THE EFFICACY OF A COMMERCIAL HERBAL FORMULATION IN THE PREVENTION OF COCCIDIOSIS UNDER NATURAL EXPOSURE TO Eimeria SPECIES

 \mathbf{BY}

MUSA, Hassana

MTECH/SAAT/2018/8001

DEPARTMENT OF ANIMAL PRODUCTION, SCHOOL OF AGRICULTURE AND AGRICULTURAL TECHNOLOGY, FEDERAL UNIVERSITY OF TECHNOLOGY

MINNA

SEPTEMBER, 2021

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A THESIS SUBMITTED TO THE POSTGRADUATE SCHOOL FEDERAL
UNIVERSITY OF TECHNOLOGY MINNA, NIGERIA IN PARTIAL FULFILMENT OF
THE REQUIREMENTS FOR THE AWARD OF THE DEGREE OF MASTER OF
TECHNOLOGY IN ANIMAL PRODUCTION

SEPTEMBER, 2021

ABSTRACT

This study was under taken to determine the Efficacy of a Commercial Herbal Formulation in the Prevention of Coccidiosis under Natural Exposure to Eimeria Species in Animal Production Teaching and Research Farm during the period of March to May 2020. A total of 180 day old broiler chicks were randomly divided in to three treatments, in a completely randomized design with 3 replicates containing 20 birds each. The groups were designated T1, T2, and T3 respectively. Treatment one was administered Ruchamax at the rate of 15g in water, treatment two was given amprolium at 100g and treatment three was served as control. Data collected on weight gain, carcass characteristics, feed intake, feed conversion ratio, nutrient digestibility and hematological parameters were subjected to analysis of variance using SPSS statistical software. The results showed that the carcass characteristics, the nutrient retention and weight gain were not significantly (p>0.05) different. Significant (p<0.05) differences were observed in hematological indices between the treatment groups. The serum parameters between the treatments were not significantly different. The results of the feacal samples showed that Ruchamax was effective against coccidiosis due to the reduction in the level of infection along the weeks. It was concluded that the weekly coccidiosis result shows the safety and efficacy of Ruchamax as an anticoccidial agent. The findings of the present study showed that 15g of Ruchamax produced comparable result to amprolium in control, treatment and prevention of coccidiosis in broiler chickens.

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CHAPTER ONE

1.0 INTRODUCTION

Poultry meat and eggs are essential foods for satisfying the nutritional requirements of the ever-growing human population. Poultry represent nearly 33% of global meat production and is a source of protein that plays an essential role in human nutrition (Abbas *et al.*, 2010).

Chicken is the most common type of poultry in the world. Chicken meat provides not only high quality protein, but also essential vitamins and minerals (Yousaf, 2018). Also, chicken meat has high level of vital polyunsaturated fatty acids, like omega (n)-3, which can be effortlessly digested and efficiently absorbed (Kharde and Soujanya, 2014). Adding to dietary value, chicken is further common owing to the fact that it is quite easy to prepare and is cheaper than other types of meat. Chickens are extensively consumed as a source of meat either directly (as broiler chickens) or indirectly (as spent layers). The term broiler refers to chickens that have specifically been bred for meat, they develop rapidly. Broiler strains are created from hybrid crosses between Cornish White, New Hampshire and White Plymouth Rock (Lawal *et al.*, 2016).

Coccidiosis is a major disease problem in the poultry business, it is a parasitic disease of the intestinal tract of animals caused by single-cell protozoa. Coccidian organisms can infect birds, livestock and humans as they are typically species-specific (Gharekhani *et al.*, 2014). Coccidiosis is initiated by any of the species of the genus Eimeria, discretely or in combination. It leads to the extensive damage of the intestinal epithelium which results in reduced food efficiency and body weight gain (Ali *et al.*, 2019). The most common and

pathogenic species that affects the poultry industry worldwide is the *E. tenella* with 100% morbidity and a high mortality due to extensive destruction of the digestive tracts of chickens (Lawal *et al.*, 2016).

Coccidiosis is largely controlled by the sanitary measures and the use of chemotherapeutic agents or chemical anticoccidial agents Nonetheless, the development of drug resistance to all the drugs introduced so far by the causative parasites and the intensifying cost of drug development have significantly reduced the commercial incentive to develop new chemical anticoccidial. Additionally, currently, buyers demand poultry products that are free from residual antiparasitic drugs. Therefore, the development of alternate, safer and ecologically friendly anticoccidial agents have become priority in most parts of the world (Ali *et al.*, 2019).

Ruchamax is a powerful herbal formulation, which contain 28 different herbs and some minerals. It is used as an appetizer, restorative, carminative, stomachic, and stimulant. It is also aid optimum absorption and utilization of nutrients and hence improves feed conversion ratio, productivity and weight gain (Walia *et al.*, 2011). Ruchamax® is mostly used in ruminant and pseudo ruminant to treat poor digestion, impaired ruminal function, debility, dyspepsia and convalescence but there is very scarce literature on its use on Poultry.

Each 15g of Ruchamax contains; *Acorus calamus*–0.20, *Andrographis paniculata*–0.80, *Azadirachta indica* -0.50, *Balanites roxburghii* –0.50, *Calotropis procera* – 0.08, *Centratherum anthelmenticum* –0.50, *Commiphora mukul* – 0.20, *Curcuma longa* – 0.20, *Eclipta alba* – 0.50, *Embelia ribes* -0.20, *Gardenia gummifera* -0.20, *Phyllanthus emblica*-0.50, *Piper longum* -0.10, *Piper nigrum* -0.10, *semecarpus anacardium* -0.04, *Solanum*

nigrum-0.40, Terminalia chebula -0.50, Tinospora cordifolia -0.10, Terminalia belerica, Allium sativum -0.50, Zinziber officinale -1.00, Trychyspermum ammi -0.50, Trigonella foenum-graecum-0.50, Woodfordia fruticose -0.40, Copper sulphate -0.05, Ferrous sulphate -0.20, Sodium chloride -1.0, Ammonium chloride-0.20 (Walia et al., 2011). These individual constituent herbs are scientifically well known to possess appetizer, restorative, carminative, stomachic and herbs activity.

1.1 Statement of the Research Problem

Growing threats of coccidian specie resistance and antimicrobial drug residues has put a lot of pressure on human health and in general the food industry, intensified efforts in checkmating microbial resistance has led to condemnation of certain meat products especially those containing intolerable levels of antimicrobials thereby affecting the agricultural value chain. Intensive poultry production also increase the spread and prevalence of coccidiosis. Additionally, strong evidence of the presence of the residues of coccidiostats in the meat of treated birds has not been adequately addressed. Therefore, there is a strong desire to use some natural alternative agents to replace the existing coccidiostats.

1.2. Justification of the Study

The present study was carried out to enlighten the poultry farmers to attain the knowledge of coccidiostats of plant origin as alternative to those of chemical origin in order to prevent harmful drug residues in the meat of treated birds, reduce coccidia specie resistance and also to reduce farmers input cost. The need to try Ruchamax on poultry arise as a result of the fact that it contains natural herbal ingredients that have been used alone or in

combination on poultry, and was scientifically proven to be safe and effective as an anticoccidial agents.

1.3. Aim and Objectives of the Study

The aim of the study was to determine:

The effect of administering Ruchamax on coccidia organisms and performance of broiler chickens.

The objectives of the study were to;

- i. Determine the effect of Ruchamax on performance of broiler chickens
- ii. Assess the effect of Ruchamax on heamatology and immunoglobulin profile of broiler chickens.
- iii. Evaluate the effect of Ruchamax on nutrient retention of broiler chickens and
- iv. Identify the anticoccidial effect of Ruchamax formulation on coccidia organisms in broiler chickens.

CHAPTER TWO

LITERATURE REVIEW

2.1. Causative Agents of Coccidiosis

2.0

Avian coccidiosis is a common prevalent ailment allied with substantial fiscal losses to poultry farmers globally. It is capable of affecting birds raised for any production purposes and in any production systems. The organisms that cause poultry coccidiosis is comprised of a wide range of single-celled protozoans of the genus Eimeria. Nine Eimeria species, E. acervulina, E. brunetti, E. maxima, E. necatrix, E. praecox, E. mitis, E. tenella, E. mivati, and E. hagani, have been identified from chickens. Eimeria brunette, Eimeria maxima, Eimeria necatrix, Eimeria tenella are the most pathogenic (Abudabos et al., 2017).

Each of the nine species occurs in a single host species or a cluster of closely allied hosts and they raid the lining of the intestine or ceca, causing death or decreased production in poultry. Infection by coccidian organisms in ample amounts produces clinical manifestations of the disease (Gharekhani *et al.*, 2014). Likewise, subclinical infections, which considerably have influence on productivity and food security, are common due to extensive drug resistance, high parasite prevalence, and ecological perseverance (Yang *et al.*, 2019).

Avian coccidiosis is recognized as the parasitic disease with the utmost fiscal impact on poultry productions globally. It is a form of protozoan maladies characterized by dysentery, enteritis, emaciation, drooping wings and poor growth. Feed and water intake are reduced. Weight loss, development of culls, declined egg production, and high morbidity and mortality may accompany outbreaks (Barbour *et al.*, 2015). The disease has a great

economic impact in poultry productions partly as a result of resistance of the organisms to anticoccidial drugs (Ghayour-Najafabadi *et al.*, 2018).

Natural products are a principal basis of novel natural drugs and their usage as an alternate medication for management of numerous diseases has been increased in the past few years. In contrast to the formulated drugs, the herbs and spices have less or no side effects. They are likewise more affordable, indicate better patient tolerance and are eagerly accessible for low socioeconomic populace (Abudabos et al., 2017). The use of innocuous and effective medicinal plants can lessen producers' input costs, preserve the resource base, enhance biodiversity and safeguard animal wellbeing (Allen *et al.*, 1997).

2.2. Poultry House Management in Control of Coccidiosis

Eimeria has a great reproductive capacity, hence virtually it would be very problematic to keep the environment around the birds free from these parasites. The international control of chicken coccidial infections in the poultry industry is becoming more difficult owing to the high stocking density of the flocks. Besides, the current global climate warming increases coccidial proliferation (Ziomko *et al.*, 2005). Likewise, the resilient nature of the coccidial oocyst's wall aids shielding the oocyst from desiccation and from the harsh effects of chemical disinfectants, hence ensuring its long term survival in the farm environment. This survival helps in its consequent distribution across the diverse poultry houses, through numerous routes including mainly workers, equipment, rodents, and/or insects (Muthamilselvan *et al.*, 2016). Study showed that the greater perils of coccidiosis are ecological and management factors, in addition to sanitary measures that are exercised on the farm, including visitors, presence of feeders and drinking systems that are difficult to

clean, and the high likelihood of carryover of this parasite from earlier infected flocks (Adulugba *et al.*, 2017).

2.3. The Coccidian Oocyst

The oocyst is deliberated an unusually hard and obstinate structure. It is resilient to mechanical and chemical destruction and to proteolytic degradation (Wondimu et al., 2019). In actual fact, oocysts continue to be viable and infectious after treatment with diluted sodium hypochlorite or after storage with the strong oxidizing agent potassium dichromate (1 to 2%) or with sulfuric acid. In addition, each layer of the oocyst wall reacts very differently to chemical treatments (Pawestri et al., 2020). The oocyst wall is a structure that is poised of a bilayer, with an outward layer that can be as thick as 500 to 600 nm but that is eventually compressed to 200 nm or less. An inward zone of about 40 nm splits the outer and inner layers, with the latter being about 40 nm dense (Quiroz-Castañeda and Dantán-González, 2015). The oocyst is non infective when being in its unsporulated state and becomes infective when it sporulates (Abbas et al., 2010). It has been established that an unsporulated oocyst can subsist up to seven months in the ceca material and that the sporulated oocyst can subsist up to 602 days in the exogenous environment (Barelli et al., 2018). Essentially, the composition of the two walls confers the oocyst with an exceptional resistance. The oocyst wall comprises largely of proteins and lipids. Additionally, carbohydrates covalently bonded to proteins have also been reported, with varying percentages (1.5– 19%). The inner wall is poised of a protein matrix entrenched with as well as layered with lipids, while the outer wall is composed of quinone tanned proteins as well as proteintyrosine crosslinks. Although the presence of cysteine has been described, no cysteine

bridges have been noticed (Pop *et al.*, 2019). The proteins provide the oocyst with great structural stability against high temperatures because the oocysts are sensitive to extreme heat or cold and desiccation. It is probable that the external coating shields the oocyst from mechanical destruction while the lipid-rich internal coating shields it from biochemical attack (Elkhtam *et al.*, 2014).

2.4. Control of Coccidiosis by Holistic Approaches of Natural Products

Anticoccidial drugs were used successfully over the past 50 years. Their use resulted in a swift development of poultry industry and the affordable poultry products to the consumers. Nonetheless, the growing resistance to Eimeria species has stirred the efforts to search for alternative control strategies (Elkhtam *et al.*, 2014). Holistic approach is presently searched and it is attempted by the application of herbal products for control of coccidiosis. The foremost researched herbs and their studied anticoccidial properties are discussed below.

2.4.1. Artemisinin

Artemisinin is a Chinese herbal extract from Artemisia annua (annual wormwood), and A. sieberi. Artemisinin has an antimalarial effect in humans; its mode of action is ascribed to the fact that artemisinin has an endoperoxide group that act against the parasite by producing extremely reactive free oxygen radicals (oxidative stress) (Muthamilselvan et al., 2016).

Obi *et al.*, (2019) was the first to report that A. annua extracts have an anticoccidial activity against E. tenella, as deduced from the herb's impact on improved weight gain, improved feed conversion and reduced lesion scores. Dried *Artemisia annua* leaves were evaluated for their anticoccidial activity in poultry as feed additives. The leaves were included in poultry diet at a 5% concentration for a period of 3 weeks equivalent to 17 ppm

of pure Artemisinin. This supplemented diet resulted in a significant protection against lesions of E. tenella in poultry. When pure artemisinin was added to the feed for a period of 4 weeks at different concentrations of 2, 8.5 and 17 ppm, it was able to significantly reduce the oocysts output from single and dual spp. infection by E. t*enella* and *Eimeria acervulina* (Allen and Fetterer, 2002).

Similar study by Health *et al.*, (2006) reported the shielding activity of artemisinin against E. *tenella* and E. *acervulina*, but not against E. *maxima*. When broiler chickens are fed the extract at concentrations of 1 or 2.5mg/kg. Artemisinin produces reactive oxygen species (ROS) through degradation of iron-implicated peroxide complex and, therefore, induced oxidative stress. Likewise, ROS impede sporulation and cell wall development in Eimeria species. Oxidative stress results in imbalance of oxidant or antioxidant species in the host due to microbial and parasitic infections including coccidiosis (Ola-fadunsin, 2013).

A study to evaluate the effect of *Artemisia sieberi* extracts on 21-day old broiler chickens infected with E. *tenella*, E. *maxima*, E. *necatrix*, and E. *acervulina*. indicated that chickens treated with A. *Sieberi* extract has reduced oocysts count per gram of feces as well as better growth performance parameters such as feed intake and weight gain when equated with the effects observed with monensin treatment (Ali *et al.*, 2019). Related results were recorded when day old broilers infected with E. tenella were treated with granulated extract of A. *sieberi* obtained from petroleum ether (Health *et al.*, 2006).

2.4.2. Aloe vera

Aloe vera has been mentioned in the literature for its healing and therapeutic effects, possessing more than 75 biologically active compounds in it. The Aloe vera immunostimulatory properties in broilers infected with Eimeria species was reported by

Abbas *et al.*, (2010). Aloe vera extracts improved the humoral and cellular immune reactions in chickens. It stirred the macrophages-phagocytic activity, cytokines production, and induced better antibody production. Additionally, Aloe vera reduced oocyst output and lesion scores. The results of this research were in agreement with those of Yang *et al.*, (2019), who tested the effects of Aloe vera against E. maxima infection. These researchers indicated that oocyst shedding and lesion scores were considerably reduced in the Aloe vera treated groups; though, no data were presented in relation to production performance of the chicken included in their experimental design (Pd *et al.*, 2018).

2.4.3. Azadirachta indica (Neem)

Neem comprises numerous active ingredient like nimbidin, sodium nimbolide, gedunin, azadirachtin, mahmoodin, Gallic acid, epicatechin and catechin,margalone, margolonone and isomargolonone, cyclictrisulphides, polysaccharides GIa, GIIa, GIIIa, GIb,NB-II peptidoglycon which all add to its useful medicinal effects (Kharde and Soujanya, 2014). Neem leaves and its components have been proven to exhibit immunomodulatory, anti-inflammatory, antihyperglycemic, antiulcer, antimalarial, antifungal, antiviral, antioxidant, antimutagenic, anticarcinogenic properties (Rajesh and Veterinary, 2019).

This plant is usually found in African and Asian countries, and it is recognized for its therapeutic effects against various infectious diseases, including coccidiosis. The neem fruit, at an inclusion rate of 150 g/50 kg feed, had an anticoccidial activity against E. tenella, as concluded from oocyst output and mortality in broilers (Abbas et al., 2010). The water extract of neem leaves has also an anticoccidial activity against E. tenella similar to that obtained by Baycox drug (Indrasanti et al., 2017). Neem had an anticoccidial effect against a mixed infection by different species of Eimeria. This effect was equivalent to that

produced by Amprolium drug (Biu *et al.*, 2006). Neem also improved cellular immune response and antibody production (Biu *et al.*, 2006). More research on the mode of action and dose-related adverse effects of neem on birds is necessary because the bitterness may affect feed intake, and thus warrants further investigation.

2.4.4. Garlic

Garlic comprises at least 33 sulfur compounds, some enzymes, 17 amino acids and minerals. The Sulphur compounds are responsible both for garlic's pungent odor and many of its medicinal properties like lowering cholesterol level. Studies are also showing immunostimulatory function of garlic extracts. Injury to the garlic bulb triggers the enzyme alliinase, which metabolizes alliin to allicin. Allicin has antimicrobial effects against many viruses, bacteria, fungi and parasites (Health *et al.*, 2006). Ola-fadunsin, (2013) in his study observed improvement in the hematological indices of the infected birds treated with extracts of Moringa oleifera, garlic and combination of Azadirachta indica and Khaya senegalensis respectively. Allen & Fetterer, (2002) also reported that the supplementation of active ingredients of garlic (propyl thiosulphinate oxide and propyl thiosulphinate) had decreased faecal oocysts excretion and greater antibody response against Eimeria acervulina in broiler chickens, he found that garlic metabolites enhance chickens' production performances and reduce the oocyst output in chickens challenged with E. acervulina, due to a direct cytotoxic effect on the coccidian sporozoites (Dar *et al.*, 2014).

2.4.5. Moringa oleifera

Moringa oleifera is one of the most important medicinal plants and it is the most widely cultivated specie of the family Moringaceae. Every part of Moringa is said to have beneficial properties that can serve. Numerous parts of Moringa oleifera minerals, vitamins and are a good source of protein, beta carotene and amino acids. The Moringa plants provides a rich and rare combination of quercetin, zeatin, caffeoylquinic acid and kaempferol. Various part of the Moringa plants such as the leaves, roots, seed, bark, fruits, and flowers act as cardiac and circulatory stimulants, antioxidant, antibacterial, antifungal and antimalarial properties, and are being used for the treatment various diseases. In addition Ola-fadunsin, (2013) reported the use of Moringa oleifera acetone extract on broiler chickens naturally infected with Eimeria species (Elkhtam *et al.*, 2014).

2.4.6. Ginger

Ginger has been reported to enhance the growth performance and digestibility in broiler chickens and effective in treating and controlling coccidial infection (Zhang *et al.*, 2008).

2.5. Plants and Compounds that Modulate Host Immunity against Eimeria

From an evolutionary point of view, birds possess a complete immune system comprising of innate and adaptive immune responses. Both immune responses are responsible for coccidial clearance and vaccine immunization. Medicinal plants often have immunomodulatory compounds which enhance antimicrobial immune responses to sustain homeostasis of poultry health. Hence, immunoregulatory plant extracts and compounds could be used as an alternative means to reinforce immune response against avian coccidiosis (Wondimu *et al.*, 2019).

2.5.1. Arabinoxylans, wheat (*Triticum aestivum*), and sugar cane (*Saccharum officinarum*)

Awais *et al.*, (2011) reported that arabinoxylan, a bioactive compound from wheat bran, improved coccidiosis in chickens as shown by body weight, oocyst count, and gut lesions. In contrast, Awais and Akhtar showed that dissimilar extracts of sugarcane juice and bagasse protected against coccidiosis in chickens as indicated by body weight gain, oocyst shedding, lesion score, and anticoccidial indices. The data from both findings discovered that wheat bran arabinoxylan and sugar cane offers host protection against Eimeria infection through natural and adaptive immune response. Cell-mediated immunity appeared to be an important factor in response to coccidiosis in birds when likened to humoral immunity (Awais *et al.*, 2011).

2.5.2. Propyl thiosulfinate and propyl thiosulfinate oxide

A study indicated that garlic compounds, propyl thiosulfinate (PTS) and propyl thiosulfinate oxide (PTSO), could alter the expression levels of 1,227 transcripts related to intestinal intraepithelial lymphocytes (IEL) in chickens. PTSO/PTS was shown to activate transcription factor, NF- κ B, which plays a key role in regulating the immune response upon infection (Elkhtam *et al.*, 2014).

Therefore, it seems that a combination of PTSO and PTS rendered chickens more resistant to experimental E. *acervulina* infection and augmented adaptive immunity, including a higher antibody response and greater splenocyte proliferation, compared with control chickens. Another in vitro study showed that PTS could stimulate splenocyte proliferation and directly kill the sporozoites, pointing to the same conclusion (Elkhtam *et al.*, 2014).

2.6. Plants and Compounds with Multiple Mechanisms to Inhibit Coccidiosis

2.6.1. Curcumin and curcuma longa

One study showed that C. *longa* exhibit anticoccidial activity (Muthamilselvan *et al.*, 2016). Another study reported that curcumin (diferuloylmethane), an active compound in C. *longa*, consistently destroyed sporozoites of *E. tenella*. Likewise, a combination of A. *annua* and C. *longa* indicated anticoccidial efficiency in broilers challenged with a mixture of E. *acervulina* and E. *maxima*. Additionally, curcumin was reported to improve coccidiosis resistance as demonstrated by increase in body weight and reduced oocyst shedding and gut lesions. Consistently, curcumin elevated host humoral immunity to Eimeria species and reduced gut destruction in poultry (Quiroz-Castañeda and Dantán-González, 2015).

2.6.2. Polyacetylenes and *Bidens pilosa*

Yang *et al.*, (2019) reported that B. *pilosa* exhibited anticoccidial activity in chickens infected with E. *tenella* as compounds by survival rate, fecal oocyst count, gut pathology, body weight, and bloody stool. Though the active compounds in B. pilosa responsible for anticoccidial action are not known, this plant is a rich source of phytochemicals, such as 70 aliphatics, 60 flavonoids, 25 terpenoids, 19 phenylpropanoids, 13 aromatics, 8 porphyrins, and 6 other compounds. Interestingly, one polyacetylene (1-phenyl-1, 3-diyn-5-en-7-olacetate) and one flavonoid (quercetin-3, 3-dimethoxy-7-0-rhamnoglucopyranose) in this plant have been proposed to be active compounds against the protozoan parasite, Plasmodium (Abudabos *et al.*, 2017). However, the identity of the active compounds needs to be further ascertained (Muthamilselvan *et al.*, 2016). Compared to anticoccidial drugs, B. *pilosa* was shown to produce little or no drug resistance in Eimeria. Botanicals developed low resistance in Eimeria probably because different compounds target multiple pathways related to drug resistance (Yang *et al.*, 2019).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Area of Study Description

The study was conducted at the Animal Production Teaching and Research Farm, in the Department of Animal Production, School of Agriculture and Agricultural Technology, Federal University of Technology Minna, Gidan Kwano campus. Minna is sited in the southern guinea savannah vegetation zone of Nigeria and is described by two seasons; wet (March to October) and dry season (November to March). It has altitude of 75cm above ocean level with a land region of 6784 kilo meter square and lies between scope 9°c 37' North and longitude 6°c 33 East, the mean yearly precipitation is 1300mm take from an especially long record of 50 years, temperature once in a while falls beneath 22 degree Celsius, the top are 44 degree Celsius (February to March) and 35 degree Celsius (November to December) (Afolayan *et al.*, 2017).

3.2 Experimental Animals

A total of 180 healthy day old broiler chickens of both sexes was obtained from Mokwa hatchery Niger state and brooded under standard conditions for eleven days before the commencement of the study. Before the arrival of the birds the pen house was thoroughly washed and disinfected with izal disinfectant, the cobwebs were removed and brooding devices were kept in strategic positions. The Brooding was done for 21 days. The birds were fed standard pelletized Commercial broiler starter feed. Wood shave was used as bedding material throughout the experiment. Manual feeders and drinkers were used. The diet was provided in two phases consisting of starter phase (0–21days) and *fi*nisher (22–46 days). They were vaccinated against Newcastle disease and infectious bursal disease, the vaccines were administered in their drinking water after 12 hours of water starvation.

3.3 Experimental Drugs

Ruchamax herbal formulation was obtained from Animal care veterinary store in Minna. 15

grams of Ruchamax powder was administered in drinking water at the second, fourth and sixth week of the experiment.

Amprolium (Water soluble powder) a commercially available anticoccidial drug for the routine treatment of avian coccidiosis due to Eimeria species was used to compare the anticoccidial effects of Ruchamax powder. Amprolium acts by interfering with thiamine metabolism in the parasite. The experimental drugs were administered for three consecutive days, and the administration was repeated three times during the 45 days of experiment.

3.4. Experimental Design

The 180 purchased birds was subjected to completely randomized design (CRD) where the birds were randomly assigned to three groups of three replicates with twenty birds per replicate. Treatment 1 was administered Ruchamax powder, while Treatment 2 received amprolium in drinking water against coccidiosis and treatment 3 was served as control. A total number of twelve birds with four birds per replicate was used for serum, heamatology and Feacal sample collection at the end of the experiment.

3.5. Performance Trait

The feed was weighed and offered ad libitum to all the experimental animals. The leftover feed was collected and weighed the next morning before offering fresh feed. Body weight gain was recorded at the end of every week. Weight gain was calculated by subtracting the initial weight of birds from the final weight of the birds. Feed intake was calculated as the difference between feed supply and feed residues. Feed conversion ratio was calculated by dividing feed intake by weight gain. The feed conversion ratio (FCR) was computed at the end of each week and the final FCR was

calculated and note at the completion of experimental duration. The number of dead birds during the experiment was recorded throughout the experiment.

3.6. Feacal Sample Collection

The birds were raised from day old in deep litter while the litter was regularly wetted so that it could be damped enough to cause oocyst sporulation and subsequent infection of the birds. Feacal samples for detection of coccidia organisms were collected at weeks two, four and six and analyzed prior to and after the administration of the experimental drugs at the ministry of livestock and veterinary hospital Minna to determine the level of infection with the coccidial organism. For each of the birds, Feacal samples were collected from droppings where possible with spatula for freshly voided feces, Feacal samples were also collected by inserting a syringe in to the anus of the experimental birds. Each Feacal sample was placed in a prelabeled bottle indicating the treatment and was immediately stored in the cooler and was quickly transported to the laboratory where it was transferred to the refrigerator and stored at 4°C until processed.

3.7. Collection of Blood Samples for Hematological and Serum Parameters

Blood samples were taken from the second week to the sixth week of the study for determination of serum parameters and haematological parameters. Blood samples were collected through the jugular veins or wings of the birds. Two milliliters (mls) of blood sample was collected at the second and third week, one ml for heamatology and one ml for serum. Four mls of blood samples were collected during weeks, four, five and six, two mls was poured in the serum plain bottles and the remaining two mls was poured in anticoagulant bottles for heamatology. Samples were stored in ice after collection and was transferred immediately to the laboratory where it was stored in a refrigerator for 24 hours.

The serum sample was transferred into serum bottle before being transferred into the machine. The serum parameters determined includes Total protein, Albumin, aspartate aminotransferase (AST), Alanine aminotransferase (ALT), Alanine phosphatase (ALP) and globulin while the haematological parameters determined are Haemoglobin, packed cell volume (PCV), red blood cell (RBC), white blood cell (WBC). Differential leukocytes count (DLC) such as Lymphocytes, Monocytes, Eosinophils, Basophils parameters were also determined. Other parameters includes mean corpuscular haemoglobin (MCH) mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC) and Platelet respectively.

3.8. Carcass Yield

At the end of the experimental period, at 46 days of age, a total of six birds with two birds per replicate were sacrificed and used for the carcass yield. The birds were slaughtered and the following parameters were weighed and recorded accordingly; Live weight, Slaughter weight, Dressing weight, Eviscerated weight, Head, Neck, Back, Breast, Thigh, Drumstick, Shank, Wing, Heart, Liver, Spleen, emptied Gizzard and Intestine, Gall bladder, Lung, Trachea, Crop and Provetriculous.

3.9. Digestibility Study

At the expiration of 42 days, three broilers from each treatment making a total of nine birds were selected and arranged in hygienic, discrete and disinfected metabolic cages. Three days of acclimatization were allowed before the commencement of the digestibility study. A known weight of feed, which matched their previous daily feed intake was fed during the metabolic trial. Excreta collection was done daily for a period of four days. The daily excreta voided for each bird was dried overnight while, total collections per bird were

pooled at the expiration of 4 days metabolic trial. Dried excreta samples were used to determine the proximate compositions in Animal Production laboratory.

3.10. Statistical Analysis

The data was compiled using Microsoft excel 2016 and analyzed by analysis of variance (ANOVA)with SPSS (statistical package for social sciences) version 15.0 Groups were compared using the least significant difference (LSD) at P<0.05.

CHAPTER FOUR

4.0 RESULTS

4.1. Growth Performance of Broiler Chickens Administered Ruchamax (0-3 weeks)

The result of growth performance of broiler chickens administered Ruchamax is presented in Tables 4.1. The results showed that final weight, daily weight and the feed conversion ratio (FCR) were not significantly different (p>0.05). The initial weight and feed intake were also not affected statistically among the treatments. The final weight ranged from 403.33 to 413.33g. The average feed intake was higher in the control group although not to a significant effect. The average feed intake ranged from 44.05-56.92g. The average weight gain between the treatment groups did not differ significantly. The daily weight results ranged from 17.25-17.41g respectively. The feed conversion ratio between the treatment groups was not influenced (p>0.05) statistically. The feed conversion ratio results ranged between 3.52-3.32 respectively. The birds administered amprolium had a higher energy and protein intake though not to a significant effect (p>0.05).

4.2. Growth Performance of Broiler Chickens Administered Ruchamax (4-6 weeks)

The result of growth performance of broiler chickens administered Ruchamax at weeks 4-6 is presented in Tables 4.2. The results showed that final weight, daily weight and the feed conversion ratio (FCR) were not different (p>0.05) significantly. The final weight ranged from 1213-1370g. The average feed intake ranged from 107.63-122.19g. The average weight gain between the treatment groups did not differ significantly. The daily weight results ranged from 37.44g-39.69g. The feed conversion ratio between the treatments was not significant (p>0.05). The feed conversion ratio results ranged between 2.75-2.96g respectively.

Table 4.1: Growth Performance of Broiler Chickens Administered Ruchamax (0-3 weeks)

(
Parameters	T1	T2	T3	SEM	P/value	L/S	
Initial							
weight(g)	41.13	41.32	41.09	0.13	0.77	Ns	

Final						
weight(g)	406.67	413.33	403.33	3.64	0.59	Ns
average feed						
intake (g)	47.39	44.04	56.92	8.64	0.86	Ns
average						
weight gain						
(g)	17.41	17.71	17.25	0.17	0.61	Ns
Feed						
conversion						
ratio	2.72	2.52	3.32	0.51	0.84	Ns
Protein						
intake	8.53	7.93	10.24	1.55	0.86	Ns
Energy						
intake	142.17	132.11	170.75	25.91	0.86	Ns
Protein						
efficiency						
ratio	2.66	3.03	2.49	0.59	0.95	Ns
Energy						
efficiency						
ratio	0.16	0.18	0.15	0.04	0.95	Ns
Survival%	95.00	95.00	83.33	3.31	0.28	Ns

ns = not significantly (p>0.05) different: SEM - standard error of mean, T1 = Ruchamax, T2= amprolium, T3= control

Table 4.2: Growth Performance of Broiler Chickens Administered Ruchamax (4-6 weeks)

_						
Parameters	T1	T2	T3	SEM	P/value	L/S
Initial weight (g)	413.33	403.33	406.67	3.64	0.59	Ns
Final weight (g)	1213.33	1270.00	1236.67	15.55	0.37	Ns
average feed intake (g)	107.63	122.19	109.44	7.81	0.76	Ns
average weight gain						
(g)	37.44	40.82	39.69	0.84	0.27	Ns
Feed conversion						
ratio Protein	2.88	2.96	2.75	0.17	0.90	Ns
intake	22.60	25.66	22.98	1.64	0.76	Ns
Energy intake	305.67	347.01	310.82	22.18	0.76	Ns
Protein efficiency						
ratio	1.69	1.69	1.74	0.10	0.98	Ns
Energy efficiency						
ratio	0.13	0.12	0.13	0.01	0.98	Ns
Survival %	100.00	97.78	96.58	0.95	0.38	Ns

 $ns = not \ significantly \ (p>0.05) \ different: \ SEM - standard \ error \ of \ mean, \ T1=Ruchamax, \ T2=Amprolium, \ T3=Control$

4.3 Nutrient Digestibility of Broiler Chickens Administered Ruchamax

The nutrient digestibility result is presented in Table 4.3. The result showed that the crude protein values were not significantly (p>0.05) different. The crude protein values ranges from 65.33-67.94%. The birds in all the treatment groups had similar crude protein values though not significant. The crude fibre results ranged between 24.70-31.48%, while the ether extract values ranges from 65.16-71.96% respectively. The ash content result ranges from 5.10 and 6.25. The Metabolisable energy value was 79.54-81.53 respectively. The birds in the control group (T1) had similar ether extract with the birds in T2 (Ruchamax.)

4.4. Carcass Characteristics of Broiler Chicken Administered Ruchamax.

The results of carcass characteristics of broiler chickens administered Ruchamax is presented in Table 4.4. The results showed that the live weight, slaughtered weight, dressing percentage were not significant (p>0.05). The values for the live weight ranges from 1650-1899g. However, birds administered Ruchamax (T1) had the highest live weight while the birds in the control group (T3) had the lowest value. The dressed weight result ranged from 1550-1775. The birds in T2 (amprolium) and T3 (control) had similar values (p>0.05). The slaughtered weight results ranged between 1625 and 1802 respectively. The eviscerated weight ranged between 1350 and 1550.

The result of the cut parts such as drumstick and back were not significantly influenced (p> 0.05). The result of drumstick % ranged between 10.92-11.11%. Birds on T2 (Amprolium) and T3 (control) had similar values (p>0.05). The back % results ranged from 11.01-13.02%. Birds on T1 (Ruchamax) and T2 (Amprolium) had similar (p>0.05) back % values. The results of the internal organs such as liver and lungs were also not influenced (p>0.05) significantly. The liver % ranged between 1.84-2.47%.

Table 4.3: Nutrient Digestibility in Broiler Chickens Administered Ruchamax

Parameters	T1	T2	Т3	SEM	P.value	LOS
Nfe	79.54	78.30	81.53	4.30	0.96	Ns
Crude						
Protein	66.97	65.33	67.54	6.81	0.99	Ns
Crude						
Fibre	26.14	24.70	31.48	13.04	0.98	Ns
Ether	70.44	65.12	71.96	4.50		
extract					0.85	Ns

ns = not significantly (p>0.05) different: SEM - standard error of mean

T1 = Ruchamax, T2 = Amprolium, T3 = Control

Table 4.4: Carcass Characteristics of Broiler Chickens Administered Ruchamax

Parameters Parameters	T ₁	T ₂	T ₃	SEM	P.value	LOS
LIVEWEIGHT	1899.25	1697.65	1650.10	58.70	0.18	NS
SLAUGHTER	1077.20	10) / .00	1020.10	20.70	0.10	110
WEGHT	1802.50	1650.00	1625.00	54.68	0.45	NS
DRESSED WEIGHT EVISCERATED	1775.00	1550.00	1550.00	66.77	0.35	NS
WEIGHT	1550.00	1350.00	1400.00	55.78	0.39	NS
Expressed as Percent	age of Dre	ssed weigl	ht			
HEAD (%)	2.66	2.72	2.84	0.04	0.23	NS
NECK (%)	6.36	5.95	6.03	0.16	0.67	NS
BACK (%)	11.92	11.01	13.02	0.42	0.11	NS
BREAST (%)	23.29	20.68	20.01	1.22	0.62	NS
THIGH (%)	12.59	13.60	13.86	0.32	0.28	NS
DRUM STICK (%)	10.92	11.11	11.07	0.18	0.94	NS
SHANK (%)	4.64	5.06	4.84	0.21	0.79	NS
WING (%)	11.51	11.30	11.83	0.22	0.72	NS
Organs Expressed as	Percentag	e of Live	weight			
HEART (%)	0.36	0.36	0.33	0.01	0.44	NS
LIVER (%)	2.47	2.34	1.84	0.26	0.70	NS
SPLEEN (%)	0.06	0.06	0.04	0.01	0.58	NS
GIZZARD (%)	1.69	2.21	2.35	0.15	0.15	NS
INTESTINE (%)	5.07	6.83	7.23	0.57	0.31	NS
GALL BLADDER						
(%)	0.10	0.09	0.10	0.00	0.88	NS
LUNG (%)	0.54	0.73	0.49	0.05	0.09	Ns
TRACHEA (%)	0.10	0.17	0.10	0.02	0.41	NS
CROP (%)	3.90	5.75	6.85	0.70	0.24	NS
PROVENTRICULUS	0.45	0.40	0.50	0.05	0.45	NG
(%)	0.47	0.48	0.52	0.02	0.67	NS

ns = not significantly (P>0.05) different; SEM = standard error of mean, T1 = Ruchamax, T2= Amprolium, T3= Control

The result of lung % ranged between 0.49 and 0.73 %. The birds in T2 (Amprolium) had the highest value while birds in T3 (control) had the lowest value. The heart % ranged between 0.33 and 0.36%. The birds in T1 (Ruchamax) and T2 (Amprolium) also had similar values (p> 0.05). The results of the spleen% ranged between 0.04 and 0.06%. The birds in T1 (Ruchamax) and T2 (amprolium) had similar values (p>0.05), the cut parts such as the head%, neck % shank %, breast %, thigh %, wing %, crop % proventriculous %, trachea % and internal organ such as gizzard were not significantly (p>0.05) different.

4.5 Haematology and Serum Biochemistry of Broiler Chickens Administered Ruchamax

The results of the haematology and serum biochemistry of broiler chickens administered Ruchamax is presented in figure 1.0. and 2.0 The result showed significant (p>0.05) differences in the haemoglobin, red blood cell (RBC), white blood cell (WBC), the mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) while there was no significant (p<0.05) difference in the platelet and parked cell volume (PCV) respectively.

The result showed significant influence (p>0.05) in the albumin, while no significant effect was observed in the total protein, globulin, aspartate amino transferase, alanine amino transferase, and alanine phosphatase between the treatment groups respectively. The trend of the significant differences observed in the heamatological indices were in a descending order across the weeks.

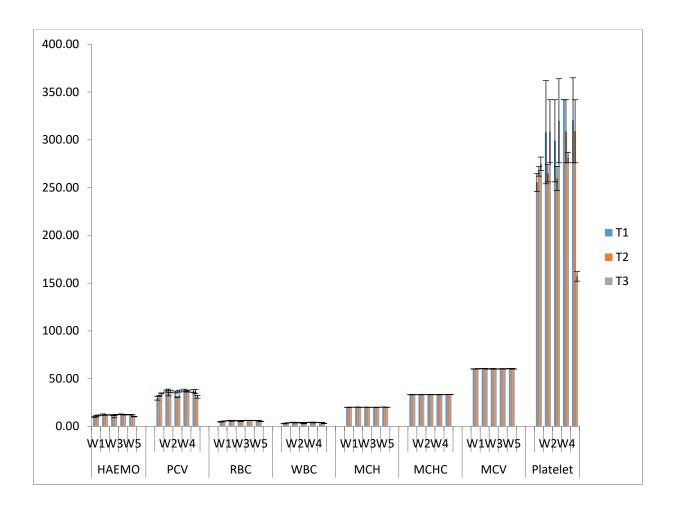


Figure 4.5: heamatology of broilers administered Ruchamax at different weeks

Haemoglobin W1, Week one T1= Ruchamax

Parked cell volume W2, Week two T2= Amprolium

Red blood cells W3, Week three T3= Control

White blood cells W4, Week four

Mean cupurscular haemoglobin W5, Week five

Mean cupurscular haemoglobin concentration

Mean cupurscular volume

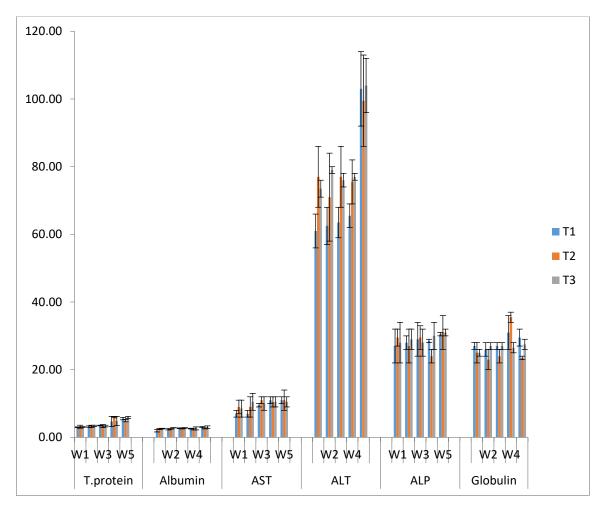


Figure 4.5.1: Serum biochemistry of broilers administered Ruchamax at different weeks

Aspertate amino transferase	T1= Ruchamax	W1= week one
Alanine amino transferase	T2= Amprolium	W2= week two
Alanine phosphatase	T3= Control	W3= week three
		W4= week four
		W5= week five

Table 4.6: Weekly Prevalence of Coccidia Organisms Among Broilers Administered Ruchamax

Treatment	Week2	Week3	Week4	Week5	Week6
T1	+	+	+	_	_
T2	+	+	+	+	_
T3 Chi	+	++	+++	+++	+++
square P.value	2.00 0.37	1.14 0.57	3.44 0.18	4.60 0.1	5.78 0.02

_ = negative; + = positive T1= Ruchamax, T2=

Amprolium, T3= Control

Table 4.6.1: Weekly percentage of broiler chickens found with coccidia specie

Weeks	T1	T2	T3	
2	25	25	25	
3	25	25	50	
4	25	25	75	
5	0.00	25	75	
6	0.00	0.00	75	

Four (4) animals examined per treatment

4.6. Weekly Prevalence and percentages of Coccidia Species among Broiler Chickens Administered Ruchamax

The result of the weekly prevalence and percentages of coccidia among broiler chickens administered Ruchamax is presented in tables 4.6 and 4.6.1, the result showed that Ruchamax was effective against coccidiosis due to reduction in the level of infection along the weeks. The level of infection and percentages among the treatments was similar in week one however, the level of infection and percentage of coccidia organisms increased in the T3 (control) at weeks four, five and six as a result of the fact that it was administered no treatment, while it was totally absent at week six in T1 (Ruchamax) and T2 (amprolium) respectively. The positive sign indicate presence of infection while the negative sign indicate no infection. The single plus sign shows 25 percent presence of coccidia infection, while the double plus sign shows 50 percent presence of infection, the three plus signs indicate 75 percent presence of coccidial infection respectively.

CHAPTER FIVE

DISCUSSION

The growth performance of broiler chickens administered Ruchamax and Amprolium indicated that the treatments had no significant effect (p<0.05) on final weight, daily weight, average feed intake and feed conversion ratio (FCR). Due to the scarcity of literature on the study of Ruchamax® on poultry, the findings of the present research is being compared to the constituents of Ruchamax®. The findings of this study is in line with the study of Allen and Fetterer, (2002), who reported that a combination of A. annua and C. longa did not significantly influence growth performance but indicated anticoccidial efficiency in broilers challenged with a mixture of E. acervulina and E. maxima. Additionally, curcumin was reported to improve coccidiosis resistance as demonstrated by increase in body weight and reduced oocyst shedding and gut lesions. Consistently, curcumin elevated host humoral immunity to *Eimeria* species and reduced gut destruction in poultry. Furthermore, Kim et al., (2013) reported that the supplementation of active ingredients of garlic (propyl thiosulphinate oxide and propyl thiosulphinate) had decreased Feacal oocysts excretion and greater antibody response against *Eimeria acervulina* in broiler chickens, he found that garlic metabolites enhance chickens' growth performances but not to a significant effect and reduce the oocyst output in chickens challenged with E. acervulina, due to a direct cytotoxic effect on the coccidian sporozoites. The findings of this study is not in line with the assessment by Ola-fadunsin, (2013) who reported significant differences in body weight gain, production performance and the anti-coccidial effect of *Moringa oleifera* acetone extract on broiler chicken naturally infected with several Eimeria species.

Significant (p<0.05) difference was observed in haemoglobin, red blood cell (RBC), white blood cell (WBC), the mean corpuscular volume (MCV), mean corpuscular haemoglobin

(MCH) and mean corpuscular haemoglobin concentration (MCHC) between the treatments while no significant (p<0.05) difference was observed in the platelet and parked cell volume (PCV). The result of this research agrees with the result of Ola-fadunsin, (2013), who observed improvement in the haematological indices of the infected birds treated with extracts of *Moringa oleifera*, garlic and combination of *Azadirachta indica* and *Khaya senegalensis* respectively. The significant differences observed in the haematological parameters may be due to the ability of Ruchamax to elevate them during fight against coccidiosis.

The nutrient digestibility of broiler chickens administered Ruchamax indicated no improvement in dry matter, crude protein, ash content and ether extract. The findings of this study is different from the findings of Obi et al., (2019) who was the first to report that A. annua extracts have an anticoccidial activity against E. tenella, as deduced from the herb's impact on weight gain, digestibility, improved feed conversion and reduced lesion score. Barelli et al., (2018) also reported Ginger to enhance the growth performance and digestibility in broiler chickens and effective in treating and controlling coccidia infection. The carcass parameters between the three treatments was not significantly (p>0.05) influenced. There were no significant differences in live weight, slaughtered weight, eviscerated weight and dressed weight among the treatment. The cut parts and the internal organs such as drumsticks, back, liver and lungs were not significantly influenced. The study agrees with the study of Yang et al., (2019) who reported no significant ((p>0.05) differences in body weight and carcass % following administration of Biden pilosa in chickens infected with E. tenella. The study also reported that the plant exhibited anticoccidial activity and reduced fecal oocyst count. However, the finding of this study does not agree with findings of Biu *et al.*, (2006) who reported an improvement in the carcass yield of birds following administration of 800 mg/kg *Azadirachta indica* extract and 100% survival rates and cessation of oocyst production.

The serum albumin between the treatments was significantly (p<0.05) influenced. While no significant differences was observed in total protein, globulin, aspartate amino transferase, alanine amino transferase and alanine phosphatase. The findings of this study is in line with the findings of Dar et al., (2014), who reported that garlic administration increases the values of serum albumin, globulin and total proteins due to its anti-inflammatory and immunomodulatory action that repair the organ lesions induced by *Eimeria* species. Ruchamax was effective against coccidiosis due to reduction in the level of infection along the weeks. The level of infection and percentages among the treatments was similar in week one however, the level of infection and percentage of coccidia organisms increased in T3 (control) at weeks four, five and six as a result of the fact that it was administered no treatment, while the level of infection was totally reduced in T1 (Ruchamax) and T2 (amprolium). The result indicates that Ruchamax® was effective against coccidiosis as compared to the anticoccidial drug amprolium, this might be due to the fact that Ruchamax® contains natural herbal plants such as ginger, garlic, turmeric, and neem which has been proven by researchers to contain high anticoccidial effect, for instance Ali et al., (2019) reported that Garlic and Ginger (Allium sativum and Zingibar officianale) significantly reduced oocyst count in broiler chickens. In addition, Elkhtam et al., (2014) also in his study indicated the anticoccidial effect of garlic and turmeric in broiler chickens experimentally challenged with coccidian oocysts, this may be due to the fact that this plants contains antioxidant, antiparasitic, antibacterial and anti-inflammatory properties. He stated according to his findings that herbal powders induced anticoccidial effect which was concentration dependent and increased by increasing the concentration of the tested powders.

CHAPTER SIX

6.1. Conclusion

It was concluded that supplementation with Ruchamax powder exhibits anticoccidial activity which was comparable with that of the anticoccidial drug and evidenced by prevention. The study also indicate the safety and efficacy of Ruchamax in control, treatment and prevention of coccidiosis. Farmers can safely administer Ruchamax to broiler birds both as prophylaxis and treatment.

6.2. Recommendation

More research should be conducted on the anticoccidial effect of Ruchamax to ascertain the findings of the present research, Ruchamax should be administered at higher inclusion level to determine it toxicity effect. Additionally, future studies on alternative control approaches should be focused on incorporation of already proven alternatives into an effective control program so that farmers could control coccidiosis in an effective way with minimal use of drugs.

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