



Effect Addition of the Extract *Nigella sativa* on the Histological and Physiological Changes of the Domestic Chicken Experimental Infected with *Eimeria maxima*

Sukayna Jabbar Mushattat¹ and Jabbar Abadi Alaridi²

¹Kufa University, faculty of science/Iraq

²Kufa University, faculty of Education for Girls/Iraq

Abstract

The results of the current study showed the effect of the black bean powder *Nigella sativa* on some of the parameters of the domestic chicken with the *Eimeria maxima*. The results showed a significant effect of the *Nigella sativa* extract in RBC, WBC and PCV. The results of the study showed a significant decrease in $P < 0.05$ ratio of heterozygous cells to lymphocytes and the number of eggs in the feces of the chick larvae (0.50 g / mL) compared to positive control. While the results showed a high morale ($P < 0.05$) in the value of the puppet images of the size of the cells and the level of blood clot (Hb) for the treatments fed the alcoholic extract of the black bean (0.50 g / ml) compared with the positive control. In terms of biochemical characteristics, the results showed a high morale ($P < 0.05$) in the level of the blood biochemical characteristics of total protein and cholesterol levels of the black bean extract (0.50 g / mL) compared with positive control, 0.05) at the level of calcium in the positive control group compared to the treatments that were fed a high morale decrease ($P < 0.05$) in the calcium level in the positive control group compared with the treatments fed with the alcoholic extract of the black bean (0.50 g / ml). The study showed a clear effect of the alcoholic extract of black bean seeds *N. sativa* in the restoration of the damaged intestinal tissues due to the experimental infection of parasites *Eimeria maxima*, which was characterized by a lack of damage in the vesicles and lack of lymphocyte infiltration and re-lining the lining of the intestinal epithelium, And lack of bowel bleeding due to injury.

INTRODUCTION

Coccidiosis is widespread throughout the globe and is caused by Oli from the Apicomplexa Division, which belongs to the Eimeriidae family [1]. Most species in poultry belong to the *Eimeria* species, which causes infection in different parts of the intestine for the rapid reproduction of host cells, causing severe damage to the intestinal mucosa, leading to diarrhea, drought and mortality, especially in small ages [2]. The incidence of coccidiosis in poultry is one of the most important problems facing poultry raising in the world, despite the progress achieved in the prevention of these infections and the incidence of coccidiosis is the high proportion of most laboratories for diagnosis of poultry diseases in many countries of the world.

The problem of parasitic infection has been diagnosed since the moment of focusing on the poultry meat industry and most antiperspirants aim to eliminate the causes, but the total elimination of coccidiosis is not possible, there is a strategy to control more than the elimination of parasite. In spite of the old use of preventive and curative drugs that provide good control over the disease is still a problem arising from the continuous breeding of drug resistance of the parasite, which in turn stimulates researchers to find new ways to control this problem [3].

Researchers have worked in many countries of the world since the beginning of the last century in all areas that can lead to the reduction of this disease, some of them stressed on the side of education and some of them focused on the therapeutic side using antacids, and some gave the other side of the importance of immunity to control The disease has been followed by several immunological programs, including the natural exposure of the disease with drugs or extraction of non-living materials of the parasite as a material for immunization or immunization by natural exposure to the parasite, or using the weak prey of the parasite [4].

Plants derived from plants are specialized and have no undesirable effect on humans, other organisms or the environment [5]. *Nigella sativa* is a plant herb grown in many parts of the world in North Africa, Asia and the Arabian Peninsula. The scientific name of the plant is *Nigella sativa*, which is a short plant. The plant contains a capsule containing three-dimensional white seeds that quickly turn black when exposed to air. The black bean contains two types of first oil fixed oil and may have little effect and second oil pilot

is attributed to the drug effect. Volatile oils are a 1.5% volatile volatile substance with aromatic aroma and pale yellow color. The most important compounds are thymoquinone, paracimina and dihydroquinin[5]. Due to the lack of studies on the effect of plant extracts on the infection of chicken with coccidiosis and on the immune response against the kidneys, the present study aimed at studying the effect of the black bean extract *Nigella sativa* on the experimental infection *E. maxima* parasites and the response of the immune system to chickens against infection

MATERIALS AND METHODS

The experiment was incubated in semi-closed rooms of 4 x 4 x for the period from 1/9/2017 to 1/12/2017. The hall was sterilized with 40% formalin and potassium permanganate by 2/1. 5-liter inverted and 38 cm plastic feed containers which were used during the first week of the experiment and then replaced with 40 cm plastic pliers and 45 cm diameter suspended cylindrical containers. The hall was also equipped with a controlled lighting source per week according to the need for airing, The ground was then sprinkled with a 5 cm sawdust. The total number of broiler chickens under study 80 of broiler chickens, after the breeding of domestic chickens from the age of one day and give the program full vaccine (except anti coccidiosis) obtained from the veterinary hospital and the breeding of poultry and until the age of 20 days was conducted The following experiments:

G1- 20 domestic chickens were given a normal diet without developing an injury after 20 days (negative control), G2- 20 chickens were given a normal diet with the development of coccidiosis after the age of 20 days (positive control), G3-20 Fowl I was given a regular meal with black seed plant extract *Nigella sativa* at a concentration of 0.25%, G4-20 Fowl I was given a regular meal with black seed plant extract *Nigella sativa* at a concentration of 0.50%.

After reaching the age of 20 days, the infection was developed by cutting 10,000 live egg Oocyst with a plastic pipette by mouth and studying the physico-chemical and immunological characteristics before and after the experiment 7 days after the experiment with 14 days.

The eggs were collected from the intestines, which were infected with the fungus. The cases were taken to the main veterinary hospital in Najaf with the help of specialized veterinarians. By collecting the lining of the intestine with the contents of the two

arteries, the isolated contents were mixed with potassium dichromate 2.5% well and then sprayed the suspension with a small holes to remove the fibers and suspended materials, and then put the suspension in a clean and sterile glass flask and covered his mouth with aluminum foil and holes in small holes to allow access to the air and placed in a water bath rocking at 28 m 24-48 hours and complete the volume with a solution of potassium dichromate from time to time to dry the suspension, and to ensure the maturity of bags of eggs was taken drop of the suspension and put on Placed on a clean glass slide and covered with the slide cover and examined under the optical microscope with an X40 force. It was observed that the sagging egg bags had 4 spore bags and each bag containing two baguins [6].

Histopathological study, the samples were taken from the intestines and kept in a 10% formalin solution for histological examination to show the histological changes caused by injury according to the method [20].

For the preparation of extract of the black bean *Nigella sativa*, applied 20 g of black bean powder in extraction extract to extract the extracted materials sequentially using the Soxhlat extractor and 200 ml of organic solvent Hexane and for 24 m for dry matter, then dissolve each gram of this substance in 10 ml of distilled water to obtain the original solution. Stock solution Concentration (0.1) g / ml This process was repeated many times in order to obtain the required quantities (0.25%)g/ml, (0.50%)g/ml.

Statistical Analysis

Analysis was conducted using the method Laest Significant Difference L.S.D. of the experiment and the level of probability $P < 0.05$ to find a moral differences between the rates of measurements used in this experiment.

RESULTS

Table (1): Effect of the studied coefficients in H / L, OpG for different ages (mean + standard error).

Groups	before infection		After infection 7 day		After infection 14 day	
	H/L	OpG	H/L	OpG	H/L	OpG
G1(negative control)	0.41 ±0.04	0	0.40 ±0.04	0	0.41 ±0.02	0
G2(positive control)	0.41 ±0.02	0	0.60 ±0.05	37740 ±432	0.89 ±0.04	28632 ±879
G3 (<i>Nigella sativa</i> (0.25%)g/ml)	0.40 ±0.06	0	0.61 ±0.01	1122 ±786	0.55 ±0.03	930 ±110
G4 (<i>Nigella sativa</i> (0.50%)g/ml)	0.42 ±0.03	0	0.60 ±0.0	4067 ±765	0.51 ±0.02	466 ±100
L.S.D $P < 0.05$	0.236		0.132	93.63	0.155	83.621

Table (2): Effect of studied treatments on PCV, RBC, WBC, Hb in serum for different ages (mean + standard e

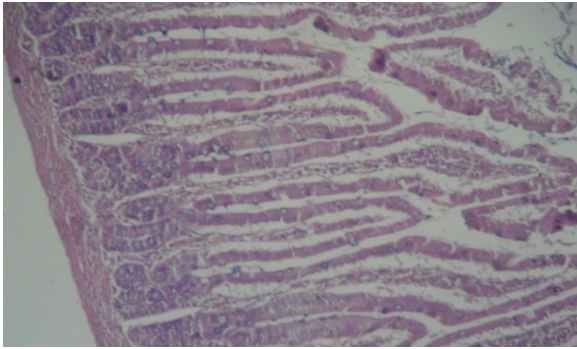
Groups	before infection				After infection 7 day				After infection 14 day			
	PCV	Hb	RBC	WBC	PCV	Hb	RBC	WBC	PCV	Hb	RBC	WBC
G1	31.0 ±0.0	10.0±0.00	2.85±78	100±25	31.75±0.1	10.50±0.21	2.86±54	104±780	31.8±0.8	10.5±0.10	2.87±74	110±98
G2	30.5±0.3	10.0±0.33	2.85±98	101±59	25.6±0.2	8.2 ±0.11	2.22±38	114±659	21.3 ±0.2	7.1 ±0.09	2.21±67	116±92
G3(0.25%)g/ml	30.5±0.2	10.1±0.15	2.84±67	100±98	30.0±0.1	10.0±0.14	2.80±86	111±987	29.0±0.5	9.0 ±0.18	2.81±72	112±86
G4(0.50%)g/ml	30.4±0.1	10.0±0.16	2.85±87	101±56	30.2±0.0	10.0±0.06	2.82±48	113±769	30.0±0.5	10.0±0.27	2.84±94	111±71
L.S.D. $P < 0.05$	1.306	1.355	1.639	1.009	1.209	1.113	1.263	2.337	2.341	1.683	1.506	2.317

PCV = %, RBC = × 106 / micron, WBC = × 103 / micron, Hb = gm / dl

Table (3): Effect of studied treatments on the level of cholesterol, glucose and total protein and serum calcium for different ages (mean + standard error)

Groups	before infection				After infection 7 day				After infection 14 day			
	Glucose	cholesterol	total protein	Ca	Glucose	cholesterol	total protein	Ca	Glucose	cholesterol	total protein	Ca
G1	198.4±0.5	100.3±3.10	4.1±0.09	1.4±0.01	199.0±5.6	105.5±4.0	4.8±0.16	1.7±0.03	200.5±2.5	109.9±4.1	4.9±0.17	2.0±0.4
G2	198.5± 8.3	100.0±2.7	4.1±0.12	1.4±0.03	199.1±6.4	067.2±1.5	3.9±0.31	1.0 ±0.01	199.6±3.7	57.5 ±1.1	2.5±0.08	0.9±0.05
G3(0.25%)g/ml	197.5±9.4	100.0±2.8	4.2±0.32	1.4±0.00	198.2±11.8	99.3 ±1.7	4.0±0.31	1.3 ±0.02	198.4±1.3	100.2±3.2	3.9±0.21	1.8±0.3
G4(0.50%)g/ml	197.3±8.7	100.2±3.0	4.2±0.22	1.4±0.03	197.5±3.7	100.0±3.0	4.1±0.11	1.2±0.02	198.5±3.2	100.2±2.9	4.0±0.05	1.2 ±0.2
L.S.D. $P < 0.05$	1.007	1.632	1.320	0.00	1.639	2.637	0.327	1.392	1.209	3.625	1.661	1.053

Glucose = mg / 100 ml, cholesterol = mg / 100 ml, total protein = mg / ml, calcium = ml / l



Image(1) : shows the normal tissue structure in the intestines in the control group G1.

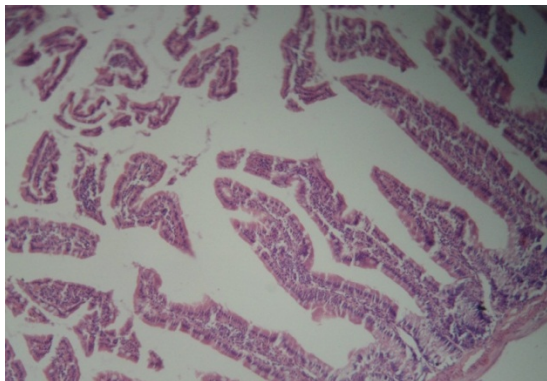


Image (2): The reduced damage to intestinal tissues caused by infection with *E. maxima* in the treated group G4 was shown to be with the *Nigella sativa* extract.

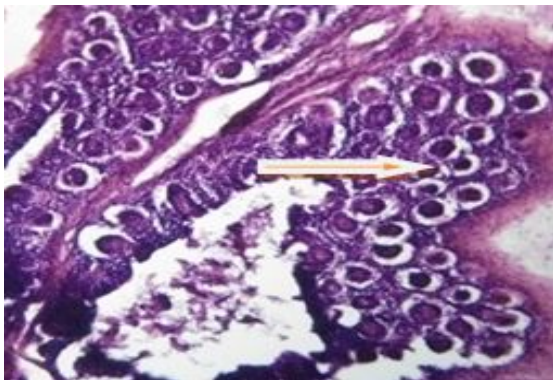
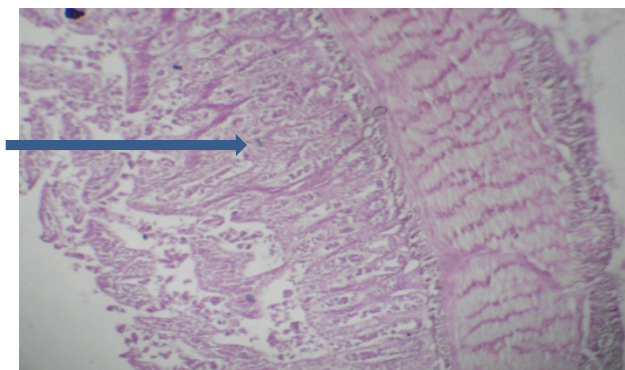
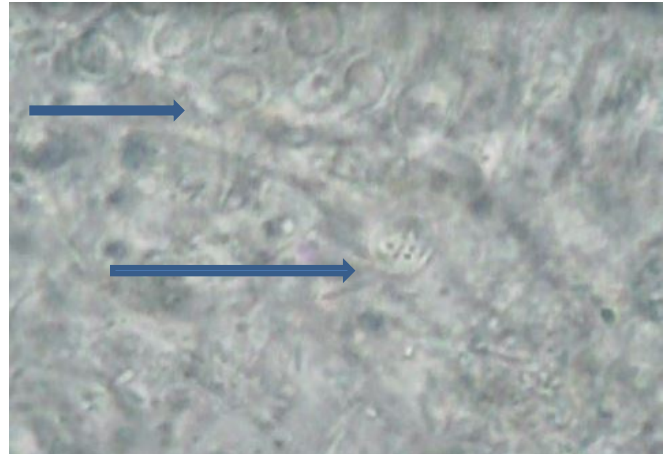


Image (3): The damage to intestinal tissues caused by infection with *E. maxima* in the group G2 was shown to heavy injury and Gametocyte.



Image(4): The reduced damage to intestinal tissues caused by infection with *E. maxima* in the treated group G3 was shown to be with the *Nigella sativa* extract.



Image(5): Gametocyte in the intestinal tissues caused by infection with *E. maxima* in G2.

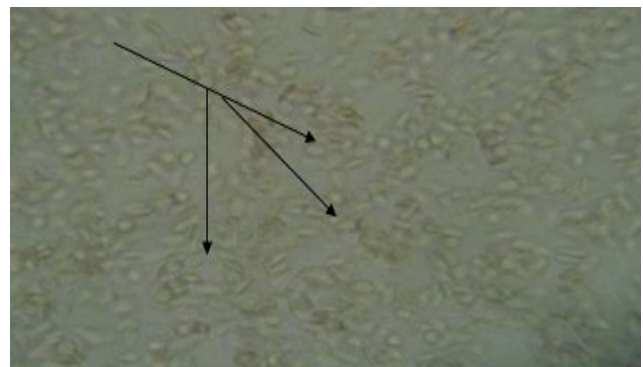


Image (6): showing a large number of Oocyst in a swab of chicken feces in G2 shows the severity of the injury.

DISCUSSION

The dose of *E. maxima* in the infected groups experimented with the emergence of signs of infection six days after the time of infusion, which resulted in the lack of feed intake and the collection of sick birds with each other and the wings drooping and feathers. These signs were due to the parasite passing through the stage of the infiltration phase in which the parasite penetrates the superficial layer of the eyes. This resulted in damage to the capillary blood vessels and bleeding. There was a difference in clinical signs between the treated groups with the *Nigella sativa* extract and control groups. The most powerful in the positive control group G2 being a group that dealt with the regular diet of feed without any additives, The groups treated with *Nigella sativa* G3 and G4 showed fewer clinical signs compared to the positive control group G2, with the lowest clinical markers in the treatment group with the black bean extract at 0.50% concentration. This is due to the role played by the extract in reducing the severity of the lesion with the experimental infection *E. maxima* by restoring the growth of damaged and damaged bowel lining and stimulating the defense cells in the body as well as the work of the black bean extract as antioxidants and thus reduce the effects of *E. maxima* [7].

The prevalence of *E. maxima* eggs in the G2 compared to the other groups is evidence of the severity of the injury. The differences in the preparation of the eggs are considered to be one of the strongest indicators on which many researchers rely to assess the severity of the infection and the treatments used to control coccidiosis. The reason for the gradual decrease in the number of eggs exposed to feces for different weeks of readings may be due to the presence of these chicks on the black bean extract *Nigella sativa*, which stimulated the gut and epithelial

lining of the intestines to stimulate the secretion of immune proteins[8].

Lymphocytes are considered to be the most important white blood cells, as their numbers change. As a result, these two cells are thought to have the ability to eat or resist infectious or alien substances. The increase in heterozygous cells and the reduction of lymphocytes in the blood of susceptible birds ,the reduction of the H / L ratio in the treated groups with the addition of the black bean extract *Nigella sativa* may be due to the containment of chick larvae in these groups on appropriate amounts of vitamins. These results are consistent with[9] On the vitamin Nat breeding to the diets of broiler meat concentrations 40.70, 100 and 400 mg / kg feed led to increased lymphatic cells and reduce hetero cells and this is in line with [9], who confirmed that the use of vitamins perhaps the reason for the low proportion of heterozygous cells to lymphocytes is to increase the secretion of adreno-cortical hormone Tropic Hormone, which is excreted from the gland. the gradual reduction in H / L in the G3 and G4 extracts may be due to the rise of lymphocytes and this is consistent with [10] Which indicated that the lymphocytes The T-helper is increased in proportion to the T-suppresser lymphocytes due to increased immune response Which is developing with age, and the decrease in this percentage indicates the low stress of birds as a result of low body temperature as the stress intensity is linked to body temperature. The rise in H / L in G2 is due to the fact that these parasites cause This is consistent with[11], which observed an increase in the number of white blood cells of the chick three weeks after being infected with a different dose of parasite eggs. Hetrophils increased their percentage Of the first week after injury, while the superiority of m The negative control group G1 on all experimental parameters in the H / L ratio is due to the fact that the treatment is not infected. The positive control group G2 recorded the highest increase in H / L ratio Being exposed to a large number of antigens for the first time, resulting in increased heterozygous cells being represented as the first line of defense and being non-treated with black seed seed extract *Nigella sativa*[11].

This study showed that the parasite has an important effect on PCV in the affected chicks, relying on the phenomenon of reduced PCV is one of the important pathological effects due to infection with this parasite that commensurate with the severity of the infection. And the cause of this phenomenon is due to several factors, including bleeding and coinciding with the incidence of parasitic infection with the secondary effects associated with this infection in the intestine of the lack of feed intake and lack of utilization of food as a result of changes in digestion and absorption of those substances[8].

In the groups treated with black bean extract, this improvement may be due to the percentage of size of the blood cells compressed in the experimental treatments to the use of plant food additives and vitamins and antioxidants, especially the black bean extract *Nigella sativa* at 0.50%, which plays an important role in promoting the distribution Oxygen to tissues or perhaps this improvement is due to increased blood volume as a reflection of the increased need for oxygen by cells [9]reported that adding plant extracts to 100- 150 mg Kg / feed had an effect A significant increase in the volume of compacted blood cells, .aud cause low blood cells stacked the second positive control treatment of G2 (infected without treatment) To anemia in the affected chicks[14] . The stability of hemochlobin levels and the number of red blood cells (RBCs) in the treatment of negative control G1 because they are not infected with parasites. The improvement of hemochlobin levels in them is due to the containment of their diets on extra amounts of vitamins found in the extract of black seed seeds *Nigella sativa* which increases the readiness of iron digestion and absorption, The researcher, Jaiswal et al.[12], confirmed that the addition of plant-derived vitamins to the diet increases the

digestion efficiency of iron by 43% higher than the basic one. This leads to an increase in the efficiency of the generation of hemoglobin. Thyrate is due to the work of vitamins found in the extract as an antioxidant and is also one of the choking compounds in the iron union with the mucus that works on melting iron And make it available for absorption by absorbed cells [13].

While the improvement in G4 more than others may return to the concentration used 0.50% of the extract, which works to increase absorption and regulate growth in the construction of the cell wall of the tissue, which increases the concentration of hemochlobin indirectly. While the reduction in H2 concentrations in the positive control group G2 was due to anemia in the infected chicks with poor feed consumption and decreased nutrient uptake to compensate for the blood components [14].

The increase in the total number of white blood cells in G2 is more than the rest of the treatment because it was exposed to a large number of bags eggs parasite *E. maxima* for the first time, which led to an increase in the total number of white blood cells, which means a significant increase in the severity of the disease, While the total number of white blood cells in the treated groups with the black seed seed extract *Nigella sativa* was also high but relatively less than in G2. This was due to the effect of the plant extracts of vitamins, antioxidants and volatile oils on the overall growth rate in aggregates of G3, G4 and stimulate the proliferation and cell division, This is due to the natural immune response due to the entry of *E. maxima* eggs bags, which are antigens, which led to an increase in the number of white blood cells that form part of the immune system and thus stimulate the immune response humoral and cellular, while the increase in the total number of white blood cells in totals Control is greater because it is a non-immune group, which resulted in the first exposure of this number of eggs *E. maxima* to increase the total number of white blood cells and this is consistent with the mechanism of the mechanism [14].

The stability of glucose concentrations is consistent with the[15], which showed that vitamin A and E extracts were orally administered to diabetic rats with a dose of 100, 200 and 300 mg / kg bw Physical Homeostasis resulting in reduced glucose compared to untreated and untreated treatments. [15]pointed to the high concentration of plasma glucose for meat-borne parasites, which contain the dietary supplements of vitamins. The reason for this was attributed to the effectiveness of vitamins in improving overall growth and thus improve the digestion of nutrients and absorption of the digestive tract and thus regulate Blood level, and the slight increase in the coefficients may be due to the decomposition of the calcification result Increased secretion of the stimulating hormones of the cyanogenic enzymes.

The levels of cholesterol in the infected groups improved empirically with *E. maxima* with the addition of the black bean seed extract *Nigella sativa* due to improved bird health, reduced intestinal lesion, improved digestion and absorption and stimulated cholesterol synthesis in the liver. This is consistent with Which reported that the vitamins effects in the re-growth of damaged tissues, which may be due to metabolic changes in the liver, and the low cholesterol levels in the positive control group G2 is due to failure to synthesize the liver because of damage caused by the severity of infection *E. maxima* parasites that caused necrosis And damage to cells Which has negatively affected the liver's normal functioning [16].

The improvement in protein levels in the *Nigella sativa* extract, especially the extract group, at a concentration of 0.50% was consistent with [16], which indicated that the addition of the protein extract containing vitamins 100 and 150 mg / kg feed Improved overall protein levels in blood plasma due to the effects of metabolic vitamins and their ability to stimulate the production of immune proteins ,the total protein level in the chicken serum is

directly proportional to the level of antibodies and body weight, which is a direct reflection of changes in metabolic rate and metabolic level in plasma blood. And proteins, especially albumin, the transfer of carbohydrates, vitamins, fatty acids and some hormones such as thyroxine important in the process of metabolic representation, and thus observed improvement in total protein levels in groups infected experimentally *E. maxima* parasites and treatment with the addition of plant extract *Nigella sativa* [16].

Protein levels may not be affected in the early days of experimental infection in untreated groups with increased vitamins due to the degradation of the calcium and the protein found in the muscles negatively affected by the high corticosteron hormone associated with stress and disease causing anorexia. Therefore, the liver increases the secretion of this hormone for the purpose of glucose synthesis which the bird needs as an energy source [16].

The results of the current study showed the effect of calcium level in the serum, as it showed a significant decrease in G2 compared to G1, the calcium deficiency can be explained that parasite after a period of injury and the vesicle around the parasite inside the cells becomes parasite able to Calcium is withdrawn from the host cells to the inner perimeter of the surrounding vesicle because the tubulo vesicular network of the parasite vesicle is highly correlated with calcium [17].

The process of host cell invasion is accompanied by the secretion of proteins by the parasite that are accompanied by an increase in intracellular calcium, as well as the parasite exit of the destroyed cells. This is induced by the low concentration of extracellular potassium ions along with the high concentration of intracellular calcium. Calcium is also necessary for the parasitic movement of the parasite [17]. Fat oxidation affects the membrane's viability and the change of membrane-associated enzymes and ion channels, leading to ion transport disruption and increased calcium. MDA binds membrane elements and affects ion exchange in cell membranes, thus altering the ionicity and efficiency Enzymes [18]. The improvement of the calcium level in the treated groups with the addition of the extract *Nigella sativa* is due to the work of plant vitamins and volatile oils found in the extract to reduce the harmful effects resulting from the pyrooxidosis associated with parasitic infection by acting as antioxidants, and by acting as a strong antioxidant and cellular catalyst To the immune system, as well as to enter the cellular membranes and thus protect the body cells from damage caused by the oxidation caused by parasitic infection, through the destruction of hydrogen peroxide and hydroperoxides in the plasma and Saito Block cells and prevent their movement around the cell and its interaction with its membranes and enzymes containing sulfahydride groups Causing significant damage to the cell and its cellular processes, as well as inhibiting the release of creatinine kinase from cells into plasma and thus reducing the effects of calcium entering the cells [19].

The results of the histological study showed that the treatment of the black bean extract of *Nigella sativa* with a concentration of 0.50% had a high effect on the repair of damaged cells due to the experimental infection. The histological images showed the recurrence of the intestinal flora, ie, the repair of the tissue, compared to the positive control group, Significantly in the lining of the intestines and zebras as a result of penetration by the first generation and the second of the parasites of the *E. maxima* parasite in addition to the infiltration of inflammatory cells within the intestinal epithelium [20], This may be due to the active compounds found in the black bean extract added to the drinking water throughout the breeding period, including Thymoquinone and its derivatives Nigel lone and Sterols. These compounds have an important role in stimulating the immune system, high blood cell count and re-tissue in the affected intestines.

REFERENCE

- [1] Cynthia, M. K. & Scott, L. (2000). The Merck Veterinary Manual. Coccidiosis .8th ed., Amerck and Aventis Company, USA.
- [2] Levine, N. D. (1985). Apicomplexa. In: Lee, J. J.; Hutner, S. H. and boves, E. C. (eds). An illustrated guide to the protozoa. Society of Protozoologists, USA. pp. 349-352.
- [3] Williams, R. B. & Catchpol, J. (2000). Anew protocol for challenge test to assess the efficacy of live anticoccidial vaccine. Vaccine. 18:1178-1185.
- [4] Anderson, G. & Jorgensen, W. K. (2004). Live vaccines for their species of *Eimeria* RIRDC publication No. 03/143 RIRDC project No. DAQ-259.
- [5] Trout, J. M., and Lillehoj, H. S. (1995). *Eimeria acervulina* infection:evidence for the involvement of CD8_T lymphocytes in sporozoite transport and host protection. Poult. Sci.,74: 1117–1125.
- [6] Barwick, W. M.; Stevenson, G. T.; Johnson, R. V.; Casores, D. R. and Hymas, T. A. (1970). Coccidiosis evaluation of techniques for battery testing of field collected *Eimeria* oocysts .Exp. Parasitol.28:37-41.
- [7] Jimolu, G. G. (2004). Studies on poultry coccidiosis in Tiyo Wereda, Arsi Zone, Oromia Regional State. MS.C. Thesis. College of Veterinary Medicine, Addis ababa university. :9.
- [8] Lee, H. A. ; Hong, S.; Chung, X. and Kim, O. (2011). Sensitive and specific identification by polymerase chain reaction of *Eimeria tenella* and *Eimeria maxima*,important protozoan pathogens in laboratory.avian Facilitre Lab.Anim.Res;27: 255-258.
- [9] Galal, A.; EL-motaal, A.; Ahmed, A. M. H. and Zaki, T. G. (2008a). Productive performance and immune response of laying hens as affected by dietary propolis supplementation. Internationa Journal of Poult. Sci., 7 : 272-278.
- [10] AL-Quraishy, S. ; Abdel-Baki; A. S. and Dkhil, M. A. (2009). *Eimeria tenella* infection among broiler chicken in Riyadh city, Saudia Arabia.J. King Saud Univ. Sci.;21:191-193.
- [11] Kaingu, F. B.; Kibor, A. C.; Shivairo, R.; Kutima, H. and Waihana, R. (2010). Prevalence to gastrointestinal helminthes and Coccidia in indigenous chicken from different agro climatic zones in Kenya. Afr. Res. ;5:458-462.
- [12] Jaiswal, A. K.; Venugopal, R.; Mucha, J.; Carothers A. M. and Grunberger, D. (1997). Caffeic acid phenethyl ester stimulates human antioxidant response element-mediated expression of the NAD (P) H:Quinone oxidoreductase (NQO1) gene. Cancer Res., 57: 440-446.
- [13] Haro, A.; Lopez-Aliaga, I; Lisbona, F.; Barrionuevo, M.; Alferez, M. J. M. and Campos, M. S. (2000). Beneficial Effect of Polle and/or Propolis on the Metabolism of Iron, Calcium, Phosphorus,and Magnesium in Rats with Nutritional Ferropenic Anemia. J. Agric. Food Chem., 48 : 5715-5722.
- [14] Islam, M. S.;Lucky, N. S. ; Islam,M. r. ;ahad, A. ;Das,B. R. ;rahman, M. m. and siddiu,M. S. i. (2004). Haematological parameters of fayonmi, assil and local chickens reared in SyIhet region in bangladesh.Int.J.Poult.Sci;3: 144-147.
- [16] Mondal, D. K.; Chattopdhay, S.; Batabyal, S.; Bera, A. K. and Bhatteechrya, D. (2011).Plasma biochemical indices at various stages of infection With a field isolate of *Eimeria tenella* in broiler chicken. Vet. World,4: 404-409.
- [17] Coppens, I. and Joiner, K. (2001). Paraste-host cell interaction in toxoplasmosis: new avenues for intervention, expert review in molecular medicine. 56: 36-41.
- [18] Bulut, M.; Selek, S.; Gergerioglu, H.; Savas, H.; Yilmaz, H. ; Yuce, M. and Ekici, G. (2007). Malondialdehyde levels in adult attention-deficit hyperactivity disorder. J Psychiatry Neurosci. ; 32(6): 435–438.
- [19] Çetin, E.; Silici, S.; Çetin, N. and Güçlü, B. K. (2010). Effects of diets containing different concentrations of propolis on hematological and immunological variables in laying hens. Poult. Science., 89:1703-1708.
- [20] Humanson, m. (1967). Anineal tissue techniques , 2 th ed. Freeman, W. H. and Company, San Francisco and London, pp.569.