DISCUSSION

The results obtained for the oocyst counts in this study compared favourably with those of Malcolm (1976).

The observed progressive increase in the number of oocysts excreted in the faecal samples of the infected birds could be as a result of progress in the infection. However, the sharp increase in the number of oocysts as a result of the administration of aqueous garlic suggests that the extract affects the maturation of the life stages of *Eimeria spp.* thereby shortening the prepatent period. The prepatent period is reported to be four days (Liliehoj and Trout, 1993). The enhanced release of these oocysts in the faecal samples may be as a result of the associated diarrhoea. The observed decrease in the oocyst count as the treatment progressed may be an indication of recovery of the birds from the disease.

In an effort to explain the observed enhanced maturation and release of faecal oocysts in birds treated with the aqueous garlic extract, we assayed the serum lipase activity. The observed statistically significant ($P<0.05$) decrease in the enzyme activity (Table 2) as a result of coccidial infection suggests that the pancreas is not affected by the disease. However, the elevated serum lipase activity as a result of treatment with the plant extract may be due to the contraction of the sphincter of Oddi of the pancreas. Substances like the opiates which contract the sphincter of Oddi are known to cause elevation of serum lipase if activity (Tietz, 1987). Another possible explanation for the observed enhanced release of faecal oocysts on administration of the plant extract may be the presence of high concentration of tannin in garlic (Mgbojikwe et al., 2000). Tannins are known to have effects on the intestine. When the stomach and bowels are relatively empty, tannic acid coagulates the superficial layers of the gastrointestinal mucus membrane. *Eimeria* species particularly *E. brunetti* infects the intestine with most damage being caused to the lower ileum, colon and proximal areas of the caeca (Liliehoj and Trout, 1993).

When compared against conventional coccidiostats, aqueous garlic extract may not be acting like amprollium which is known to antagonize thiamine thereby depriving the parasite of this essential vitamin (Frank, 1985) nor the sulphonamides which are no longer in demand because of their side effects which may include kidney damage (Anon, 2001).

In conclusion, aqueous garlic extract is effective against avian coccidiosis and its mechanism of action may be through the induction of maturation of the life stages of the causative organism, shortening of the prepatent period and flushing out the oocysts. More work, however, needs to be done to determine the active principle and possibly the species of the Eimeria against which it is active.

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