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Vol. 22(2) May-August 2024

CONTENTS

Impact of *Zea nicaraguensis* introgression on Kernel Trait Variability in maize lines **231**
SENTHILKUMAR V., PRIYA GARKOTI., THOTLA NARESH, MAYANK TIWARI, ANIRUDH T. V. and NARENDRA KUMAR SINGH

Improving *Brassica juncea* performance through hybrid breeding strategies: a focus on combining ability and heterosis analysis **244**
ANU SINGH, USHA PANT, PREETI LOHANI, A. S. JEENA and ANIL KUMAR

Study of Nano Urea application under graded n rates on growth, productivity and nitrogen use efficiency of transplanted rice (*Oryza sativa* L.) **251**
S.K.YADAV , D.K.SINGH, PRATIMA ARYA and YUVRAJ SINGH

Isolation, screening and characterization of Drought tolerant Plant Growth Promoting bacteria from Indian Himalayas **261**
PRIYANKA KHATI, PANKAJ KUMAR MISHRA and LAKSHMI KANT

Impact of Glomalin-Related Soil Proteins on *in vitro* Finger Millet (*Eleusine coracana* (L.) Gaertn.) seed germination **272**
AMIT SINGH RANA, SUGANDHA PANT, ASHOK KUMAR VERMA and ASHUTOSH DUBEY

Rating scale of pedological development in humid moisture regime of guava growing soils in north-east region of Haryana **279**
DHARAM PAL and DINESH

Coating micronized elemental sulphur powder on prilled urea: process and product evaluation **286**
P. O. SURESH, N. R. PATEL, R. JAT, R. A. PANIA, A. K. MISHRA, P. B. VAISHNAV

Multi-year temporal analysis of sheath blight incidence in rice using geostatistical technique **297**
AMIT BIJLWAN, RAJEEV RANJAN, MANENDRA SINGH, RAJ KUMAR SINGH, RAJEEV KUMAR SRIVASTAVA, KRISHNA PRATAP SINGH and RAVINDRA KUMAR SINGH RAJPUT

Efficiency assessment of classifiers for sugarcane area mapping: A machine learning approach with Google Earth Engine **305**
POOJA YADAV, AJEET SINGH NAIN and SHIVANK DEVLIYAL

Calibration and performance evaluation of the APSIM and CERES-Wheat model in the foot hills of Western Himalayas **319**
NEHA PAREEK, A.S. NAIN, P. K. SINGH, HEMANT KUMAR, SHRUTI V. SINGH, MANJARI SINGH,PRIYANKA SWAMIand SANTOSH KUMAR

Population dynamics of major insect pests of sesame and their correlation with meteorological factors **330**
BHUMIKA RAWAT, M. S. KHAN, ASHUTOSH and DEEPIKA JEENGAR

***In-vitro* screening of *Trichoderma* isolates for their antagonistic potential against *Rhizoctonia solani* causing aerial blight of Soybean** **335**
ARUNKUMAR, BHUPESH CHANDRA KABDWAL and ROOPALI SHARMA

Physiological and biochemical responses of okra seed (*Abelmoschus esculentus* L.) to botanicals and containers during storage **350**
SUNIL KUMAR, S. S. JAKHAR,ANIL KUMAR MALIK and AXAY BHUKER

Effect of integrated weed management practices on growth parameters in vegetable pea (*Pisum sativum* L.) **357**
NEELIMA RAWAT, MANOJ RAGHAV, DHIRENDRA SINGH, ALKA VERMA, NAVNEET PAREEK, HITAISHI KURIYALand IMAMUDDIN SHAH

Maximizing Chrysanthemum (*Dendranthema gradiflora*T.) growth and yield: Unveiling the superiority of Black Polythene Mulch **360**
HARSHITA BORA, MAMTA BOHRA and K. C. SINGH

Utilization of ultrasonicated edible coating to prolong shelf life of fresh cut- onion **368**
NEHA RAWAT, SATISH KUMAR SHARMA, ANIL KUMAR, NAVIN CHANDRA SHAHI, ASHUTOSH DUBEY, CHARU BISHT, ARCHANA GANGWAR

Effect of cooperative societies on food security status of cassava farming households in delta state, Nigeria **378**
IZEKOR, O.Band OKOROR O.T.

Strategies for Improving Agricultural practices: A case study of tomato growers from Uttarakhand **388**
TAMANNA JOSHI and ASHUTOSH SINGH

Physico-functional and sensory qualities of instant custard powder incorporated with resistant starch from Grand Naine banana **398**
SRUTHY. P. M., SHARON. C. L., SEEJA THOMACHAN PANJIKKARAN, A. N. JYOTHI, ANEENA E. R.and LAKSHMI P. S.

Development and quality evaluation of rice-based meal replacer with chocolate flavour for adults **404**
ATHIRA RAJ, SUMAN K.T., BEENA A. K., SEEJA THOMACHAN PANJIKKARAN, SHARON C. L., LAKSHMY P. S., DELGI JOSEPH C.and SREELAKSHMI A. S.

Effect of bleaching on optical properties of *dhaincha (Sesbania aculeata)* pulp **411**
SURABHI DAS, ANITA RANI, MANISHA GAHLOT, SAKSHI and NIDHI SISODIA

Evaluation of genetic and non-genetic factors affecting first lactation traits in crossbred cattle **421**
NAYLA FRAZ, B. N. SHAHI, R. S. BARWAL, C. V. SINGH and A. K. GHOSH

Mushroom (*Agaricus bisporus*) waste as a replacement for deoiled rice bran and its impact on immunocompetence against Ranikhet (Newcastle) disease virus in Rhode Island Red Chicken **426**
MANAS ARORA, R. KUMAR, A. TEWARI, A. KUMAR, J. PALOD and B.C MONDAL

Effect of *Aloe vera* leaf extract on pathological lesions of *Escherichia coli* infected broiler chickens **433**
MAMTA KUMARI, RAJENDAR P. GUPTA, DEEPIKA LATHER, PREETI BAGRI, RENU SINGH, SARVAN KUMARand KOMAL

Effect of metronidazole on hematological parameters in Common Carp (*Cyprinus carpio*) **443**
ANIKA SHARMA, MADHU SHARMA, TARANG SHAH and PRASANJIT DHAR

Reproductive and productive performances of Japanese Quails (*Coturnix japonica*) under agro-climatic conditions of Assam **449**
DEBAJIT DEKA, ARFAN ALI, ASHIM KUMAR SAIKIA, MRIDUL DEKA, UTPAL JYOTI SARMA, MANORANJAN NEOG and RANJIT KUMAR SAUD

Performances of Turkey birds under backyard system in agro-climatic condition of Assam **454**
DEBAJIT DEKA, ARFAN ALI, ASHIM KUMAR SAIKIA, MRIDUL DEKA, MANORANJAN NEOG, RANJIT KUMAR SAUD and UTPAL JYOTI SARMA

Nutraceutical supplements for managing pain and inflammation: A special focus on palmitoylethanolamide and astaxanthin **459**
AKHTER RASOOL, DIVYA CHAVAN, PULI VISHNUVARDHAN REDDY, JAN MOHD MUNEEB and IRTIQA MANZOOR

Characterization and use of hydrochars from wheat straw, fruit peels, and sewage sludge: A potential biofuel source **470**
KARAN SATHISH and SHWETA SARASWAT

Battery assisted single wheel weeder for medicinal plants **479**
SANDEEP KUMAR SAROJ and JAYANT SINGH

Chat GPT: Perception of students towards AI tool **486**
ARPITA SHARMA KANDPAL and POOJA GOSWAMI

Effect of *Aloe vera* leaf extract on pathological lesions of *Escherichia coli* infected broiler chickens

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ABSTRACT: The emergence of antibiotic resistance in poultry is common due to the continuous use of antibiotics for controlling major bacterial diseases like *Escherichia coli* infection. So, this study was planned to evaluate the use of plant extract *Aloe vera* as an alternative to fight against *E. coli* infection. The effect of *A. vera* leaf extract supplementation was assessed based on the severity of clinical signs and pathological lesions in *E. coli*-infected broiler chicks. Broiler chicks were supplemented with aqueous *Aloe vera* Leaf (AVL) extracts @ 20 ml per liter of water from day one of age. On the 8th day of age, group A2 and B2 birds were given *E. coli* O78 @ 10^7 CFU/0.5 ml intraperitoneally. The birds were sacrificed after 28 days of *E. coli* inoculation and examined for gross and histopathological lesions. Gross lesions observed were fibrinous pericarditis and perihepatitis, airsacculitis, peritonitis, enteritis, congestion in different visceral organs such as lung, spleen, liver, kidneys and intestines, and atrophy of bursa of Fabricius. Histopathological changes observed were fibrinous pericarditis and myocarditis, fibrinous perihepatitis; congestion, hemorrhages and mild oedema and infiltration of lymphocytes and macrophages in the alveoli and bronchiole. There was depletion of lymphocytes in the bursa of Fabricius. The protection on gross and histopathological lesions of *E. coli* infection due to *A. vera* leaf extract was 14.19% and 17.5%, respectively.

Key words: *Aloe vera, E. coli,* gross lesions, histopathological lesions, poultry

The poultry industry in India is one of the fastest growing agricultural sectors in the country. Commercial broilers are grown throughout the world and broiler chicken is becoming the most affordable, delicious and nutritious protein. Demand of poultry meat is growing globally. However, the birds are affected by many infections that lead to economic loss as well as increased risk of antibiotic resistance. Avian colibacillosis caused by *Escherichia coli* (*E. coli*) is among the important bacterial infections affecting poultry industry. The acute form is characterized by septicemia resulting in death and in its sub-acute form by airsacculitis and fibrinous polyserositis. The majority of economic losses result from mortality, decreased egg production, and condemnations and also costs of vaccination, chemotherapy, and eradication programs (Kabir, 2010). The frequent use of antibiotics is resulting in resistance to pathogenic microorganism, affecting the feed efficiency and growth performance of poultry birds. To find a remedy, we can use herbal preparations as medicines. In ethnoveterinary practice, various herbal and medicinal plants have been used to prevent chicken diseases (Waihenya *et al.,* 2002).

Aloe vera, is another important medicinal plant which is a succulent plant growing by spreading offsets. Its ethno veterinary use dates far back into history as in the Greek, Egyptian and Roman eras (Bassetti and Sala, 2005). While raw leaf juice was traditionally used as laxatives (Wintola *et al*., 2010), and to treat burns and cuts (Pugh *et al*., 2001). Isolates from *Aloe vera* were shown to inhibit microbes like *Escherichia coli, Klebsiella pnemoniae*, *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Staphylococcus aureus, Candida albicans* and *Penicillium* sps**.** (Stuart *et al*., 1997; Devi *et al*., 2012). Clinical studies have revealed several immunomodulatory properties. Currently, it is extensively used in the cosmetic industry for its anti-inflammatory cure of skin disease (Reuter *et al*., 2008), wound and burn healing effects (Jia *et al*., 2008) reported.The extract of *Aloe secundiflora*

is used in the control of fowl typhoid in chickens (Waihenya *et al*., 2002). Johnson *et al*. (2011) found that aqueous and alcoholic extract of *Aloe vera* had strong antibacterial activity *in vitro* against *S. aureus* and *E. coli*. So, the present study was undertaken with the objectives to study the effect of *A. vera* extracts on pathological changes of *E. coli* infection in broiler chicks.

MATERIALS AND METHODS

Guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals, Gov. of India were followed to conduct the experiment. Approval for the study was granted by the Institutional Animal Ethics Committee of the University on Date 13/12/2013/No. VPHE/IAEC/ 3381-3400, Agenda item No. 5."

Healthy and unvaccinated day-old Cobb broiler chicks (180) were procured from a local hatchery and reared under cage system in the departmental animal house under strict hygienic conditions. The chicks were kept in well-ventilated and well-lit rooms, maintained at optimum temperature. All the birds were provided with commercially prepared chicken starter feed and fresh water *ad libitum* throughout the experiment.

Aloe vera leaves were collected from the farm of the College of Agriculture, CCSHAU, Hisar. The leaves were washed and pulp/gel was taken out after removing the outer covering. The pulp was blended for 3 minutes. The blended material was squeezed through muslin cloth and stored at 4°C till further use (Githiori *et al*., 2003). *E. coli* (serotype O78) isolated from natural cases was used @ of 1 x 10^7 CFU of *E .coli*/0.5 ml per bird for producing infection (Jindal *et al.,* 2003).

One hundred day old chicks were divided into groups viz., Group A, containing sixty birds each. The birds of group A were kept as control without any supplementation. The birds of group B were supplemented with aqueous AVL extract @20 ml per liter water. After seven days of age, the birds of both the groups were further divided into two subgroups (Group A into A1 and A2, Group B into B1 and B2) of 25 and 35 birds, respectively. *E. coli*(@ 1 x 10^7 CFU/ 0.5ml) inoculum was injected intraperitoneally to the birds of group A2, and B2 whereas 0.5 ml sterile NSS was injected to groups A1 and B1.

Clinical signs and mortality were noted daily.Viable bacterial cell count of *E. coli* in liver of the infected groups (A2, and B2) was determined at 7, 14, 21 & 28 days post infection under aseptic conditions by plate counting method described by Cruikshank *et al.* (1975).

At the end of the experiment, the birds were sacrificed and thorough post mortem examination was done to note the gross lesions. Tissues were collected for histopathologicalexamination. For this,the formalin fixed tissues were washed in running tap water, dehydrated in graded ethyl alcohol, cleared in benzene and embedded in paraffin wax (melting point 60-62^{0}C). Sections of 3-4 µm thickness were cut using semi-automatic microtome (Yorco, YSI-062) and stained with hematoxylin and eosin (Luna, 1968). The colibacillosis specific gross lesion score (GLS) and histopathological lesion score (HLS) in different experimental groups were calculated for different organs/tissues at scale of 0 to 4 as detailed below:

0 = No lesion; 1 = Mild lesions; 2 = Moderate lesions; 3 = Moderately severe lesions; 4 = Severe lesions

Per cent mean gross and histopathological lesions were calculated with following formula (Witter,1982).
Per cent mean gross/histopathological lesion score (GLS/HLS) of organs = [Mean GLS/HLS of an organ / 4(Maximum GLS/HLS of an organ)] x 100
Overall per cent mean GLS/HLS of organs irrespective of post infection period= [Sum of per cent mean GLS/HLS of an organ at different post infection period / 5(Total number of post infection periods)] x 100

Overall per cent mean GLS/HLS of organs

irrespective of post infection period and organs = [Sum of per cent mean GLS/HLS of organs irrespective of post infection period / 6(Total number of organs)] x 100

Per cent protective effect due to *A. vera* leaves extract supplementation in *E.coli* infected chicks was calculated on the basis of GLS and HLS as per method of Witter (1982) using following formula:
Per cent protective effect due to AVL extract supplementation in *E.coli* infected chickens = [(Overall % mean GLS/HLS in group A2 – Overall % mean GLS/GLS in group B2) / Overall per cent mean GLS/HLS in group A2] x 100

The data for various parameters were subjected to statistical analysis by applying Analysis of Variance (ANOVA) technique using Statistical Package for Social Sciences(SPSS) 17th version and by Z test. Statistically significance between means was tested using Duncan Post hoc comparisons tests at $P<0.05$.

RESULTS AND DISCUSSION

No clinical signs were observed in any of the control groups A1, and B1 throughout the experiment. Clinical signs of inappetence, dullness, depression started to appear at 24 hours post infection in the group A2 (control infected, non-supplemented). Thereafter the chicks showed anorexia, listlessness, ruffled feathers and drooping of head and neck. They also exhibited reluctance to movement and were huddling together near the light source. At 7 DPI some of the infected birds exhibited respiratory distress and mild diarrhoea, and dehydration. The wings and legs were stretching out and the birds were not moving. The severity of clinical signs declined in second week post infection, and considerably reduced in third week post infection. Clinical signs were not observed in fourth week post infection. The chicks of group B2 (infected, AVL extract supplemented) also started showing clinical signs after 24 hours of infection and the severity of the signs was almost equal to that of group A2 i.e. control infected group. However, the chicks appeared did not show any clinical signs from the mid of third week post infection.

The severity of clinical signs of experimental colibacillosis observed in the present study was at peak on 7 DPI. More or less similar clinical signs have also been reported by other workers (Pandey *et al.,* 1998; Satyajit *et al*., 2013; Verma and Swamy, 2013; Kumari and Gupta, 2014a; Abd El-Tawab *et al*., 2015) in natural and experimental cases of colibacillosis. The results of clinical signs indicated that *A. vera* leaves did not show any significant effect on amelioration of clinical signs of colibacillosis.
There was no mortality in non-infected groups A1, and B1. Total mortality observed in group A2 was 25.71% (three on 2 DPI, one on 3 DPI, two on 6 DPI, one on 8 DPI and two on 11 DPI). In group C2, mortality started at 2DPI and continued up to 9 DPI with a total mortality of ten chicks (28.57%). The difference was insignificant.Mean viable bacterial (*E. coli*) cell counts in the liver of the infected groups of broiler chicks are presented in Table 1.

The mean viable cell count in group B2 was lower as compared to group A2 from 7 DPI onwards though the difference was non-significant. These findings suggested that the bacterial load was decreased to some extent due to supplementation of AVL extract. It is reported that *A. vera* leaf extract has antibacterial effect in *in-vitro* conditions (Kumari and Gupta, 2014b), so it is possible that in *in-vivo* conditions also it inhibits the growth of bacteria resulting in lower bacterial count noticed in organs.

Pathological studies

No pathological changes could be observed in chicks from non- infected groups (A1 and B1) at different intervals throughout the experiment.

Group A2 (*E. coli* infected): On 7 DPI, fibrinous pericarditis and perihepatitis was observed (Fig. 1). It resulted in adhesions with abdominal wall as well as with other visceral organs. Congestion was noticed in various visceral organs such as liver, heart, lungs, spleen, proventriculus and kidneys. Lungs revealed thin fibrinous layer over its surface. Spleen was slightly enlarged and pin point necrotic foci were noted. Bursa of Fabricius was atrophied and intestines congested. On 14 DPI, heart revealed

severe pericarditis and liver was covered with thick fibrinous mass. Severe peritonitis was noticed along with adhesions of intestines with abdominal organs. Lungs were congested, and consolidated. Bursa of Fabricius was atrophied. On 21 DPI, the severity of lesions was reduced. On 28 DPI, there were no significant lesions except heart and liver showed mild congestion.

Group B2 (*E. coli* infected + AVL extract): On 7 DPI, fibrinous layer was present on heart and liver (Fig. 2) along with adhesions with abdominal wall. Lungs were congested and covered with fibrinous exudate. Spleen showed enlargement, congestion and few necrotic foci. Intestine revealed mild congestion. Mild atrophy of bursa of Fabricius was observed. The severity of lesions was almost same as in group A2 at 7 DPI. At 14 DPI, the severity of lesions in heart was almost same as in group A2 but hepatic lesions were not as severe as in group A2. On 21 DPI, mild congestion was observed in heart, liver, lungs, spleen and intestine. On 28 DPI, no gross lesions were noticed in any of the organs.

Polyserositis have been reported by other workers (Baliarsingh *et al.,* 1993; Verma and Swamy, 2013; Kumari and Gupta, 2014a) in natural and experimental colibacillosis. It was noted that gross lesions in supplemented infected group B2 were of less intensity at different intervals as compared to group A2. Mild protective effect of AVL extract on gross lesions might be due to its antioxidant, antibacterial and immunomodulatory effect (Kumari *et al.*, 2023). Kumari and Gupta (2014b) notice growth inhibitory effect of *A. vera* extract on *E. coli* although complete antibactericidaleffect was not observed. The cellular immune response in broilers was also observed to be improved by *A. vera* administration. This suggests that the reduction of free radical concentration, the reduced number of bacterial count in organs and the improved cellular immunity might have reduced the severity of lesions to some extent.

Histopathological lesions

Group A2 (*E. coli* infected): On 7 DPI, heart revealed fibrinous pericarditis characterized by presence of large amount of fibrin and infiltration of heterophils and lymphocytes along with myocarditis characterized by congestion, haemorrhages and fragmentation of cardiac muscles, oedema and infiltration of mainly heterophils (Fig.

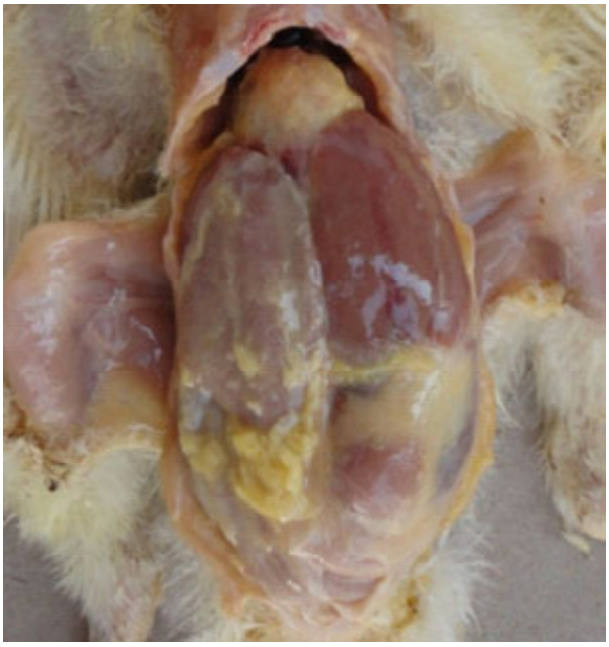

Fig. 1: (Group A2, 7DPI) Thick fibrinous layer/mass on the surface of heart and liver (arrows) resulting in adhesions of visceral organs

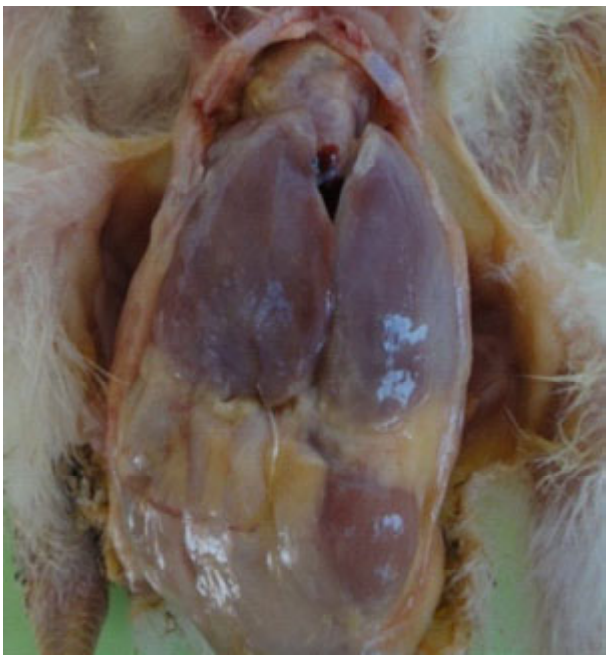

Fig. 2: (Group C2, 7DPI): Moderate fibrinous layer on the surface of heart and liver (arrow)

3). Lungs revealed pleuritis and bronchopneumonia characterized by congestion, perivascular oedema and mononuclear cells infiltration in alveoli and bronchioles. In the spleen depletion of lymphocytes in the white pulp, congestion and focal necrotic areas were noted. Bursa of Fabricius revealed atrophy of bursal follicles, depletion of lymphocytes and increase in inter follicular space due to reticulo-endothelial cells proliferation, infiltration of heterophils and oedema. Intestines revealed mild congestion in the mucosa. Perihepatitis and Proventriculitis was also noticed (Fig. 4).

On 14 DPI, heart revealed severe pericarditis characterized by infiltration of mononuclear cells mainly lymphocytes and macrophages along with accumulation of fibrin. Myocarditis was observed adjacent to pericardium characterized by congestion and severe infiltration of lymphocytes. Liver revealed perihepatitis characterized by infiltration of mononuclear cells mainly lymphocytes and hepatitis characterized by inflammatory cells mainly mononuclear cells around portal triad area and mild fatty changes. In lungs, there was pleuritis characterized by presence of fibrin and infiltration of lymphocytes. In the parenchyma, pneumonic lesions were observed characterized by infiltration of lymphocytes and macrophages. Spleen revealed severe depletion of lymphocytes along with proliferation of reticulo-endothelial cells, necrotic areas and congestion of blood vessels. The severity of lesions in spleen was severe on 14 DPI as compared to 7DPI. Bursa of Fabricius revealed severe atrophy of bursal follicles characterized by depletion of lymphocytes and vacuoles formation along with infiltration of few heterophils and reticulo endothelial cells proliferation in the inter-follicular spaces. Intestines exhibited enteritis characterized by congestion, mononuclear cells infiltration in mucosa and focal necrosis of villi. Proventriculus revealed serositis characterized by presence of fibrin and infiltration of heterophils and lymphocytes in the serosal layer.

On 21 DPI, pericarditis was evident in heart though of mild degree. Similarly, liver revealed mild perihepatitis and hepatitis at focal areas associated with infiltration of few lymphocytes and macrophages. Mild congestion was noticed in various organs such as spleen, lungs, kidneys and

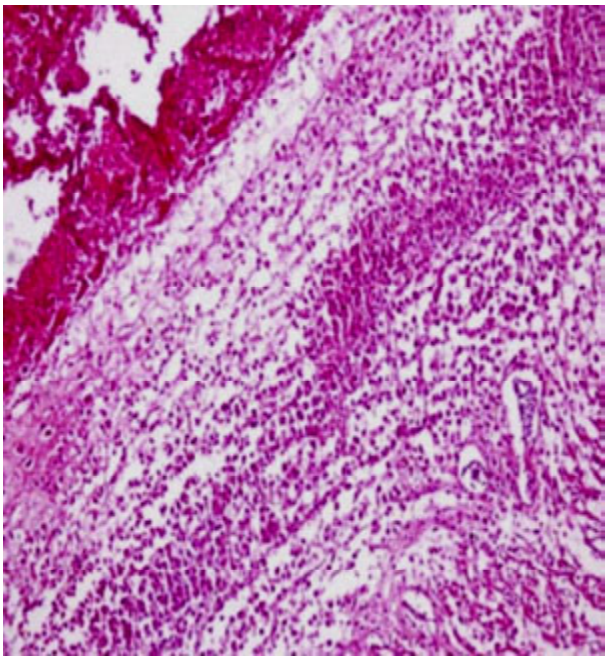

Fig. 3: (Group A2, 7DPI) Heart- Severe pericarditis characterized by fibrin layers (arrow) mixed with large number of heterophils and few lymphocytes (H&E X 200)

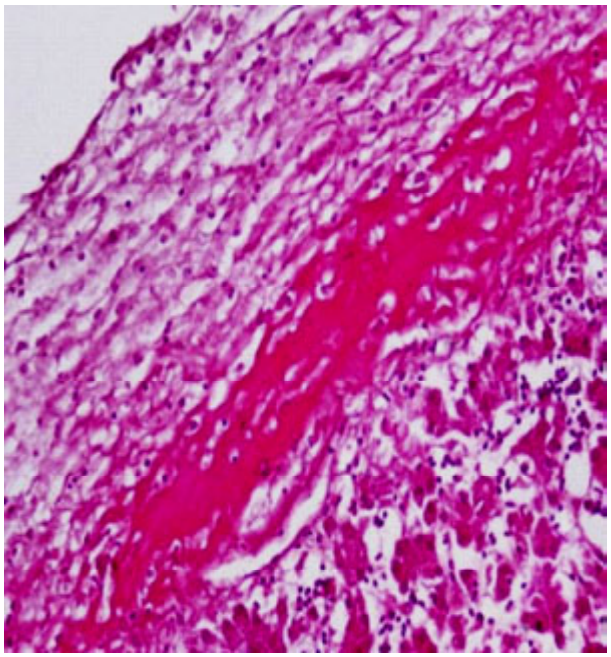

Fig. 4: (Group A2, 7DPI) Liver- Perihepatitis characterized by thick layers of fibrin (arrow) associated with heterophilic and lymphocytic infiltration. (H&E X 400)

Table 1:Mean viable bacterial cell count (X10^{6}CFU/gram of liver, mean ± S.E.) in *E. coli* infected groups of broiler chicks

Groups	Days post infection			
	7	14	21	28
A2	1.22 ± 0.32[a]	3.06 ± 1.00[a]	0.014 ± 0.01[a]	0.002 ± 0.001[a]
B2	1.02 ± 0.38[a]	2.10 ± 0.49[a]	0.007 ± 0.003[a]	0.00 ± 0.00[a]

a: Means with superscript in the column did not differ significantly (P < 0.05)

intestines. Bursa of Fabricius and spleen revealed mild depletion of lymphocytes. On 28 DPI, mild congestion was noticed in heart and liver. There were lymphocytic aggregations in the liver and spleen. Other organs did not reveal any significant lesions.

Group B2 (*E. coli* infected + AVL extract)

On 7 DPI, heart revealed pericarditis characterized by presence of fibrin and infiltration of heterophils and lymphocytes. In liver, there was perihepatitis, congestion, mild fatty changes in hepatocytes along with infiltration of heterophils and lymphocytes. Lungs revealed severe broncho-pneumonia characterized by congestion and infiltration of lymphocytes, heterophils and macrophages in the alveoli and bronchioles. The severity of lesions in lungs of AVL extract supplemented infected group was comparatively higher than other infected groups A2 and B2. Spleen revealed necrosis and depletion of lymphocytes in white pulp along with reticulo-endothelial cells proliferation. Bursa of Fabricius revealed depletion of lymphocytes in the follicles. Intestines revealed enteritis characterized by infiltration of heterophils and lymphocytes in the mucosa along with desquamation of the mucosal epithelium.

On 14 DPI, severe pericarditis and myocarditis characterized by accumulation of fibrin and infiltration of lymphocytes and macrophages was observed. The severity of lesions in heart on 14 DPI was of higher magnitude as compared to 7 DPI. Liver revealed perihepatitis characterized by infiltration of lymphocytes, few heterophils and macrophages. Lungs revealed congestion and mononuclear cells infiltration in the alveoli and parenchyma. In spleen, there were necrotic foci with depletion of lymphocytes in the white pulp and the severity of the lesions was significantly less compared to group A2. In bursa of Fabricius too, mild depletion of lymphocytes was noticed in the

Table 2: Per cent mean gross lesion score in various organs in different experimental groups

Organs	Groups	Days post infection				
		0	7	14	21	28
Heart	A2	0	80	80	30	0
	B2	0	75	75	20	0
Liver	A2	0	85	80	15	0
	B2	0	75	60	15	0
Lungs	A2	0	60	60	15	0
	B2	0	65	45	15	0
Spleen	A2	0	60	50	20	0
	B2	0	55	35	15	0
Bursa	A2	0	55	55	10	0
	B2	0	50	35	10	0
Intestine	A2	0	25	25	5	0
	B2	0	25	20	5	0

Table 3: Per cent mean histopathological lesion score in various organs in different experimental groups

Organs	Groups	Days post infection				
		0	7	14	21	28
Heart	A2	0	75	55	25	5
	B2	0	65	70	10	0
Liver	A2	0	75	45	25	5
	B2	0	60	50	10	5
Lungs	A2	0	65	60	25	10
	B2	0	85	55	15	10
Spleen	A2	0	55	60	15	0
	B2	0	55	30	15	0
Bursa	A2	0	75	70	30	0
	B2	0	55	30	10	0
Intestine	A2	0	10	15	0	0
	B2	0	25	5	0	0

Table 4: Overall per cent mean lesion scores irrespective of post infection period and organs in different experimental groups

Lesion score	Groups	Overall per cent mean lesion scores irrespective of post infection period and organs
GLS	Group A2	27.00
	Group B2	23.17
HLS	Group A2	26.67
	Group B2	22.00

follicles which was significantly lower as compared to group A2. In proventriculus, there was mild fibrin deposition in serosa and infiltration of mainly heterophils. Intestines revealed enteritis characterized by mild destruction of villi and infiltration of heterophils.

On 21 DPI, in the heart infiltration of lymphocytes, plasma cells and macrophages along with few fibrin strands was noticed. In liver, there was congestion of blood vessels and mild infiltration of lymphocytes around the portal triad. Aggregations of lymphocytes were noticed in the liver parenchyma. In spleen secondary follicles were observed. Mild congestion was noticed in lungs. Nothing abnormal was noticed in the other organs like proventriculus, intestine and kidney. Bursa of Fabricius was almost normal with presence of lymphocytes in the medulla of the follicles.

On 28 DPI, nothing abnormal was observed in any of the organs except presence of lymphocytic aggregations in liver and secondary lymphoid follicles in spleen.

Histopathological changes observed in heart due to colibacillosis in the present study were fibrinous pericarditis and myocarditis characterized by accumulation of fibrin, infiltration of heterophils and lymphocytes in early stage and macrophages in later stage. Similarly, liver revealed fibrinous perihepatitis and hepatitis along with degenerative/necrotic changes in hepatocytes at different intervals of post infection. Similar changes have been reported by other workers (Jindal *et al.,* 2003; Verma and Swamy, 2013; Kumari and Gupta, 2014a) in chickens infected with *E. coli*.

These histopathological features of fibrinous pericarditis and perihepatitis were of lower magnitude in AVL extract supplemented infected groups at different intervals. Antioxidant activity of *A. vera* might have contributed for hepatoprotective and cardioprotective potential of *A. vera* as these extracts have ability to scavenge highly reactive free radicals formed in the cells during normal metabolism as well as infection (Rajasekaran *et al.,* 2005; Jain, 2012; Kumari *et al*., 2023). The differences in severity of hepatic and cardiac lesions between the control infected and extract supplemented infected groups might also be attributed to anti-lipoperoxidative activity and anti-inflammatory activity of the extracts (Mahmoud *et al*., 2012). Hepatoprotective and cardioprotective potential of *A. vera* leaf extract has also been demonstrated by different workers in laboratory animals and poultry (Waihenya *et al*., 2002; Kaithwas *et al*., 2011; Kumari *et al*., 2023). The improved antioxidant activity due to *A. vera* administration helps to neutralize the free radicals formed during the *E. coli* infection, this results in less damage to cells and hence lower magnitude of inflammatory lesions observed in liver and heart.

In lungs, congestion, hemorrhages and mild oedema were noticed in early stage and pneumonic lesions at later stage of the infection. Similar respiratory lesions have been reported by other workers (Baliarsingh *et al.,* 1993; Gangane *et al*., 2006; Verma and Swamy, 2013; Kumari and Gupta, 2014a) in *E. coli* infected chickens. The AVL extract supplemented infected group exhibited severe lung lesions as in control infected group.

Spleen and bursa of Fabricius revealed necrosis and depletion of lymphocytes in the control infected group indicating immunosuppression due to *E. coli* infection AVL extract supplemented infected groups revealed significantly less lesions in spleen and in bursa of Fabricius as compared to control infected group. These results revealed that colibacillosis caused immunosuppression and are in accordance with findings of Hegazy *et al.* (2010) and Kumari and Gupta (2014a) who also reported depletion of lymphocytes mainly in bursa of Fabricius in *E. coli* infection in chickens. The difference in severity of lesions in the extract supplemented groups might be due to immunomodulatory effect of these extracts (Wang *et al.*, 2007; Akhtar *et al*., 2012; Channa *et al*., 2014; Shokraneh *et al*., 2016; Kumari *et al*., 2023). Intestines revealed congestion and enteritis in broiler chicks infected with *E. coli* though the lesions were mild. Proventriculus, in the present study revealed proventriculitis characterized by infiltration of heterophils and mononuclear cells in the serosa. The severity of lesions in these organs

did not differ much between infected groups.*E. coli* causes depletion of lymphocytes in lymphoid organs while *A. vera* administration improves the immunity and thus helps to reduce the damage to organs. Although at the present dose level considerable difference were not observed between infected and infected plus supplemented group.

Lesion score

Based on the colibacillosis specific mean gross and histopathological lesion scores (GLS and HLS) in various organs (heart, liver, spleen, bursa of Fabricius, lungs and intestines) in different experimental groups, the overall per cent mean GLS and HLS in different organs irrespective of post infection period was calculated (Table 2 and 3). The lesion score was lower in supplemented infected group as compared to non-supplemented infected group. Overall per cent mean lesion scores irrespective of post infection period and organs in different experimental groups is presented in Table 4. The protective effect of AVL extract on gross and histopathological lesions of *E. coli* infection was found to be 14.19% and 17.5%, respectively.

Overall histopathological findings in different organs suggested that*A. vera* leaves extract caused only mild reduction in the severity of lesions due to *E. coli* infection. Although, *A vera* is widely used for the external treatment of minor wounds, skin irritations including burns, bruises and abrasions, and general inflammatory skin disorders (Mwale and Masika, 2010) and possess good antibacterial activity in *in-vitro s*tudies (Kumari and Gupta, 2014b). The literature reported that *A. vera* leaf extract also possess antioxidant, anti-inflammatory and immunomodulatory activities so the present study was done to observe the effect of *A. vera* in broilers against *E. coli*infection but the results of *in vivo* experimental study in broiler chicken revealed that at the present dose the differences in lesion score were not considerable and *A. vera* do not help considerably in reducing the infection systemically.

CONCLUSION

It is concluded that, *A. vera* leaves extract supplementation did not show considerable ameliorating effect on pathological changes of *E. coli* infection at the dose used in the present study. Further research is required using different doses to explore its use against bacterial infections as literature suggests that it possess good antibacterial and antioxidant activities.

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