DEPARTMENT OF VETERINARY PATHOLOGY
Bihar Veterinary College, Patna- 800014
(Bihar animal Sciences university Patna, Bihar)

Title o of thesis: **Clinico-Pathological Studies and therapeutic effects of febuxostat on visceral gout in vanaraja chicken"**.

Name of student: Dr. Kanchan Rawal Admission number: BVC/M/VPP/003/2017-18

Major discipline: Veterinary Pathology Minor discipline (s): Veterinary Medicine
Date of thesis thesis submission: Total pages of the thesis: 73
Major Advisor: Dr. Deepak Kumar

Visceral gout have prime economic important in poultry due to increased incidence causing production loss, regular mortally and lack of availability of specific treatment. The present research work was conducted on four groups of vanaraja chickens to establish the effect of febuxostat and bottle gourd juice at various dose levels in preventing gout with urates in aceclofenac induced toxicity. Group I was kept as control. Group II was administered with acelofenac at dose of 20 mg/kg vanaraja chickens through feed. Group III was administered with febuxostat (2mg/kg) and aceclofenac (20 mg/kg). Groups IV, were administered with aceclofenac at dose rate of 20 mg/kg vanaraja chickens through oral feed respectively for 28 days. The vanaraja chickens observed daily for any abnormal behavioral clinical signs and mortality as well as weekly body weight during the experiment. Survived vanaraja Chicken from each group were sacrificed by cervical dislocation at the end of experiment and blood samples were collected before sacrifice for hematology and biochemical analysis. A detailed post mortem examination was performed on vanaraja chickens which died during the experiment as well as gross lesions were recorded. Tissue samples (liver, kidney, heart,) were collected in 10% neutral buffered formalin for histopathological examination and. In aceclofenac group II and group IV on the day 14, 21 and 28 day revealed 27.77 %. mortality and typical clinical signs,, decreased body weight, significant increased Hb, PCV, TLC, TEC and lymphocyte, uric acid, creatinine, albumin, SGPT, SGOT and alkaline phosphatase and histopathological changes were observed. End of the 28 day visceral gout affected vanaraja chickens in group II and group IV treated with febuxostat and bottle gourd juice. Aceclofenac administration at the dose rate of 20 mg/kg and above for period of 28 days, induced visceral gout in vanaraja chickens Febuxostat and bottle gourd juice treated groups II and IV suggested that which is well as safe to use without any side effects in vanaraja chickens. This suggested beneficial effect of febuxostat and bottle gourd juice in elimination of urate deposition from the kidneys and thus preventing the visceral organ.

Dr.Kanchan Rawal
(M.V.Sc Scholar)

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CLINICO-PATHOLOGICAL STUDIES AND THERAPEUTIC EFFECTS OF FEBUXOSTAT ON VISCERAL GOUT IN VANARAJA CHICKEN

Dr. Kanchan Rawal

Admission. No –BVC/ M/VPP/003/2017-18

SUBMITTED TO

BIHAR ANIMAL SCIENCES UNIVERSITY,

(BIHAR VETERINARY COLLEGE)
PATNA, BIHAR

In partial fulfillment of the requirements
For the degree of

Master of Veterinary Science
IN

(DEPARTMENT OF VETERINARY PATHOLOGY)

BIHAR VETERINARY COLLEGE
PATNA 800014

2019

CERTIFICATE-I

This is to certify that the thesis entitled "**Clinico-Pathological studies and therapeutic effects of febuxostat on visceral gout in vanaraja chicken.**" Submitted in partial fulfillment of requirement for the award of the degree of **Master of Veterinary Science (in the discipline of Veterinary Pathology)** of the faculty of post-Graduate Studies, **Bihar Animal Sciences University, Patna,** is the bonafide research work carried out by **(Dr. Kanchan Rawal)** daughter of (Mrs. Sita Devi and Late. Shri Sita Ram Rawal) under my supervision and that no part of this thesis has been submitted for any other degree or diploma.

The assistance and help received during the course of this investigation have been fully acknowledged.

Dr. Deepak Kumar

(Signature and name of the Major Advisor)

Place:

Date:

CERTIFICATE- II

This is to certify that the thesis entitled "**Clinico-Pathological studies and therapeutic effects of febuxostat on visceral gout in vanaraja chicken"**submitted by **(Dr. Kanchan Rawal, Admission No- BVC/M/VPP/003/2017-18),** daughter of Mrs. Sita Devi and Late Shri Sita Ram Rawal to Bihar Animal Sciences University Patna, Bihar in partial fulfillment of the requirement for the degree of **Master of Veterinary Science** in the discipline of **Veterinary Pathology** has been approved by Advisory committee after an oral examination of the student in collaboration with an External Examiner.

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(Kanchan Rawal)

Place: (Signature & Name of the student)

Date:

CONTENTS

ABBREVIATIONS

%	Percent
&	And
µ	Micron
µl	Microliter
U/L	Unit Per liter
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AKP	Alkaline Phosphatase
B.wt	Body Weight
BCG	Bromo Cresol Green
DNA	Deoxyribose nucleic acid
e.g.	Exampli gratia
et al.	Et alibi
FDA	Food And Drug Administration
Etc	Etcetera
ED50	Half maximum effective dose
DLC	Different leucocyte count
G	Gram
GDP	Gross Domestic Product
GGT	Gamma Glutamyl transferase
H	Hour

H&E	Haematoxylin and Eosin
IAEC	Institutional Animal Ethics Committee
ILFC	Integrated Livestock farm Complex
i.e	id est
IBD	Infectious Bronchitis Disease
IUPAC	International unit of pure and applied chemistry
IP	Indian pharmacopeia
K_3EDTA	Tripotassium Ethylene DiamineTetraaciticacid
LAP	Leucine Amino peptidase
LDH	Lactate Dehydrogenase
L	Litre
Ltd	Limited
Min	Minute
Ml	Millilitre
mg	Milligram
Mg/dl	Milligram per Decilitre
Mg/g	Milligram per gram
Mg/kg	Milligram per Kilogram
Mg/ml	Milligram per Millilitre
NSAIDs	Non-steroidal Anti Inflammatory Drugs
PG	Prostaglandin

PCV	Packed Cell Volume
Ppm	Parts Per Million
PCT	Proximal Convoluted Tubule
Pvt. Ltd	Private Limited
SE	Standard Error
SGPT	Serum Glutamic pyruvic Transaminase
SGOT	Serum Glutamic Oxaloacetic Transaminase
TLC	Total Leukocyte Count
TEC	Total Erythrocyte Count
TSP	Total Serum Protein
UA	Uric Acid
U/L	International Unit per Litre
Viz	Namely
WBC	White Blood Cell
XO	Xanthene Oxidase
XOR	Xanthene Oxido-reductase

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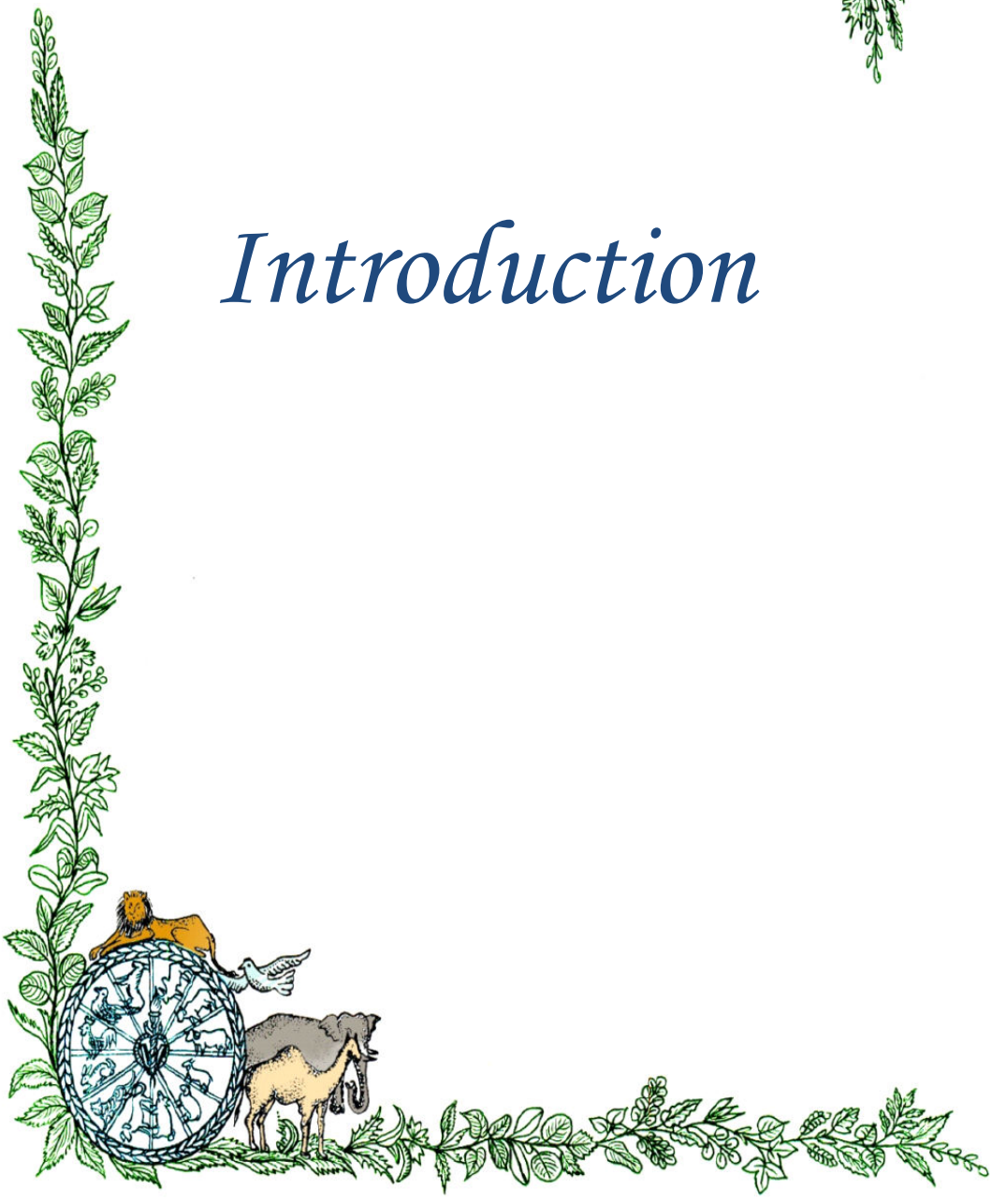
Introduction



1 Introduction

India is among developing country, Poultry industry is also developed and now days it contributed a major part in the Indian economic. Indian poultry industry is one of the fastest growing segments of the Agricultural sector. Poultry industry in India is a vibrant, fast growing and dynamic sub-sector of agriculture. Poultry population in India is 729.20 million (12.39%increase).

Vanaraja chicken bird is one of the colored birds for both egg and meat. Vanaraja bird (10-20) can be reared under free range condition for egg purpose. In case commercial farming these bird can be reared under intensive and semi intensive condition providing all necessary inputs just like commercial broiler, backyard poultry breed with an egg production of 150 to 200 and 1.5 kg body weight at 10 to 14 weeks of age is visible in rural village condition. Ayyagari (2001) described genetic architecture of 'vanaraja' germplasm developed directorate on poultry, Hyderabad. This dual purpose bird was developed by crossing Red Cornish males with random bred –meat control population females. These birds were famous for their attractive plumage, better survivability, immune competence and large sized eggs.

From last 3 decade, has been increase poultry production remarkable. This is due to the mainly improvement of stock performance through better management and disease control practices. Birds are suffer from the most infectious and non-infectious diseases and metabolic disease and disorder. Infectious disease can be controlled by medication and vaccination. In case of metabolic diseases, which Couse considerable loses to the poultry industry needed different approach for the control. In poultry commercial varieties of broiler and layer introduced new metabolic diseases related with cardiovascular system like ascites, urinary system like visceral gout etc. these diseases are becoming major cause of mortality today and cause heavy losses to the farmers. Sandhyarani (2019)

In the present days of high demand for meat and eggs. In poultry bird is genetically engineered for higher productivity and its selection is based on production parameters. In this process health of the vital organs is ignored. It has resulted in increased incidence of several metabolic disorders. The kidney is one such vital organ of birds with diverse metabolic and excretory functions. Gout is a common metabolic disorder that results in abnormal

accumulation of urates in domestic birds. It occurs as two distinct forms, namely, visceral and articular gout. Amravathi *et al.,* 2015; Feizi*et al.,* 2012; these two syndrome differ in age of onset, frequency, sex predilection, causes, and pathogenesis, gross and microscopic lesions. Shivaprasad, (1998)

Visceral gout is a common metabolic disorder characterized by high level of uric acid in the blood lead to deposition of urates on the surface of various visceral organ, in some cases, surface of muscle and synovial sheath of tendons and deposition may also occur on the surface of heart, liver, spleen, kidney and peritoneum appear as white chalky coating,(Mudasir *et al.,* 2005) This condition can occur as an individual problem at any age but outbreaks are seen in young chicks in the first and second week of life. Clinical sign such as Chalk-like urate deposits on pericardial sac and liver capsule, Increased thirst (polydipsia), and decreased appetite, abnormal droppings. Chalky urates in their stool, feather plucking, plumage (Patel *et al.,* 2007).

Visceral gout have prime economic important in poultry due to increased incidence causing production loss, regular mortally and lack of availability of specific treatment. Visceral gout has been reported in various caged and aviary birds from different parts of the world. (Dhara *et al.,* 2010). The deposits on serosal surfaces appear grossly as a white chalky coating, and those within viscera may be recognized only microscopically. Visceral urate deposition is generally due to a failure of urinary excretion which may be due to obstruction of ureters, (Crespo and Shivaprasad, 2003, Sodhi *et al*., 2008 and Eldaghayesn *et al*., 2010).

Uric acid is the primary catabolic product of protein, non-protein nitrogen and purines in birds. It is synthesized mainly in liver of birds. Birds excrete uric acid as primary nitrogen metabolite as calcium sodium urates which are not water soluble, this is called ureotelic metabolism (phatak, 2002)

Birds are Uricotelic and lack Uricase enzyme which convert uric acid to less harmful substance. Uric acid itself is not toxic but precipitated crystal can cause severe mechanical damage to tissue like kidney, heart, lung intestine, and also in the joint (Fitz-Coy *et al.,* 1988; Nayak *et al.,* 1988; Rao *et al.,* 1993).

The cause for the development of kidney lesions is usually multi factorial and may be attributed to nutritional deficiencies, managemental problems, toxins and infectious agents. In addition, the urinary system of poultry is so unique that the blood can be directed from the

lower limbs straight into the renal parenchyma this may increase the effect of nephrotoxic drugs and/or enhance elimination by taking the compound directly to the kidney. (Sathiyaseelan *et al.,* 2018).

There is need to` search for the newer cause of visceral gout. Research published in the scientific journal, nature in January 2004 has confirmed that "Veterinary use of diclofenac is responsible for the recent devastating decline vulture population. In Veterinary practices diclofenac is ban, but other non-steroidal anti-inflammatory drugs such as aceclofenac, ketoprofen, caprofen are still used. Aceclofenac bears close structural and path morphological resemblance to diclofenac. Hinz *et al.* (2003) showed that aceclofenac could slowly but sustainably bio transform into diclofenac. This drug lethal to the birds in the dosages that would be given to livestock to reduce pain and swelling of those animals suffering from arthritis. The BNHC (Bombay Natural History Society) is calling for tighter control on the use of this drug in veterinary practice in southern Asia. (Shalya, 2012).Poultry feed contain many animal source by-product like meat, and bone meal, blood meal, bone based DCP, mutton etc,which are likely to have residues and probably responsible for visceral gout in birds.

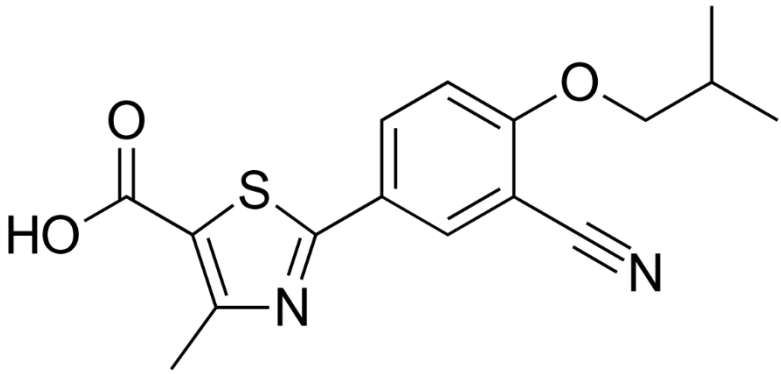
Aceclofenac (2-[2-[2-[2, 6,- dichlorophenyl) aminophenyl] acetyl] oxyacetic acid; is a non-steroidal anti-inflammatory drug, Structurally aceclofenac, diclofenac and ketoprofen are all related as Arylpropionic acid derivatives(Aronson,2009) it inhibits both cyclooxygenase-1 (COX-1) and cyclooxygenase-2 (COX-2) , with the evident selective COX-2 inhibition (Gonzalez *et al.,* 1994) . The COX- inhibitory action of Aceclofenac is due to limited but sustained biotransformation in to diclofenac has been substantiated convincingly for human subject (Hinz *et al*., 2003). Aceclofenac is rapidly absorbed from the gastrointestinal tract and 99% bound plasma protein and 100% bioavailability. This drug eliminated through primarily from renal excretion, with 70-80% of the administered dose found in urine as glucoronide and rest is excreted in faeces. In animal, aceclofenac is used in the treatment of inflammatory and painful condition of bones, joints, and musculo-skeletal systems cattle, horses, dogs and cat this drug affect metabolism of animals.

Febuxostat

Febuxostat is non-purine selective inhibitor of xanthine oxidase (XO), xanthine dehydrogenase. It is clinical development as an orally administered agent for the treatment of hyperuricaemia and gout. Febuxostat lowers serum urate levels more potential than

allopurinol and it bind to enzyme by means of a different mechanism from that used by a metabolite of allopurinol oxypurinol.

Chemical structure of Febuxostat



Febuxostat is administrated through orally and is quickly absorbed through gastrointestinal tract. The absolute bioavailability of Febuxostat was determine to be approximately 78 % in rat and in human 49%. Concomitant food intake reduce the absorption of Febuxostat. (Anonymous, 2008). Febuxostat was rapidly absorbed with a medium time to reaches its maximum plasma concentration in 1 to 1.5 hrs after the dose is taken following oral absorption, approximately 85% of the drug is absorbed. Although the rate and extent of absorption may decrease with food intake and antacid use, no clinically significant change in the effect of Febuxostat has been reported, therefore, it may be taken without regard to food or antacid consumption. There is no accumulation when it is given in therapeutic doses in daily interval. It is approximately 99.2 % protein bound, primarily to albumin. (Khosravan *et al.,* 2006)

Febuxostat is eliminated by both renal and hepatic pathways. However, renal clearance is not significant. Only a small amount of unchanged drug or metabolites is excreted in the urine. The half-life is approximately five to eight hours. Over a 24-hour period, Febuxostat results in a dose-dependent decrease in serum uric acid concentrations. As a result, total daily urinary uric acid excretions are reduced with an increase in total daily urinary xanthine excretion. (Khosravan *et al.,* 2006).

Pathway of purine metabolism and mechanism action of Febuxostat

Uric acid is end product of purine metabolism in birds and is generated in the cascade of hypoxanthine – xanthine –uric acid. Both steps in the above transformation are catalyzed by xanthine oxidase. Febuxostat is a 2- arylthizole derivative that achieves its therapeutic effect of decreasing serum uric acid by selectively inhibiting XO. Febuxostat is potent non purine selective inhibitor of XO with an in vitro inhibition Ki value less than one Nano molar.

Bottle gourd

*Lagenariasiceraria*commonly known as Bottle gourd Syn. Doodhi, syn Lauki (Hindi), Kadoo (Marathi) which is official in Ayurvedic Pharmacopoeia. It is one of the excellent fruit for human being made and gifted by the nature having composition of all the essential constituents that are required for normal and good human health (Habibur, 2003). It's time to focus attention overall constituents and character of *Lagenariasiceraria* fruit for the better human health and lives. Two varieties of this fruit drug sweet and bitter are mentioned. Botanically, both belong to same genus, the former known by the Sanskrit synonymalba and tumbi and later by the names as Iksuaku, Katutumbi and mahaphala. The sweet variety is generally used as a vegetable, while the wild variety bitter, latter is preferred for the medicinal use. The former variety is cultivated widely for its fruit and vegetable. The latter is found wild previously in most areas but now in some hot areas of country, obviously as wild and has bitter fruits and preferred for medicinal use, the difficulty in procuring and losing interest in cultivation of wild variety, the sweet and edible variety is now being used in medicine as well.(Sivarajan and Balchandra, 1994)

The plant *Legenaria Siceraria* common known as bottle gourd. It is a one of excellent fruits. Bottle gourd have many properties like, vitamins, iron and minerals. It is contain the triterepenoidcucurbitacine B,D,G,H, two sterol viz fucosterol, campesterol, flavone- C glycoside and lagenin. Bottle gourd is useful in many diseases like, cardiac disorder, ulcer and hepatic disease. Bottle gourd juice help to regulate blood pressure of hypertensive patient, because high potassium content. the edible portion of fruits is fair source of ascorbic acid, beta carotene and good source of vitamin B complex, pectin dietary soluble fibres and contains highest source of choline level-a lipotropic factor, a healer of mental disorders, along with required metabolic and metabolite precursors for brain function, amongst any

other vegetable known to man till date. It is also good source of minerals and amino acids (Nadkarni, 1992)

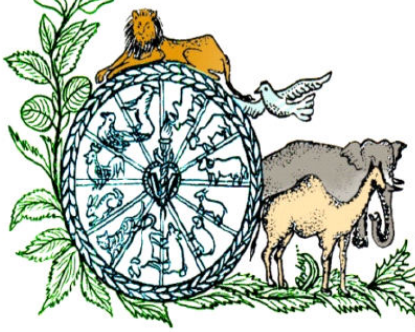
Peroxidase and polyperoxidase activity in relation to its blanching period and total enzymatic inactivation of blanched sample (i.e, residual peroxidase activity is less than one) is also reported in 180 seconds. In addition, small amount of unidentified mono-and di-caffeoylquinic acid derivative was detected. 30% inhibition of superoxide formation in xanthine and xanthine oxidase medium by methanolic extract from fruit of *Lagenariasicerari*a is reported. (Jiwjinda *et al.,* 2002)

No adequate scientific studies on visceral gout in Vanaraja chicken have been carried out in Bihar state , keeping in view its economic significance and frequent occurrence in poultry , it is necessary to study the Clinico- pathological and newly drug treatment aspect of gout with the following objectives –

Objectives

1. To investigate possible role of the high dose Aceclofenac in the causation of visceral gout
2. To Study the effect of Febuxostat on visceral gout in vanaraja chicken
3 To Study the Haemato-Biochemical changes in experimental cases of visceral Gout.
4 To Study the Histo-Pathological changes in experimental cases of visceral gout

Review Of Literature



2 Review of literature

A Brief Resume of Work done in India and abroad

Gout is a common metabolic disorder. It is condition in which high level of uric acid in the blood. Its lead deposition of urate crystal on the surfaces of various internal organs, and various joints, gout is not disease condition, but a clinical sign of severe kidney failure (except articular gout), Gout condition is commonly and practically encountered in commercial poultry birds. Extensive studies on incidence and etiology of gout have been done by several workers. Clinico-pathological, hematological, and serum biochemical changes in natural cases of gout and nephritis in poultry have also been studied by several scientists. The following review describe on the parameters which are include in the present study

Mortality rate

Jackson *et al.* (1972) reported in the first week of life 2.08 percent mortality due to yolk sac infection, visceral gout and starvation syndrome in their survey of nine batches of broiler chicks.

Bimal Chandra *et al.* (1973) recorded severe mortality due to visceral gout or gouty nephritis.

Rahamathulla and Mohiyudeen (1973) recorded the lesions in kidney and liver in poultry and in one flock they showed on autopsy uric acid nephritis associated with necrotic hepatitis, thus confirming as gouty nephritis and observed a mortality rate of 8 to 15 per cent in the flocks.

Nayak *et al* (1988) discovered in gout cases 300 broiler birds in an organized farm in the district of Nadia, west Bengal. out of 300 broiler birds 181 birds (60.33%) died. Heal so finding highest mortality rate (26.33%) during 4^{th} week followed by 3^{rd} week (20%) and 2^{nd} week (14%) of age.

Srivastava *et al.* (2001) reported that visceral gout usually occurs in broiler chicks within 3 weeks of age. Maximum mortality was observed in 6^{th} and 10^{th} day of age. He also observed maximum mortality in colder month of November and April.

Reddy *et al.* (2006) observed that 40% mortality within12 days of administration at the dose rate of 5 mg/Kg diclofenac of body weight in vanaraja.

Eldaghayes *et al.* (2010) studied the pathology of visceral gout in growing layer farm of 45000 birds located in Tripoli, Libya and observed a mortality of 489 birds (1.08%) within 10 days span.

Sharma *et al.* (2012) observed diclofenac sodium toxicity in broiler birds. They observed 40% mortality at the end of 3rd week.

Ghodasara *et al.* (2014) conducted toxicopathological studies of meloxicam, ibuprofen and diclofenac sodium in broiler chicks. They reported 25% mortality in diclofenac treated group at dose rate of 15 ppm in feed.This was noticed from day 8 to day 14 of the experiment.

Akhter and Sarker (2015) observed percent mortality in different groups fed diclofenac by oral route. Group A was treated with @ 5mg/kg body weight, group B was treated with @ 10mg/kg body weight and Group C was treated with @ 20 mg/kg body weight Diclofenac orally mixing with drinking water. They reported 30% mortality rate in group A, 60% mortality in group B and 70% mortality in group C. Birds of Control group did not show any signs and symptoms of toxicity.

Ramzan *et al*. (2015) assessed diclofenac sodium toxicity at different concentrations in relation to time using broiler chicken model. They noted percent mortality in different treatment groups showed increasing manner with greater dose of diclofenac used by Oral route. Diclofenac sodium in oral route by dose rate of 2.5mg/kg, 5.0mg/kg, 10mg/kg revealed 10, 30, 50 percent mortality respectively.

Drugs and chemical

Mubarak and Sharkawy (1999) reported gout induced in laying pullets by sodium bicarbonate toxicity which revealed significant microscopic changes and urate deposits associated with tubular necrosis.

Phatak (2001) suggested that antibiotic like gentamycin, erythromycin, sulfonamide etc. used to be excreted through kidney and tend to cause renal damage resulting in visceral gout.

Oaks *et al.* (2004) Diclofenac was responsible for the decrease of vulture population in South Asia.

Patel *et al. (*2014) administration Aceclofenac at the dose rate of more than 20 mg/kg induces toxicity symptom in layers. Pathomorphological lesions induced by aceclofenac toxicity were vascular and degenerative in nature mainly affecting kidney, heart, liver, lung, proventricular and gizzard.

Bottle gourd

Anonymous (1996) bottle gourd is good source of beta carotene, ascorbic acid, and also good source of vitamin B complex, pectin dietary soluble fibres and contains main source of choline level-a lipotropic factor, a healer of mental disorders, along with required metabolic and metabolite precursors for brain function,

Bsbi List (2007) Bottle gourd also known as white flowered gourd, synonym are *lauki* and *doodh*, which is being used as vegetable.

Kausar and Waris (2016) reported that excess uric acid is removed from the body by using different drugs which also produces harmful effect on other organs in many cases. But they were studied; treatment was done by bottle gourd which helps in removing excess uric acid from the blood without affecting other organs. Bottle gourd orally treated rats revealed a good effect on liver function test as compare to extract and Zyloric drug treated rats. He also reported that Bottle gourd is found to be the healer of mental disorder due to its highest content of choline than any other vegetables and minerals, vitamins. And any other amino acid that are present in it. Are used to synthesize neurotransmitters. Bottle gourd have different functions such as good to help digestion, urinary disorders, refreshes during summers, prevent heart disease , reduces stress etc.

Clinical sign

Cristopher *et al.* (1977) reported that induced gout in birds through the excess feeding of sodium bicarbonate which revealed symptom as of anemia and weakness.

Kaushik and Kalara (1979) recorded the incidences of gout in poultry observed symptom like body weight gain, poor growth and low feed consumption.

Mishra *et al.* (1981) defined rapid growth of initially in gout followed by dullness and loss of appetite.

Jagani (1985) visceral gout affected layer bird's revealed signs like comb emaciation, dehydration and dark colorations of wattles, underdeveloped comb and wattle.

Riddell (1987) reported that most common cause symptoms include anorexia and emaciation.

Chaudhry *et al.* (1988) defined signs of gout in broiler as loose droppings, dullness, decreased appetite and greenish diarrhea. Deaths were unexpected and growth rate was poorer.

Lumeij *et al.* (1994) recorded clinical symptom of visceral gout in broilers like, lethargy and anorexia may be prominent for hours or days. Depression became severe with the passage of time and the affected birds stopped eating and drinking.

Naidoo *et al.* (2007) observed toxicity of diclofenac in 18 week old layers. Affected birds were reluctant to move, perched in one corner of their cages, appeared to be asleep and also were anorexic. Earlier to death, the affected birds acted to be comatose and could not be roused.

Anonymous (2008) issued single dose oral toxicity study of Febuxostat in rats and dogs. Rat exhibited reduced locomotor activity after 5-25 minutes, cyanosis lying on its side and reduced body temperature after dose rate of 600mg/kg. Dogs exhibited severe vomiting, depression, reduced locomotor activity, salvation and loose stool after giving dose at the rate of 2000mg/kg.

Irtaza *et al*. (2008) recorded toxico-pathological effects of diclofenac in four avian species including broiler chicks. Clinical signs in all species included depression, somnolence, reduced body weight.

Seema (2008) detected age associated pathological effects of diclofenac sodium in Cobb-400 and WLH male chicks. Cobb-400 birds appeared dehydrated, emaciated, depressed

and lethargic with shrunken eyes. They also showed signs like feather plucking, anorexia, low feed intake and decrease growth, involvement of joints was detected in some of the birds.

Jain *et al*. (2009) recorded diclofenac induced biochemical and histopathological changes in white leghorn birds (*Gallus domesticus*). Diclofenac treated birds showed mild dullness, diarrhea, lethargy, bloody diarrhea segregation and anorexia.

Jana *et al*. (2009) noticed gout in broiler birds in West Bengal revealed that birds affected by gout indicated dullness, loss feed intake and decrease body weight, diarrhea and death.

Sharma and Vegad (2010) described depression, dehydration, ruffled feathers, dullness, moist vent and sometimes with greenish diarrhea in naturally affected gouty birds.

Aworh *et al.* (2012) discovered clinical signs in birds given diclofenac only in group-A include sitting on the hock with eyes closed, decrease feed and water intake, depression and lethargy.

Akhter and Sarker (2015) observed effect of diclofenac sodium in broilers. Birds of Control group did not show any signs and symptoms of toxicity. Birds of Group A, B and C treated with diclofenac revealed mild to severe diarrhea, segregatory behavior, dullness, blood tinged diarrhea and anorexia before death.

Effect on body weight

Shinde (2007) noticed that average body weight and average body weight gain in all diclofenac treatment groups was poor than control in broiler chicks.

Seema (2008) studied age related pathological effect of diclofenac sodium in poultry and detected significant decrease in average body weight in all treatment group cobbs 100 broilers chicks as compare to control group while in white leghorn chicks decrease in body weight was noted only in high dose (150 ppm diclofenac sodium) group.

Irtaza *et al.* (2008) recorded that body weight of birds on day 7 of the experiment did not differ significantly but on day 21of the experiment reduced significantly in broilers and pigeons of all diclofenac-treated group.

Ghodasara *et al.* (2014) discovered reduction in body weight gain was in diclofenac treated group II in comparison with the control at the end of 1st, 2nd and 3rd week. The diclofenac treated group revealed significant reduced at the end of 1st week and higher significant reduction in body weight at the end of 2nd and 3rd week as compared to control group.

Haematological study

Rahamathulla and Mohiyudeen (1973) he study in 1970 -1972 and he observed that a flock of birds was showing uric acid nephritis associated with necrotic hepatitis. The blood picture of the ailing birds which had signs of gout was studied and the number of monocyte was found to be between 18-28%.

Christopher (1977) defined that slight leucocytosis, thrombocytopenia with an increase in the lymphocyte count (shaver brand hens). The erythrocytes became more fragile and their sedimentation rates were increased. He also found a reduced in RBC count with decrease HB and PCV.

Pegram *et al.* (1982) studied no change in haemoglobin, packed cell volume in gouty birds produced experimentally by feeding oosporeine containing diet.

Chandra *et al.* (1985) reported a gradual increase of total erythrocyte counts, haemoglobin, packed cell volume, and total leucocyte counting birds in nephritis. The increase in total Erythrocyte counts, haemoglobin, and packed cell volume might be due to dehydration as a result of diarrhoea.

Koutsos *et al.* (2001) reported an experiment to determine the ability of cockatiels (Nymphicushollandicus) to adapt to high dietary levels and fed diets containing 11, 20, 35, or 35% crude protein for 11 months and found that there were no significant difference in differential count of cells.

Singh *et al.* (2013) observed significantly ($P \leq 0.05$) higher level of TEC, TLC, Hb, and PCV compared to normal birds. DLC showed significantly higher lymphocyte count in gout case as compared to normal birds but there was no significant difference observed in heterophils, eosinophils, monocyte, and basophils count.

Biochemical study

Bell *et al.* (1959) defined that plasma uric acid level in chicken was influenced by nutrition. Age. Sex and reproductive status. They observed that plasma uric acid of laying hens; non-laying hens, immature hens and cockrels were 2.27, 5.40, 4.58, and 2.8 mg /dl of plasma respectively.

Siller *et al.* (1959) reported increase in plasma uric acid level in birds either on high protein diet or during starvation.

Leveille *et al.* (1961) conducted an experiment in which chicks were fed with different levels of protein diet viz. 10, 15, 20 and 25% for 3 weeks and the influence of it on growth parameters, total serum protein and serum cholesterol was studied. Total serum protein increased significantly in chicks fed with10% protein diet to 25 % protein diet. The changes in serum protein could be recognized to variation in albumin level, the level of globulin remaining constant at all protein levels fed.

Okumara and Tasaki (1969) detected that concentration of uric acid level in the blood, kidney and liver were increased respectively to the level of dietary protein. They further reported that birds fed on 40% of casein diet showed 4 and 3 mg of uric acid per G of kidney and liver respectively.

Rajanna (1981) discovered the serum uric acid significant highest level on continuous use of coccidiostats and vitamins A deficient diet.

Ritchie *et al.* (1994) noticed uric acid is the major nitrogen metabolic waste product in birds and affects the concentration of uric acid such as age, diet and laying period.

Uma *et al.* (1997) observed blood uric acid level in gout affected bird was 25.6 to 42.2 mg/dl. Uric acid level in control birds was 6.4 mg/dl, and also observed in cases of gout blood uric acid level crossed 25.6 and severe symptom leading to death when blood serum uric acid level crossed 42.4 mg/dl.

Lin *et al.* (1976) described that the normal blood uric acid level in broilers and layers were 4.85 and 4.87 mg per 100ml of blood respectively. They observed hyperuricemia among affected birds and state that birds with 8.27 to 21.4 mg uric acid per 100 ml of blood and showed slight clinical signs and died due to gout.

Mishra *et al.* (1981) considered the experimental gout in poultry and detected the increased total serum uric acid and excess protein diet in the broilers

Chandra *et al*. (1984) described that experimentally fed five groups of 18 day old chicks each containing 60 birds, with excess protein (42.48%), elevated calcium (3.37%), urea (5%), vitamin A deficient diet and monitored diets (balanced diets) for 15 weeks continuously. Indifferent groups they notice uric acid level to be 50.0, 40.0, 12.0, and 8.5 µg per/dl plasma respectively and also found a significant highest level in AST activity in chicken fed with excess protein diet and increase in ALT activity in chicken fed with urea containing diet (6 weeks) and after 7 weeks in chicks with high protein diet.

Jagani (1985) noticed gout in the flock of layer and estimated serum uric acid in affected and normal birds and also observed the levels of the uric acid to the tune of 13.48 mg percent varying between 2.27 to 38.59 mg percent among affected birds while the normal birds showed uric acid levels as 4.72 mg per cent with range of 2.95 to 6.35mg percent.

Pathak (2001) reported that serum uric acid level in gout affected birds was observed 44mg per 100 ml of blood and in normal birds 5.7 mg per 100 ml of blood.

Sayed (2001) observed that uric acid level in normal birds range between 2 to 15 mg per 100 ml of blood and more than 20 mg per 100 ml of blood resulted in gout.

Hassanerin (2004) recorded the increased level of Hb TEC, PCV in natural case of gout. Globulin part of protein was found to be increased.

Kraljevi *et al.* (2008) investigated the enzymatic profile in blood plasma that can help in the diagnosis of organic or functional liver damages caused by gamma ray in chickens and evaluated the activity of Aspartate Amino-Transferase (AST), Alanine Amino-Transferase (ALT), Gamma-Glutamyl Transpeptidase (GGT), Leucine Aminopeptidase (LAP), Lactate Dehydrogenase (LDH) and α-Hydroxy-Butyrate Dehydrogenase (HBDH) in blood plasma and observed that throughout the experimental period, only GGT activity did not changed statistically significantly.

Dhara *et al.* (2010) recorded therapeutic experimental study on 120 sick broiler birds of two weeks affected with visceral gout and observed that mean serum uric acid level at 0 day of treatment was significantly increased (35.2±12.28) in birds of all the groups compared

to healthy birds (7.58±0.73) and recognized it to failure of excretory function of kidney of gout affected birds caused by several factors.

Feizi *et al.* (2012) assessed some biochemical factors in broiler breeder with goutsyndrome and observed significantly increased (P<0.001) uric acid level in the affected farms (8.35±0.28 mg/dl) compared to non-affected farms (5.40±0.14 mg/dl).

Feizi *et al.* (2012) investigated statistically significant (P<0.001) elevated level of serum creatinine in affected group (1.06±0.02 mg/dl) compared to control group (0.41±0.01 mg/dl) in a gout impacted broiler breeder farm.

Singh *et al.* (2013) observed in gout positive birds significantly higher level of uric acid. There was significant difference revealed in activity of ALT, AST, ALP, in gout affected birds. There were no significant changes observed in total serum protein. and serum albumin level was found to be significantly higher and globulin value lower significant in gout affected birds.

Pathology of visceral gout

Gross pathology

Jull (1938) noticed the first time gross lesions of visceral gout in poultry. He observed lesions like paleness and enlargement of kidneys and often coated with whitish sugar like material. Similar whitish depositions were also found on the lining of abdomen and the surface of liver.

Iyer (1941) reported that the birds with gout brought for autopsy were often very light with sunken eyes and also observed lesions characterized by chalky deposits in the thorax and abdomen, pericardium, liver, lungs, kidney, spleen and entire surface of intestine including mesentery and genital organ.

Runnells *et al.* (1965) noticed that gout affected poultry birds emaciated as well as dehydrated carcasses and also observed urate deposits in kidney and ureters. At time urate were so abundant that kidney appeared frosted, renal lesions, and urate crystals were found on pericardium epicardium and peritoneum.

Bokori (1966) reported gout in poultry and detected the gross lesions of white chalky deposition on different visceral organs.

Lathkar and Rajya (1968) noticed in detail the gross and morphological change in the kidneys and liver of gouty birds. Gross changes included enlarged red colored kidney, urate filled dilated ureters, swollen liver with urate deposition and deposition of urate on all the visceral organs

Nath and Singh (1972) studied the pathology of disorder of urogenital tract in poultry and found that out of 206 kidneys studied, five showed the lesion of visceral gout, the incidence being 2.43% among different kidney lesions.

Rahamathulla and Mohiyuddeen (1973) noticed that morphological changes observable in kidney and liver of gouty birds. Grossly, kidneys were swollen along with the deposition of dirty white uric acid crystals. The ureters were distended with creamy white material. Liver were congested, enlarged and friable.

Chandra and Singh (1980) reported gross lesion of uric acid nephritis in birds. They found chalky white deposition and enlargement of kidney with urate deposit in other visceral organ and body cavity in majority of the cases. In some cases the kidney of one side was atrophied with compensatory hypertrophy of the other side.

Pegram and Wyatt (1982) observed enlargement with green discoloration of gizzard in birds affected with gout.

Reddy *et al.* (1984) observed lesions such as deposition of urate on air sacs and paleness of kidney in birds died due to visceral gout.

Chaudhari (1988) investigated deposition of urate on heart, kidney, and liver. Ureters were packed with urate crystal and kidney was hypertrophied and urate deposition also observed in trachea, joint cavity and lung.

Sah *et al.* (1988) reported bursal atrophy along with the severe nephrotic lesion in few outbreaks of IBD. He was observed urate deposition on kidney, kidney enlarged, hard and brittle. The ureter were greatly enlarged and distended by pale yellow gritty materials. He was also observed white chalky urate deposition on pericardium, epicardium, and serosal surface of organs and articular surface particularly the hock joint.

Uma *et al.* (1999) studied pathology of gout in broilers and found that gouty birds appeared emaciated, dehydrated with reddish musculature visceral organs showed deposition of white urate crystals.

Sayed *et al.* (2001) described lesion of visceral gout as white urate deposits over hearts, liver, proventricular, spleen, articular surface of joints, dilated ureters and atrophy of the affected kidney with enlargement of surviving lobes.

Yewale (2010) revealed extent and severity variation in different flocks as well as in different birds. The main organs, which showed striking gross lesions, were kidney, heart, liver, proventriculus, intestine, gizzard, lungs, spleen, air sacs and joints.

Feizi *et al.* (2012) reported that inclusion of 2 per cent calcium in broilers' diet lead to some disorders in the physiologic process of kidneys consequently resulting in urates depositions on serous surfaces of viscera especially kidneys which are the symptoms of gout syndrome that makes vast mortality in flock.,

Patel *et al.* (2014) Aceclofenac administration at the dose rate of greater than 20 ppm induces toxicity symptom in layers. Pathomorphological lesions induced by aceclofenac toxicity were vascular and degenerative in nature mainly affecting kidney, liver, heart, proventricular, gizzard and lung. Were specific of visceral gout. Aceclofenac administration on at and above 20 ppm for 21 days induces visceral gout in layer chicks.

Auda (2013) observed prominent change on the serosal surface of liver, heart, kidney and ureters. The pericardium, liver and air sac appeared to be dusted with a pale yellow powder. Both kidneys swelled enlarged. With numerous white nodule on the external and sectioned surfaces. The ureters were dilated in whole length. They contained a small or large quantity of whitish fluid or muddy substance.

Mudasir *et al.* (2017) reported postmortem examination revealed a white chalk material deposit on spleen, liver, pericardial sac, and kidney, also observed enlarged kidney with necrotic foci and swelling of ureter in some birds.

Satthiyaseelan *et al.* (2018) recorded in 24/120 (20%) cases and it was characterized by swollen pale kidneys with urate deposits and dilated ureters white material. The other

noticeable findings observed were presence of chalky white masses surrounding the heart, kidneys, liver and other organs.

Histopathological changes

Jungher and Levine (1940) noticed that gouty nephritis featured by necrosis of liver and chalkiness of visceral organs in broilers.

Chandrashekaran *et al.* (1957) also described similar lesions in certain outbreaks of gout.

Lathkar and Rajya (1968) noticed that revealed vascular and degenerative changes along with urate deposition accompanied with inflammatory changes. The changes observed by them in liver consisted of deposition of urate, necrotic foci and infiltration of Heterophils.

Barron (1966) described that gouty nephritis in cases of Gumboro's disease. The kidneys were swollen due to marked urate deposition in the tubules along with degenerative changes. Sinusoidal congestion along with mild fatty changes were the additional features noticed.

Rahamathulla and Mohiyuddeen (1973) noticed that degeneration of kidney tubules and uric acid tophi in interstinum as well as inside the tubules. Infiltrating cells were predominated by heterophils. Liver showed multifocal areas of necrosis, urate deposition and infiltration of heterophils and lymphocyte.

Reddy *et al.* (1984) observed degenerative changes in lower part of renal tubules along with deposition of crystals of uric acid and urates from cases of visceral gout in poultry.

Chandra *et al.* (1988) recorded microscopic lesion on heart, liver and kidney in nephritis in poultry. He was observed in liver vacuolar and granular degeneration of hepatocyte. Sinusoids were congested and necrotic foci around the central. Kidney revealed infiltration with lymphocyte and heterophils and large mononuclear cell into perivascular, pericapsular and interstitial space.

Chaudhary (1988) discovered that needle like urate deposition on heart, liver and kidney along with infiltration of mononuclear cells and heterophils.

Sah *et al*. (1988) reported microscopic lesion of kidney in visceral and articular gout associated with IBD virus infection kidney tubules showed pink homogenous and heterothallic casts. Group of kidney tubules at places were necrosed. Multiple area of radiating uric acid crystal were surrounded by lymphocyte. Macrophages and giant cells. Interstitial tissue showed hemorrhage.

Nayak *et al.* (1988); Rao *et al.* (1993) reported microscopic changes of liver and kidney in gout. Liver revealed massive deposition of urate crystal in the sinusoid and central veins in the form of fine meshes of network and irregular sponges. The Glisson's capsules at different zone showed marked thickening with proliferating fibrous tissues and infiltration of heterophils and lymphocyte at different zone of liver. The kidney revealed complete destruction of both glomeruli and tubular structure with deposition of urate crystal in the form of spongy balls of variable sizes.

Zaragoza *et al*. (1995) reported NSAID induced hepatitis by aceclofenac and diclofenac. The liver sections showed peri-portal fibrosis, mild hydropic degeneration, and focal aggregation of lymphocytes.

Uma *et al.* (1997) reported congestion, hemorrhage, edema, and deposition of urate of varying degree along with infiltration of heterophils in heart. In kidney, urate depositions were seen in the glomeruli, tubular lumen and interstitial space along with infiltration of inflammatory cell around the urate deposits.

Gupta *et al.* (2002) noticed visceral gout where kidney revealed hypertrophy and hyperplasia of the cell of the glomeruli, dilation of the tubules, tubular casts. Necrosis of the tubular epithelium interstitial, fibroplasia and presence of numerous urate crystal of variable in the tubules. Liver revealed congestion.

Gajera (2006) studied variable degrees of congestion, mild to severe haemorrhages and deposition of urates in form of amorphous material or radiating crystalline rosette pattern in kidney, liver, heart and lungs. He observed tubular degeneration, cystic dilatation of renal tubules, proteinaceous tubular casts, deposits of urates in the tubular parenchyma and occasional infiltration of heterophils and mononuclear cells in kidneys. The urate deposits gave positive reaction with De-Galantha's stain which appeared as black needle shaped crystals. The lesions in the pericardium consisted of variable degree of congestion, haemorrhages, edema and thick masses of deposition of urates as homogenous pinkish

amorphous material. Hepatic parenchyma showed vascular congestion, haemorrhages, focal deposition of urates in rosette form which in H&E sections seen as radiating clear spaces. Some sections showed focal infiltration of mononuclear cells in the hepatic parenchyma.

Seema (2008) observed variable degrees of vascular congestion, mild to severe hemorrhages, necrosis, degeneration and deposition of uric acid crystals either in the form of homogeneous amorphous material or assuming radiating crystalline pattern (tophi) in different organs (kidney, liver, spleen, heart and lung).

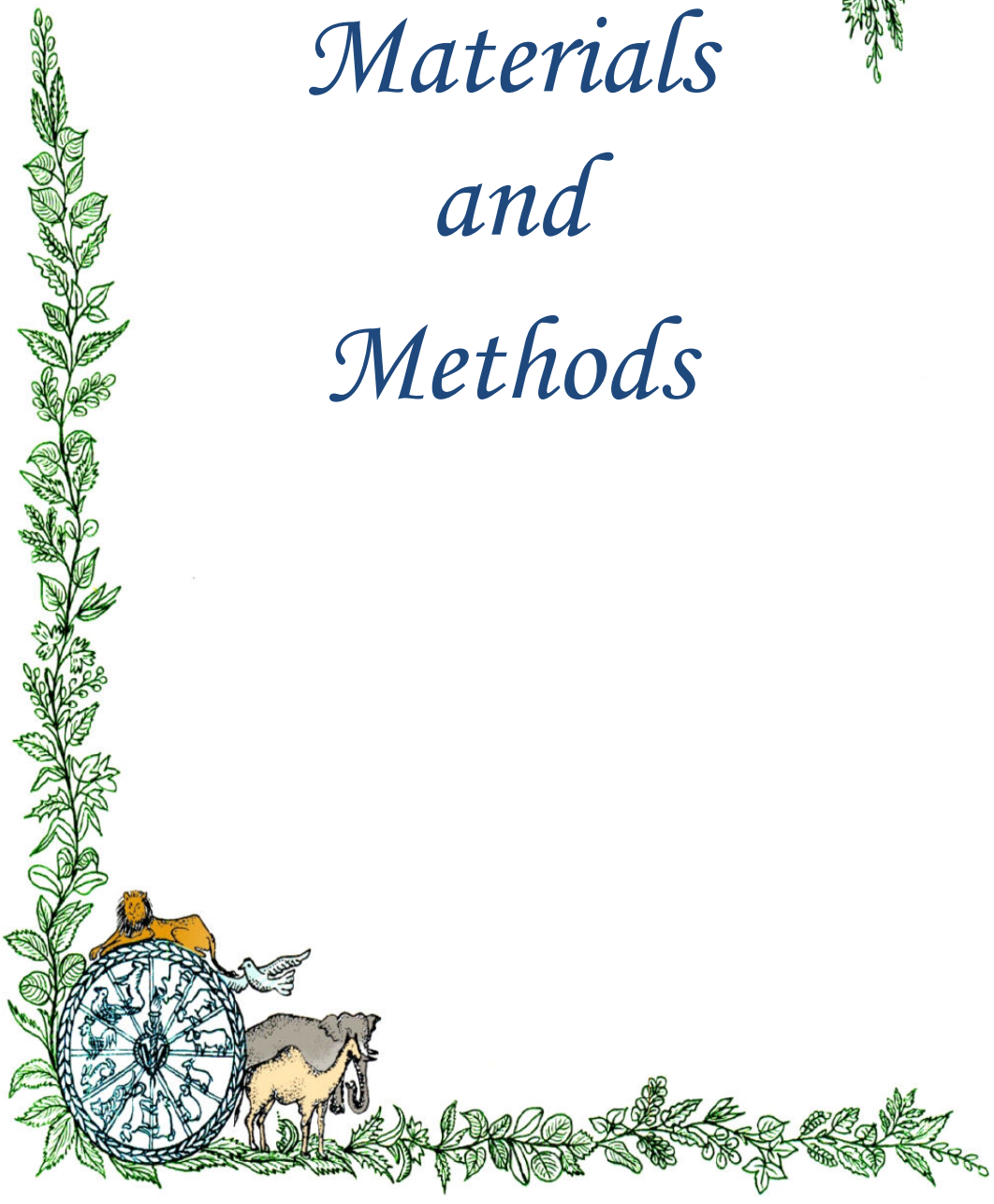
Hedaoo *et al.* (2008) microscopically found hemorrhages, focal necrosis, granular degeneration and infiltration of inflammatory cells in liver. In kidneys degenerative changes in tubular epithelium, hemorrhages in glomerular tuft, increased cellularity in glomeruli and infiltration of inflammatory cells in inertial space.

Auda (2013) observed multiple small, focal lesion on myocardium. The lesion consisted of a central zone of radiating sheaves of eosinophilic crystal like material. This zone surrounded by mononuclear and multinuclear macrophage.

Akhter and Sarker (2015) observed kidney section of birds of Group C, A, B showed tubular degeneration, urate deposition present in kidney tubules. The section of birds of group C, B, A which succumbed to toxicity showed tubular degeneration and varying sized foci of urate deposition (tophi) either in the form of amorphous material or in the radiating crystalline pattern mixed with necrotic debris due to degeneration of surrounding cells.

Mudasir *et al.,*(2017) noticed severe damage of tubules characterized by moderate to severe tubule dilation and necrosis and bluish gouty deposition surrounded by radiation of needle like urate crystal followed by granulomatous reaction of lymphocytes, macrophage, and fibroblasts with hemorrhage. The collecting renal tubules, Uri nephric ducts and ureters were also filled by gouty deposits with epithelial hyperplastic change and chronic inflammatory cellular reaction in their wall. The interstitial tissue showed edema. Congestion, hemorrhage and lympho mononuclear cell infiltration. Glomerular changes included thickening of Bowman's capsule and proliferative glomerulonephritis.

Materials and Methods



3 Materials and methods

The present experiment on ‘‘Clinico-Pathological studies and therapeutic effect of Febuxostat on visceral gout in Vanaraja Chicken” was conducted ILFC, BVC, Patna and Department of Veterinary Pathology, College of Bihar Veterinary College, Bihar Animal Sciences University, Patna.

Test Chemicals for experiment

Febuxostat

Febuxostat was procured from the market and was used drug for its therapeutic effect to determine visceral gout on the experimentally induced in Vanaraja Chicken.Febuxostat is tablet form (febustat 40, Abbott Healthcare Pvt.Ltd) having 40 mg/kg tablet.

Compound name	**Febuxostat**
Batch Number	FSA 9002
Company	Abbott
Molecular weight	316.38
Color	Titanium dioxide I.P
Store	Store at temperature not exceeding 30ºC

Aceclofenac

Aceclofenac was procured from the market and was used drug for experimentally induced toxicity in vanaraja chickens. Aceclofenac in tablet form (Zerodo 1100, Ipca laboratories Ltd. Mumbai) having 100 mg active drug. Tablet was crushed to fine powder and mixed in the mash feed in graded level.

Compound name	Aceclofenac
Batch Number	GGGO19002AS
Company	IPCA
Molecular weight	354.19
Molecular formula	$C_6H13CL2NO4$
Color	Ferric oxide yellow USP—NF &Titanium dioxide I.P
Store	Dry place, protected from light and moisture.

Bottle gourd juice

It is traditional systems of medicine have always played important role in meeting global healthcare needs.

Raw Bottle gourd was brought from market and washed under tap water. Grinding of raw bottle gourd juice was done in grinder to obtained raw bottle gourd juice in transferred into beaker. The obtained juice was left over for 24 hours in beaker. Next day supernatant was obtained into another beaker. And filtered through sieve to obtained raw clear extract. Raw clear extract used for treatment.

Experimental birds

Study was conducted on day old vanaraja chicks. Total seventy two vanaraja chicks procured from ILFC BVC, Patna and were maintained under standard managemental condition.

Housing and Management

The experimental procedure was reviewed and approved by Institutional Animal Ethics committee. Vanaraja chicks were housed in the experimental unit, ILFC, BVC, Bihar Animal Sciences University, Patna and weighted weekly during the entire period of study. Environmental temperature and lighting regimens were applied according to the vanaraja

chicken guidelines. Birds were observed daily for clinical signs, any physical or behavioral changes, body weight and mortality, and throughout the experimental.

Feeding management

All the birds received balance layer pre- starter and starter feed in mash form. Procured from ILFC, BVC, and Patna. The composition of layer pre-starter and starter feed in in mash form was as per standard guidelines. The feed offered *ad libitum* and water was freely available at all times throughout the study.

Design of experiment

Present Study was conducted on total no of seventy two (72), Vanaraja chickens, Birds were divided into four groups, and each group contain 18 chicks. Experimental trial was started after completions of one week observation period. The experimental study was conducted in 7 day old vanaraja chicks to know the effect of feeding aceclofenac drug with the special interest in causation of visceral gout and as well as role of Febuxostat for treatment of visceral gout. Groups were numbered I to IV. Group I was kept as control group and fed only mash feed. The producing group II and treatment group IV were given aceclofenac dose rate of 20 mg/kg respectively through feed every day for 28 days. The preventive group III was given aceclofenac 20 mg/kg and Febuxostat 4mg through feed every day for 28 days. After 28 days group II was treated with bottle gourd juice 20ml and group IV treated with Febuxostat 4 mg/kg. All the birds were maintained for 42 days and blood samples was collected on 7, 14, 21 and 28^{th} day. Weighing of chicks was carried out at day one and at the everyday for 42 days. Mortality and body weight were recorded throughout the period of experiment. Postmortem examination of birds died during experiment was conducted with the recording of gross lesion and collection of tissues.

After completion of treatment period that is on 42 days of the experiment , the surviving birds from all IV groups were sacrificed by cervical dislocation after collection of blood for hematology and biochemical profile analysis, the postmortem examination were conducted for histopathological observation and gross lesion were recorded. Tissue sample from organs like kidney, liver, heart, lung, spleen, and intestine were collected in 10% formalin for histopathological.

DESIGN OF EXPERIMENT

S.No.	Groups name	No. of Birds	Groups/treatment
Group I	Control group	18	Untreated control group fed normal starter feed for 42 days
Group II	Producing group	18	Fed Normal starter feed with aceclofenac @ 20mg/kg feed for experimental induce gout for 28 days, treated with bottle gourd juice 20ml through drinking water.
Group III	Preventive group	18	Fed Normal starter feed with Aceclofenac @20mg/kg +Febuxostat4mg/kg
Group IV	Treatment group	18	Fed Normal starter feed with aceclofenac @ 20mg/kg for experimental induce gout for 28 days, treated with Febuxostat 4mg/kg B.wt.

Collection of blood

The birds were maintained for 42 days and blood sample was collected on 7, 14, 21 and 28, 42day. Blood was collected from wing vein. About 2 ml of blood was collected in plain container and kept in slant position for separation of serum samples were store in the deep freeze at -20ºC for further biochemical estimation.

All birds belonging to group I to IV were observed daily for any mortality throughout the period of 42 days of experiment.

Body weight

Body weight of chicks of all the 4 groups were taken initially on day 0 and then 7, 14, 21 and 28 and 42 days of the experiment study.

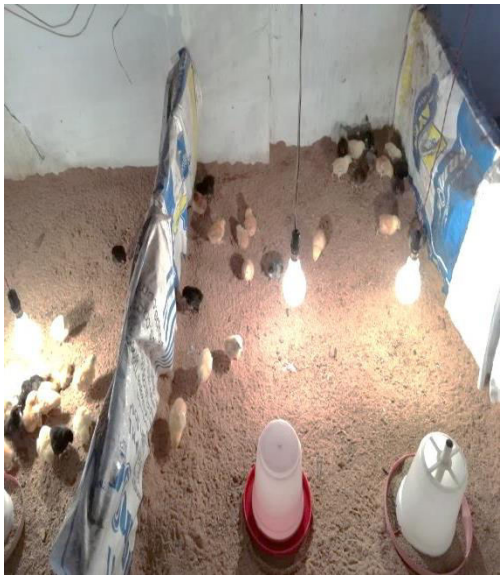


Fig.1. Grouping of Vanaraja chicks at the end of 7 day of experiment0

Fig. 2. Control group of Vanaraja chickens after the completion of 21 days of experiment

Fig. 3. collection of blood from wing vein of vanaraja chickens during experimental period

Fig. 3. Collection of blood from wing vein of vanaraja chickens during experimental period

Mortality

Mortality in different group were recorded throughout the experiment period.

Haematological estimation

Blood sample was collected on 7, 14, 21 and 28, 42 day. Blood was collected from wing vein into plain container containing K_3EDTA (Tripotassium Ethylene DiamineTetraaciticacid) for the estimation of various haematological parameters. The following parameters were carried out.

Hemoglobin (Hb)

Hemoglobin was estimated by Cyanomethemoglobin method using Spectrophotometer.

Total leukocyte count (TLC)

TLC was done with the help of Haemocytometer in Neubauer's counting chamber using Nett and Herrick's fluid as diluting fluid and the results were expressed in thousand per cubic mm.

Total erythrocyte count (TEC)

TEC was done with the help of Haemocytometer in Neubauer's counting chamber using Nett and Herrick's fluid as diluting fluid and the results were expressed in millions per cubic mm.

Packed cell volume (PCV)

PCV was estimated by micro haematocrit technique as described by Jain (1986) and the volume was expressed in volume percent (%) with slight modification.

Different leukocyte count

Freshly prepared blood smears were prepared and stain with Wright's stain (Lucas and Jamroz, 1974) percent of different leucocyte were determined by examining the smear under the oil immersion of light microscope.

Biochemical investigation

Blood was collected in vacutainer and kept in slant position for separation of serum samples were store in the deep freeze at -20ºC for further biochemical estimation. The following biochemical estimation were semi auto analyzer.

1. **Total serum protein level (g/dl)**
2. **Serum Albumin (g/dl)**
3. **Globulin (g/dl)**
4. **Alkaline phosphatase (µ/L)**
5. **Serum Uric acid level (mg/dl)**
6. **Serum Creatinine level (mg/dl)**
7. **Serum glutamic pyruvic transaminase (SGPT) (U/L)**
8. **Serum glutamic oxaloacetic transaminase (SGOT) (U/L)**

Serum uric acid level

For the purpose of serum uric acid analysis, uric acid estimation kits (uricase/PAP method) manufactured by coral were used.

Serum Albumin (g/dl)

For the purpose of serum albumin analysis, serum albumin estimation kits (BCG method) manufactured by crest bio systems company (division of Coral system Pvt, Ltd.) were used.

Alkaline phosphatase (ALP)

For the purpose of alkaline phosphatase analysis, alkaline phosphatase estimation kits (pNPP kinetic method), manufactured by crest biosystems company (division of Coral Systems Pvt, Ltd.) were used.

Serum creatinine level

For the purpose of serum creatinine analysis, serum creatinine estimation kits (Modified jaffe's kinetic method) manufactured by crest bio system company (a division of Coral Systems Pvt, Ltd) were used. The method of estimation of serum creatinine was fixed kinetic type.

Serum glutamic pyruvic transaminase (SGPT)

For the purpose of Serum glutamic pyruvic transaminase estimation, SGPT kits (Modified IFCC method) manufactured by Coral clinical systems (a division of Tulip Diagnostics (P) Ltd.) were used. The method of estimation of SGPT was modified IFCC type. It is found in a variety of tissues but is mainly found in the liver.

Serum glutamic oxalo-acetic transaminase (SGOT)

For the purpose of Serum glutamic oxaloacetic transaminase estimation, SGOT (AST) kits (Modified IFCC method) manufactured by Coral Clinical System (a division of Tulip Diagnostics (P) Ltd.) were used.

Gross lesion and histopathology

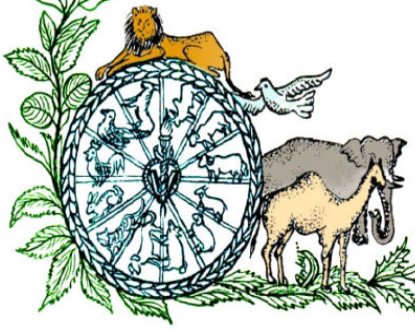
After completion of treatment period that is on 42 days of the experiment, the surviving birds from all groups, group IV were sacrificed by cervical dislocation. A detailed post mortem was performed. The gross pathological lesions if any present were recorded in individual birds from all groups i.e. from the dead as well as sacrificed birds.

For histopathological examination, tissue including kidney, liver, spleen, heart, and lung were collected in 10% formalin for histopathology and process by paraffin embedding technique. Section were cut 4-5 microns thickness with automatic section cutting microtome machine and stained with Haematoxyline and Eosin (H&E) stains (Luna 1968) .and stained slide were observed under microscope and lesion were recorded. Furthermore, heart, liver, kidney tissues were fixed in absolute alcohol and were directly cleared in xylene and embedded in paraffin. Sections were cut 8 microns thickness. Tissues were stained using De Galantha's special staining method (Luna 1968) for demonstration of urate crystal.

Data Analysis

The data obtained from mortality, body weight, hematological and biochemical profile were subjected to analysis by using standard procedures described by Snedecor and Cochran (1980). The data analysis was carried out by SPSS (Statistical package for the social science) software version 23. The One way ANOVA were used for showing significance between different groups.

Results & Discussion



4 Results and Discussion

The present study was carried out to know the Clinico pathological studies and therapeutic effect of Febuxostat, and bottle gourd to the experimentally induce aceclofenac drug (@ 20 mg/kg) in vanaraja chicken. To undertake these objectives pathological, serum biochemical and hematological were carried out in vanaraja chickens.

Clinical signs

All the vanaraja chickens belonging to group I, group II, group III and group IV were observed every day for any abnormal physical changes throughout the period of 42 days of experiment.

Vanaraja chickens under Control Group I seemed clinically healthy and did not observed typical clinical signs during all the experimental period. Vanaraja chickens kept under control group I and only fed normal starter feed and libitum water. vanaraja chickens in the aceclofenac Group II and Group IV showed typical clinical signs like dehydrated, lethargy, emaciated, anorexia, , drooling of the wings, dullness,Birds often have reddened, swollen feet that progress to blisters and sores.Shifting leg lameness, vent moist and greenish diarrhea.

The aceclofenac and febuxostat was administered in Group III did not shown any clinical signs and appeared healthy. visceral gout affected vanaraja chickens in group II treated with bottle gourd did not observed clinical signs and appear healthy, visceral gout affected vanaraja chickens in group IV treated with febuxostat did not observed clinical signs and appear healthy

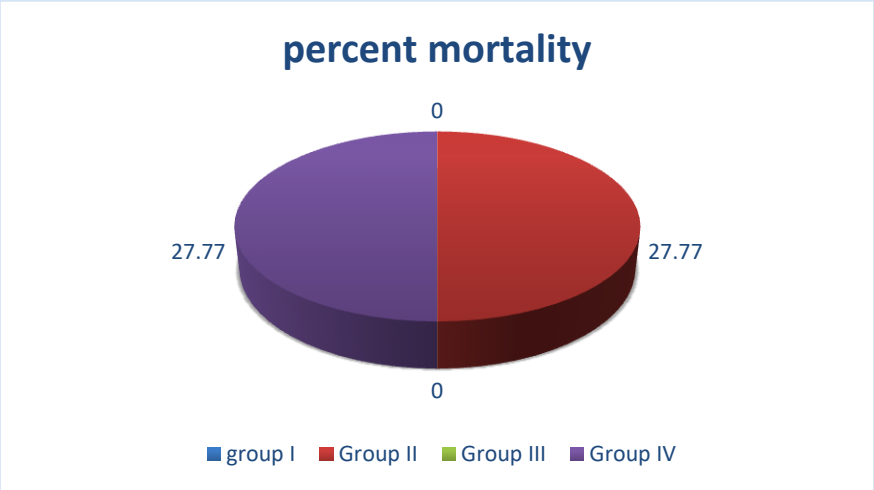
The clinical signs in aceclofenac toxicity during experiment were in agreement with findings of Patel et al., (2014).

Similar clinical signs were recorded by Seema (2008), Naidoo *et al.,* 2007, Tamta *et al*. (2009), Jana *et al.* (2009), Darbar *et al.* 2010 and Ghodasara *et al.* (2014).

In present study clinical signs like dehydration, dullness, emaciated, weakness in aceclofenac administered vanaraja chickens of Group II and Group IV may be endorsed to

Table.1 Mortality (%) in different intervals of different groups in vanaraja chickens

Days	Experimental group			
	Group I	Group II	Group III	Group IV
7	-	-	-	-
14	-	2	-	3
21	-	1	-	1
28	-	2	-	1
42	-	-	-	-
N= No. of birds	18	18	18	18
Total deaths	-	5	-	5
Mortality %	-	27.77	-	27.77



the damage caused by aceclofenac to kidney tissue resulted into impaired kidney function. Injured kidney impaired uric acid excretion resulting to hyperuricaemia and deposition of urate crystals in visceral organs. The aceclofenac and febuxostat was administered in Group III did not shown any clinical signs and appeared healthy. The present study revealed that bottle gourd and febuxostat efficiently reduce the urate crystal formation and repair the kidney function and kidney damage.

MORTALITY

As shown in table 1 and chart 1 there was no mortality in group Iand group III whereas in group II and group IV variable mortality was observed. Mortality was observed in aceclofenac induced group II (22.77%) and followed by group IV (22.77%). In group II and group IV mortality started from day 14 and was continued up to 28 days after aceclofenac induced. All died birds showed lesions of visceral gout.

Similar findings also reported by Irtaza *et al.* (2008), Seema (2008), Shinde (2008), Jain *et al.* (2009), Darbar *et al.* (2010) and Sehgal *et al.* (2011).

BODY WEIGHT

The average body weight for each group of vanaraja chickens was recorded weekly have been shown in Table 2 and chart 2

There was no observed any significant changes between control group I and preventive group III at the end of 7, 14, 21, 28 and 42 days. Decreased body weight was observed in aceclofenac administered group II and group IV as compared with control group at the end of 7, 14, 21, and 28days. Reduction in body weight were observed in bottle gourd treated group II and febuxostat treated group IV comparison with the control at the end of 42 days.

Decreased body weight revealed in the present experiment indicated that Aceclofenac dose @ 20mg/kg in feed can induce visceral gout in vanaraja chickens, its toxic effect by decreased in body weight.

In present study increased value of uric acid in group II and Group IV at end of 14 days, 21 days 28 days during the present study revealed that aceclofenac negatively affect kidney functions which was outward terms of serum uric acid level similar finding also

Table 2: Mean ± S.E. values of Body weight (grams) of Vanaraja chickens in different groups at various time intervals

Groups	7 day	14 day	21 day	28 day	42 day
Group I	87.83±2.35[a]	150.66±2.04[a]	244.83±3.40[a]	345.83±4.49[a]	1053.5±14.79[a]
Group II	90.66±1.62[a]	111.16±5.26[b]	115.83±4.86[b]	119.83±4.71[b]	344.5 ± 3.35[b]
Group III	88.16±1.74[a]	153.33±2.81[a]	250.16±3.55[a]	345.66±3.67[a]	1067.84±21.71[a]
Group IV	89.33±1.48[a]	104.33±1.33[b]	108.83±1.30[b]	113.83±1.78[b]	343.33 ± 2.43[b]

Superscript a, b, c, d in column indicates significant difference between the groups within days

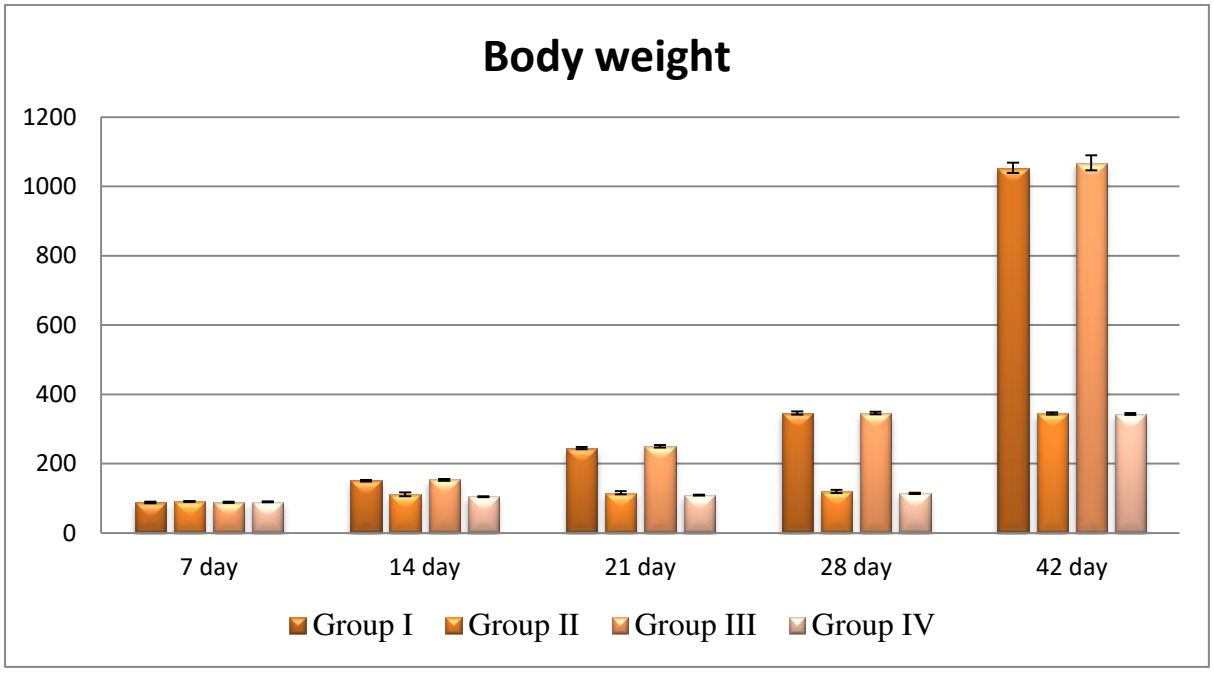


Fig. 2: Histogram showing Mean± S.E. of Body weight (grams) of Vanaraja chickens in different groups at various time intervals

reported by Seema (2006), Irtaza *et al.* (2008), Shinde (2008), Darbar *et al.* (2010), Sehgal *et al.* (2011).

In present experimental study aceclofenac induced group II treated with bottle gourd juice, it is observed that bottle gourd juice have good effect on aceclofenac induced visceral gout produced in vanaraja chickens. Experimental study on vanaraja birds shown that significant decreased body weight level after the end of 28 days to 42 days treated with bottle gourd juice. Group II showed significant slightly increased body weight .It was apparent that bottle gourd juices have protective effect on aceclofenac induced visceral gout in vanaraja chicken. In the absence of available literature the value of bottle gourd further indicate that bottle gourd juice slightly increased body weight concentration which was decreased due to aceclofenac toxicity.

In present study aceclofenac induce group IV treated with febuxostat. Experimental study in vanaraja birds showed that significant slightly increased level after the end of 28 days and 42 treated with Febuxostat. It was evident that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja birds.

Hematological parameters

Hemoglobin

Hemoglobin is an important diagnostic tool to determine Dehydration, (increased) anemia, polycythemia (increased), poor nutrition, the mean levels of Hb in different experimental groups are presented in .Table 3, Chart 3.

The Mean ± S.E value of Hb (g/dl) of vanaraja chickens of Group I, II, III and IV at the end of 7 day were 7.70 ± 0.063, 7.58 ± 0.20, 7.60 ± 0.20and 7.49 ± 0.33 respectively, No significant differences were observed in all groups at the end 7 days.

The Mean ± S.E value of Hb (g/dl) of vanaraja chicks of Group I, II, III and IV at the end of 14 days were 7.63 ± 0.12, 13.81 ± 0.22, 7.53 ± 0.12 and 13.86 ± 0.10 respectively, Mean Hb value of aceclofenac induced vanaraja chicks belong to Group II and IV showed significantly higher as compared to control Group I at the end of 14 days. The vanaraja chicks of induced with Aceclofenac + Febuxostat (Preventive Group) Group III not revealed any significant change mean value of Hb as compared to control Group I at the end of 14 days.

Table 3: Mean±S.E. values of Haemoglobin (Hb) (g/dl) of Vanaraja chickens in different groups at various time intervals

Groups	7 day	14 day	21day	28 day	42day
Group I	7.70±0.063 [a]	7.63± 0.12[a]	7.56 ± 0.20[a]	7.66 ± 0.13[a]	7.56±0 .20 [a]
Group II	7.58 ± 0.20[a]	13.81± 0.22 [b]	14.75± 0.11[b]	15.33± 0.14[b]	7.65 ± 0.15[a]
Group III	7.60 ± 0.20[a]	7.53± 0.12[a]	7.50 ± 0.20 [a]	7.43 ± .071[a]	7.66 ± 0.18 [a]
Group IV	7.49 ± 0.33[a]	13.86 ±0.10[b]	14.90 ±0.21[b]	15.48 ± 0.15[b]	7.63± 0. 17[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

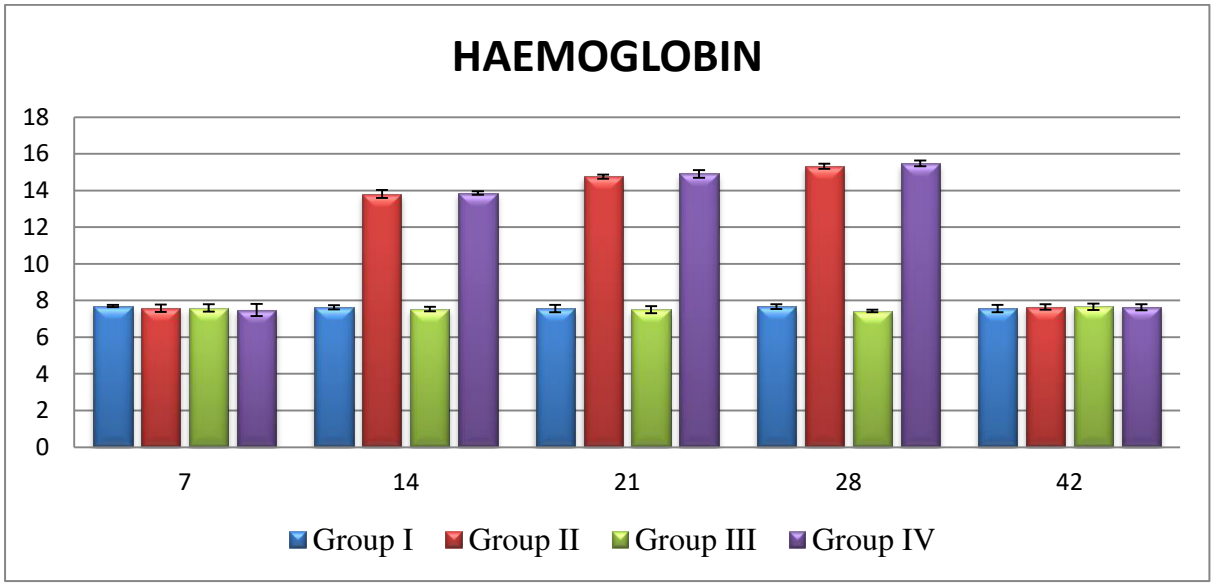


Fig. 3: Histogram showing Mean ± S.E. value of Haemoglobin (Hb) (g/dl) of Vanaraja chickens in different groups at various time intervals

The Mean ± S.E value of Hb (g/dl) of Vanaraja chickens of Group I, II, III and IV at the end of 21 days were 7.56 ± 0.20, 14.75 ± 0.11, 7.50 ± 0.20 and 14.90 ± 0.21 respectively, Mean Hb value of aceclofenac induce in vanaraja chicken i.e. Group II and IV showed significantly higher as compared to control Group I.at the end of 21 days. Mean Hb value of aceclofenac + febuxostat induced in vanaraja chicken i.e. Group III (Preventive Group) not revealed any significant changes at the end of 21 days.

The Mean ± S.E value of Hb (g/dl) of vanaraja chickens of Group I, II, III and IV at the end of 28 days were 7.66 ± 0.13, 15.33 ± 0.14, 7.43 ± 0.71, and 15.48 ± 0.15 respectively, Mean Hb value of aceclofenac induce groups i.e. Group II and IV showed significantly higher as compared to Group I at end of 28 days. The vanaraja chickens of induced with aceclofenac + febuxostat (Preventive Group) Group III not revealed any significant change mean value of Hb as compared to control Group I at the end of 28 days.

The Mean ± S.E value of Hb (g/dl) of vanaraja chickens of Group I, II, III and IV at the end of 42 days were 7.56 ± 0.20, 7.65 ± 0.15, 7.66 ± 0.18, 7.66 ± 0.18 and 7.63 ± 0. 17 respectively, Gout affected vanaraja chickens belong to group II treated with bottle gourd juice and Group II treated with Febuxostat drug revealed no significant changes in mean value of Hb compared to control Group I and Group III at the end of 42 days.

Packed cell volume (PCV)

Packed cell volume is an important diagnostic tool to know the determine of Dehydration, (increased) anemia (decreased), polycythemia (increased), poor nutrition, the mean levels of PCV in different experimental groups are presented in .Table 4, Chart 4.

The Mean ± S.E. value of PCV of Vanaraja chicks of Group I, II, III and IV at the end of 7days were 22.50 ± 0.78, 22.13 ± 0.84, 22.90 ± 1.03, and 22.15±0.42respectively. No significant differences were observed in all groups at the end of 7 days.

The Mean and S.E value of PCV of Vanaraja chicks of Group I, II, III and IV at the end of 14 week were 22.35 ± 4.63, 37.26 ± 0.22, 24.48 ± 1.05, and 37.34 ± 0.55 respectively. Aceclofenac induced Groups i.e. Group II and IV revealed comparatively higher PCV than Group I and III. No significant difference in PCV was observed between Group I and III. The vanaraja chicks of induced with Aceclofenac + Febuxostat Group III not revealed any significant change mean value of PCV as compared to control Group I at the end of 14 days.

Table 4: Mean±S.E. values of Packed cell Volume (PCV) (%) of Vanaraja chickens in different groups at various time intervals

Groups	7 day	14 day	21day	28 day	42day
Group I	22.50 ± 0.78[a]	22.35 ± 4.63[a]	25.96 ± 1.51[a]	25.71 ± 1.33[a]	24.87 ± 1.51[a]
Group II	22.13 ± 0.84[a]	37.26 ±0.22[b]	39.13 ±0.25[b]	41.79 ± 0.25[b]	24.38 ± 0.60[a]
Group III	22.90 ± 1.03[a]	24.48 ±1.05[a]	24.90 ± 2.98[a]	26.36 ± 2.11[a]	25.15 ± 0.96 [a]
Group IV	22.15±0.42[a]	37.34 ±0.55[b]	39.04 ± 0.46[b]	41.75 ± 4.21[b]	25.81 ± 1.33 [a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

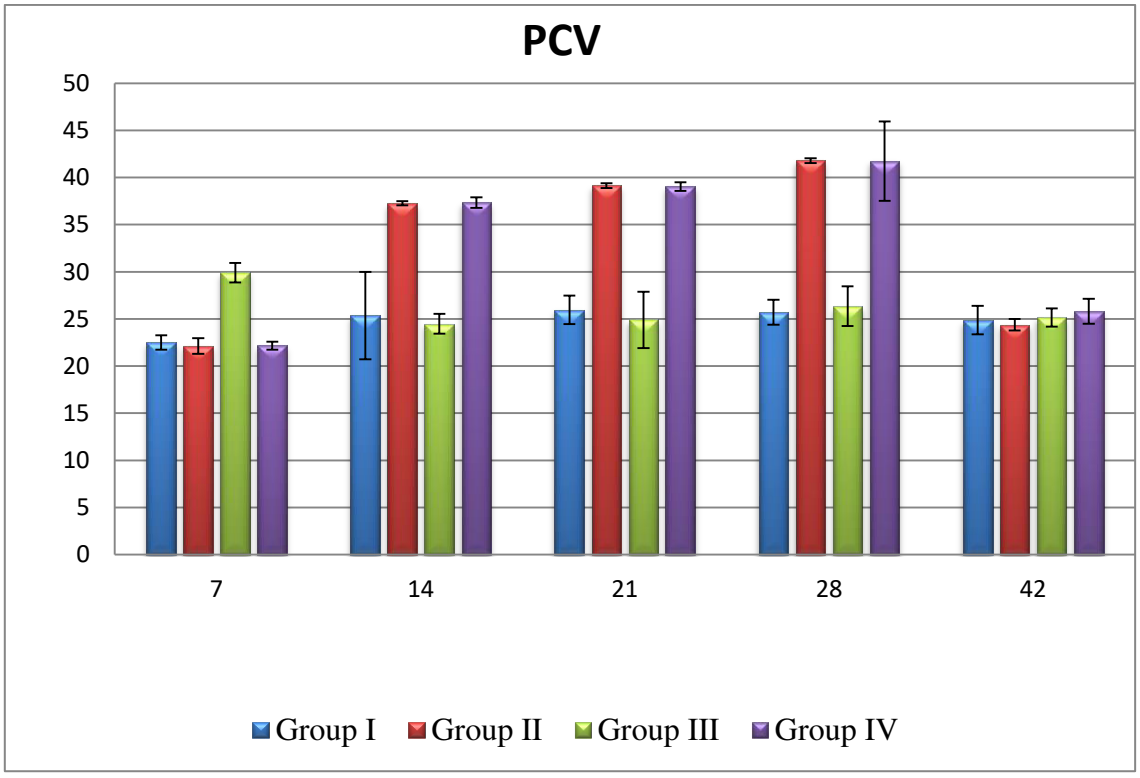


Fig. 4: Histogram showing Mean ± S.E. of PCV (%) Vanaraja chickens in different groups at various time intervals

The Mean and S.E value of PCV of chicks of Group I, II, III and IV at the end of 21days were 25.96±1.51, 39.13±0.25, 24.90±2.98, and 39.04±0.46 respectively. Aceclofenac induced affected Groups i.e. Group II and IV revealed comparatively higher PCV than Group I and III. No significant difference in PCV was observed between Group I and III. The vanaraja chicks of induced with aceclofenac + Febuxostat Group III not revealed any significant difference mean value of PCV as compared to control Group I at the end of 21 days.

The Mean and S.E value of PCV of chicks of Group I, II, III and IV at the end of 28 days were 25.71 ± 1.33, 41.79 ± 0.25, 26.36 ± 2.11, and 41.75 ± 4.21 and respectively. Induced Aceclofenac Groups i.e. Group II and IV revealed comparatively higher PCV than Group I and III. No significant difference in PCV was observed between Group I and III. The vanaraja chicks of induced with aceclofenac + Febuxostat Group III not revealed any significant difference mean value of PCV as compared to control Group I at the end of 28 days.

The Mean and S.E value of PCV of vanaraja chickens of Group I, II, III and IV at the end of 42 days were 24.87 ±1.51, 24.38 ± 0.60, 25.15 ± 0.96 and 25.81 ± 1.33 and Respectively. Visceral gout affected vanaraja chickens from group II treated with bottle gourd juice and Group IV treated with Febuxostat drug revealed no significant changes in mean value of PCV as compared to control Group I and group III at the end of 42 days.

Total erythrocyte count

Total erythrocyte count is important parameter of haematology, this is important diagnostic tool to determination of anaemia (decreased), polycythaemia, and haemoconcentration (increased). The Mean SE value of TEC in different experimental groups is presented in Table 5 and chart 5.

The Mean ± S.E value of TEC of vanaraja chicks of Group I, II, III and IV at the end of 7 days were 3.27± 0.07, 3.53 ± 0.16, 3.33 ± 0.19, and 3.89 ± 0.37 respectively.. Revealed No significant differences were observed in all groups at the end 7 days.

The Mean ± S.E value of TEC of vanaraja chicks of Group I, II, III and IV at the end of 14 days were 4.27 ± 0.15, 4.64 ± 0.20, 3.90 ± 0.15and 4.46 ± 0.09 respectively, Aceclofenac induce Groups i.e. Group II and IV revealed comparatively higher TEC than Group I and III. No significant difference in TEC was observed between Group I and III. The

Table 5: Mean ± S.E. values of TEC (10^6/µL) of Vanaraja chickens in different groups at various time intervals

Groups	7 days	14 days	21days	28 days	42days
Group I	3.27± 0.07[a]	3.19 ± 0.13[a]	3.22 ± 0. 08[a]	3.26 ± 0.09[a]	3.21± 0.13[a]
Group II	3.53 ± 0.16[a]	4.64 ± 0.20[b]	5.83 ± 0.12[b]	6.68 ± 0.30 [b]	3.56 ± 0.11 [a]
Group III	3.33 ± 0.19[a]	3.20 ± 0.08[a]	3.22 ± 0.09[a]	3.29 ± 0.10 [a]	3.29 ± 0.09 [a]
Group IV	3.89 ± 0.37[a]	4.46 ± 0.09[b]	5.60 ± 0.30[b]	6.60 ± 0.58 [b]	3.59 ± 0.19[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

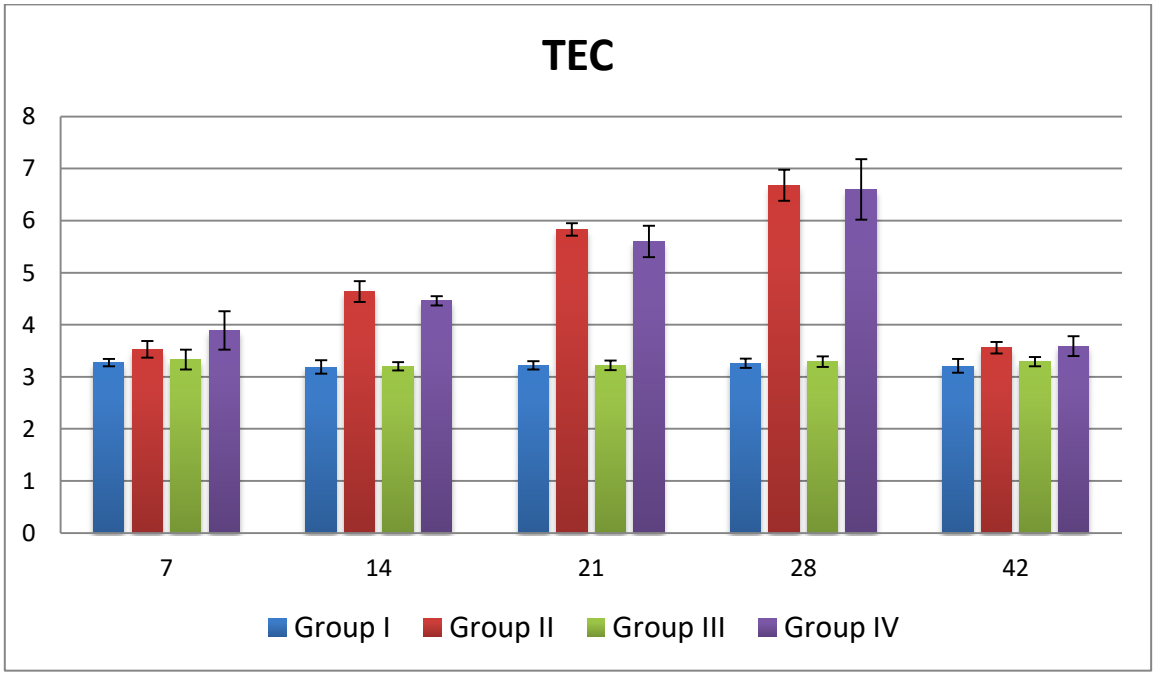


Fig. 5: Histogram showing Mean ± S.E. of TEC (10^6/µL) Vanaraja chickens in different groups at various time intervals

vanaraja chicks of induced with Aceclofenac + Febuxostat (Preventive Group) Group III not revealed any significant change mean value of TEC as compared to control Group I at the end of 14 days.

The Mean ± S.E value of TEC of vanaraja chickens of Group I, II, III and IV at the end of 21 days were 3.22 ± 0.08, 5.83 ± 0.12, 3.22 ± 0.09, and 5.60 ± 0.30 respectively, Aceclofenac induce Groups i.e. Group II and IV revealed comparatively higher TEC than Group I and III. No significant difference in TEC was observed between Group I and III. Group The vanaraja chicks of induced with aceclofenac + Febuxostat (Preventive Group) Group III not revealed any significant change mean value of TEC as compared to control Group I at the end of 21days.

The Mean ± S.E value of TEC of vanaraja chickens of Group I, II, III and IV at the end of 28 days were 3.26 ± 0.09, 6.68 ± 0.30, 3.29 ± 0.10, 6.60 ± 0.58 respectively, Aceclofenac induce group II and IV showed high TEC value as compared to group I and III. No significant difference was observed between group II and III. Vanaraja chickens in group III induced with aceclofenac + febuxostat. There was no significant mean value of TEC observed in group III compared to control Group I at the end of 28 days.

The Mean ± S.E value of TEC of vanaraja chickens of Group I, II, III and IV at the end of 42 days were 3.21± 0.13, 3.56 ± 0.11, 3.29 ± 0.09 and 3.59 ± 0.19 respectively, Gout affected vanaraja chickens belong to group II treated with bottle gourd juice and Group IV treated with Febuxostat drug revealed no significant changes in mean value of TEC compared to control Group I and group III at the end of 42 days.

Total leukocyte count

TLC is important diagnostic tool used for the determination of leucocytosis (inflammatory diseases), leucopoenia (viral diseases) and leukaemia (increased). The Mean and SE value of TLC in different groups are presented in Table 6 and Chart 6.

Mean ± S.E value of TLC of vanaraja chicks of Group I, II, III and IV at the end of 7 days were 24.71 ± 0.56, 24.87 ± 0.61, 24.66 ± 1.08, and 24.56 ± 0.47, in Group I, II and III at the end of 7 days.

The Mean ± S.E value of TLC of vanaraja chicks of Group I, II, III and IV at the end of 14 days were 25.16 ± 0.37, 38.12 ± 0.47, 23.66 ± 0.91 and 38.15 ± 0.63 respectively,

Table 6: Mean ± S.E. values of TLC (.10^3/µL) of Vanaraja chickens in different groups at various time intervals

Groups	7day	14 day	21day	28 day	42day
Group I	24.71 ± 0.56[a]	25.16 ± 0.37[a]	24.74± 0.49 [a]	24.88 ± 0.31 [a]	24.77 ± 0.51 [a]
Group II	24.87 ± 0.61[a]	38.12 ± 0.47[b]	40.46 ±0.48[b]	42.34 ± 0.34 [b]	24.85 ± 0.70 [a]
Group III	24.66 ± 1.08[a]	23.66 ± 0.91[a]	25.33 ± 0.98[a]	24.66 ± 0.61 [a]	24.85 ± 0.50 [a]
Group IV	24.56 ± 0.47[a]	38.15 ± 0.63[b]	40.32 ± 0.69[b]	42.27 ± 0.44 [b]	24.71± 0.42[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

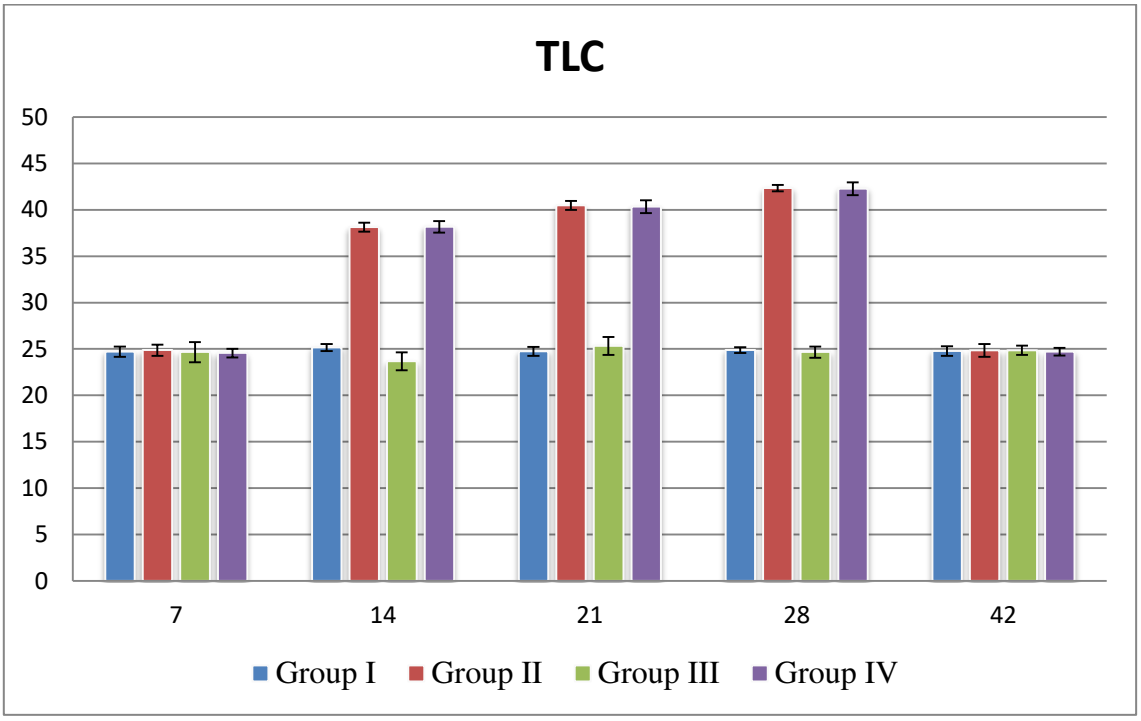


Fig. 6: Histogram showing Mean±S.E. of TLC (10^3/µL) of Vanaraja chickens in different groups at various time intervals

Administered Aceclofenac in group II and IV showed higher TLC value as compared to group I and III. No significant difference was observed between group I and III. Vanaraja chickens in group III induced with aceclofenac + febuxostat. There was no significant mean value of TLC observed in group III compared to control Group I at the end 14 days.

The Mean ± S.E value of TLC of vanaraja chickens chicks of Group I, II, III and IV at the end of 21 days were 24.74± 0.49, 40.46 ± 0.48, 25.33 ± 0.98 and 40.32 ± 0.69 respectively. Administered Aceclofenac affected group II and IV showed higher value as compared to group I and III. No significant difference was observed between group I and III. Vanaraja chickens in group III induced with aceclofenac + febuxostat. There was no significant mean value of TLC observed in group III as compared to control Group I at the end 21 days.

The Mean ± S.E value of TLC of vanaraja chickens of Group I, II, III and IV at the end of 28 days were 24.88 ± 0.31, 42.34 ± 0.34, 24.66 ± 0.61 and 42.27 ± 0.44 respectively. Induced Aceclofenac affected group II and IV showed higher value as compared to group I and III. No significant difference was observed between group I and III. Vanaraja chickens in group III induced with aceclofenac + febuxostat. There was no significant mean value of TLC observed in group III as compared to control Group I at the end 28 days.

The Mean ± S.E value of TLC of chicks of Group I, II, III and IV at the end of 42 days were 24.77 ± 0.51, 24.85 ± 0.70, 24.85 ± 0.50, respectively. Visceral gout affected vanaraja chickens belong to group II treated with bottle gourd juice and Group II treated with Febuxostat drug revealed no significant changes in mean value of TLC compared to control Group I and group III at the end of 42 days.

In the present study increased values of Hb, TEC, PCV and TLC visceral gout affected vanaraja chickens may be due to develop huddling tendency together or scatter respectively leading to low water intake. This results in progressive dehydration and simultaneously haemo–concentration. The agreement with result of (Schmidt *et al.,* 2003), Julian, 1982). Higher lymphocyte count might be due to metabolic acidosis leading to uremia, which may have stimulatory effect on bone marrow leading to leukocytosis.

Different leucocyte count

DLC in different experimental groups are presented in .Table 7, 8.9, 10, 11, Chart 7, 8.9,10,11,

Lymphocyte

The Mean ± S.E value of lymphocyte of vanaraja chicks of Group I, II, III and IV at the end of 7 days were 61.16 ± 0.30, 66.66 ± 0.49, 61.33 ± 0.33 and 60.50 ± 0.22 respectively. No significant alteration were observed in all groups at the end 7 days.

The Mean ± S.E value of lymphocyte of vanaraja chicks of Group I, II, III and IV at the end of 14 days were 60.83 ± 0.30, 71.50 ± 0.42, 60.33 ± 0.21 and 71.66 ± 0.66 respectively. Induced Aceclofenac Groups i.e. Group II and IV revealed comparatively higher lymphocyte than Group I and III. No significant difference in lymphocyte was observed between Group I and III. The vanaraja chicks of induced with aceclofenac + Febuxostat Group III not revealed any significant difference mean value of lymphocyte as compared to control Group I at the end of 14 days.

The Mean ± S.E value of lymphocyte of vanaraja chicks of Group I, II, III and IV at the end of 21 days were 60.00 ± 0.25, 71.66 ± 0.25, 60.50 ± 0.34 and 72.16 ± 0.30 respectively. Aceclofenac administered Groups i.e. Group II and IV revealed comparatively higher lymphocyte than Group I and III. No significant difference in lymphocyte was observed between Group I and III. The vanaraja chicks of induced with aceclofenac + Febuxostat Group III not revealed any significant difference mean value of lymphocyte as compared to control Group I at the end of 21 days.

The Mean ± S.E value of lymphocyte of vanaraja chicks of Group I, II, III and IV at the end of 28 days were 60.16 ± 0.16, 72.50 ± 0.42, 60.00 ± 0.25, and 72.66 ± 0.33 respectively. Induced Aceclofenac Groups i.e. Group II and IV revealed comparatively higher lymphocyte than Group I and III. No significant difference in lymphocyte was observed between Group I and III. The vanaraja chicks of induced with aceclofenac + Febuxostat Group III not revealed any significant difference mean value of lymphocyte as compared to control Group I at the end of 28 days.

The Mean and S.E value of lymphocyte of vanaraja chickens of Group I, II, III and IV at the end of 42 days were 61.50 ± 0.56, 60.00 ± 0.36, 60.16 ± 0.47, and 61.00 ± 0.63 respectively. Visceral gout affected vanaraja chickens from group II treated with bottle gourd juice and Group IV treated with Febuxostat drug revealed no significant changes in mean value of lymphocyte as compared to control Group I and Group III at the end of 42 days.

Table 7: Mean±S.E. values of Lymphocyte (%) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	61.16±0.3[a]	60.83±.030[a]	60.00±0.25[a]	60.16±0.16[a]	61.50±0.56[a]
Group II	66.66±0.4[a]	71.50±0.42[b]	71.66±0.25[b]	72.50±0.42[b]	60.00±0.36[b]
Group III	61.33±0.3[a]	60.33±0.21[a]	60.50±0.34[a]	60.00±0.25[a]	60.16±0.47[a]
Group IV	60.50±0.2[a]	71.66 ±0.66[b]	72.16±0.30[b]	72.66±0.33[b]	61.00±0.63[b]

Superscript a, b, c, d in column indicates significant difference between the groups Within days

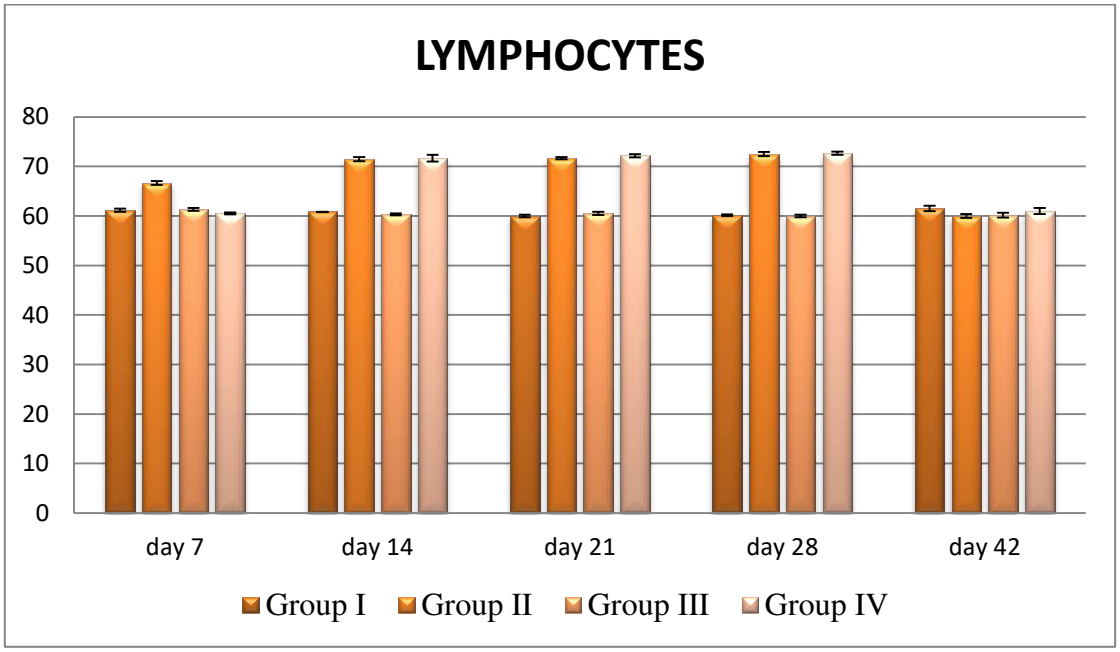


Fig. 7: Histogram showing Mean±S.E. of lymphocyte (%) of Vanaraja chickens in different groups at various time intervals

In the present study increased value of lymphocyte was observed on the day of 14, 21,28 days in induced aceclofenac group II and Group IV our finding agreement with result with Christopher (1977). Who also observed increased value of lymphocyte in gout affected birds.

Increased lymphocyte value might be due to metabolic acidosis leading to uremia, which effect on bone marrow leading leucocytosis.

Heterophils

The Mean ± S.E value of heterophils of vanaraja chicks of Group I, II, III and IV at the end of 7, 14, 21, 28 and 42 days. There was No significant alteration were observed in value of heterophils in all groups at the end 7, 14, 21, 28, and 42 days

Normal heterophils indicated that did not have any infection in vanaraja chickens. Our finding agreement with Koutsos *et al.* (2001), Christopher (1977) reported by there was no significant changes observed in heterophils.

Eosinophils

The Mean ± S.E value of eosinophils of vanaraja chicks of Group I, II, III and IV at the end of 7, 14, 21, 28 and 42 days. there was No significant alteration were observed in value of eosinophil's in all groups at the end 7, 14,21,28,and 42 days.

Normal eosinophil indicated that did not have any parasitic infection in vanaraja chicken. Our finding agreement with Christopher (1977) and Koutsos *et al.* (2001), reported by there was no significant changes observed in eosinophils.

Monocytes

The Mean ± S.E value of Monocyte of vanaraja chicks of Group I, II, III and IV at the end of 14, 21, 28 and 42 days. There was No significant alteration were observed in value of Monocyte in all groups at the end 7, 14, 21, 28, and 42 days.

Normal monocyte indicated that did not have any infection in vanaraja chicken. Our finding agreement with Christopher (1977) and Koutsos *et al.,* (2001), and Singh *et al.* (2013), reported by there was no significant changes observed in monocyte.

Table 8: Mean±S.E. values of Heterophils (%) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	27.33±0.21[a]	27.16±0.16[a]	27.00±0.25[a]	27.33±0.33[a]	27.66±0.21[a]
Group II	27.66±0.21[a]	26.66±0.21[a]	26.50± 0.61[a]	26.33±0.42[a]	27.50±0.34[a]
Group III	27.33±0.21[a]	26.83±o.16[a]	27.33±0.21[a]	27.50±0.42[a]	28.00±0.36[a]
Group IV	27.66±0.21[a]	26.83±0.30[a]	26.66±0.21[a]	26.50±0.42[a]	27.16±0.30[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

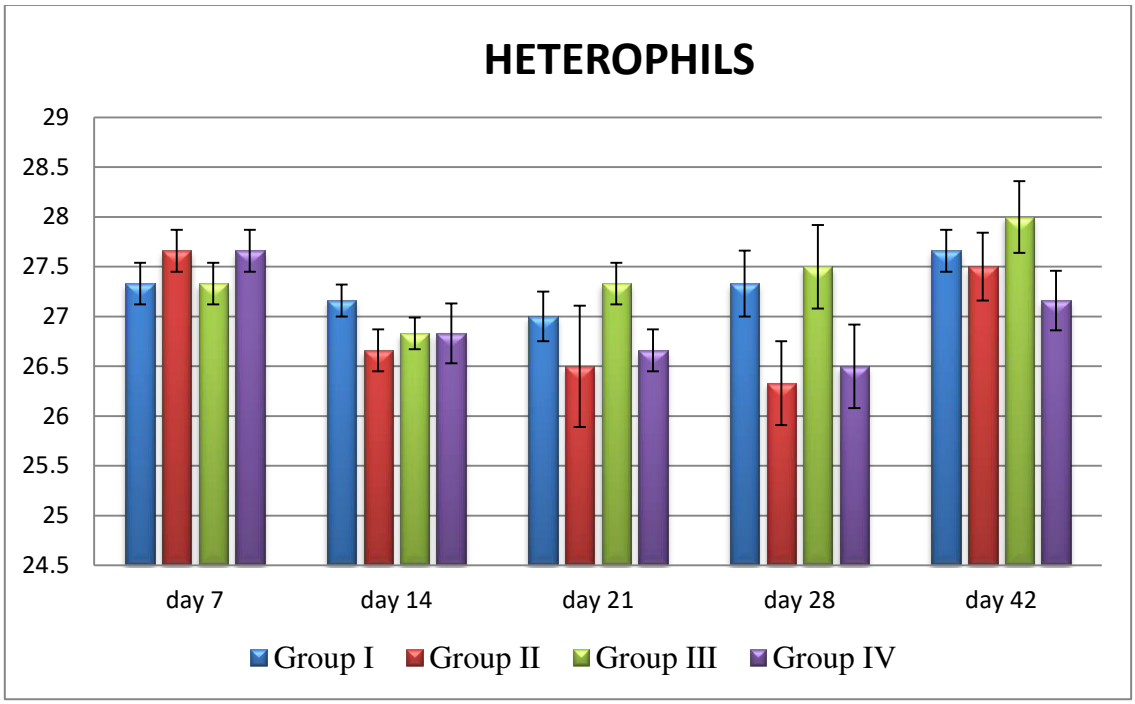


Fig. 8: Histogram showing Mean±S.E. of Heterphils (%) of Vanaraja chickens in different groups at various time intervals

Table 9 : Mean±S.E. values of Eosinophil's (%) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	2.00±0.25[a]	2.66±0.21[a]	2.33±0.33[a]	2.50±0.22[a]	1.83±0.30[a]
Group II	2.50±0.34[a]	2.16±0.16[a]	2.33±0.21[a]	2.33±0.33[a]	2.50±0.42[a]
Group III	2.00±0.25[a]	2.50±0.22[a]	2.33±0.33[a]	2.00±0.25[a]	2.50±0.22[a]
Group IV	2.83±0.30[a]	2.66±0.33[a]	2.50±0.33[a]	2.50±0.22[a]	1.83±0.30[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

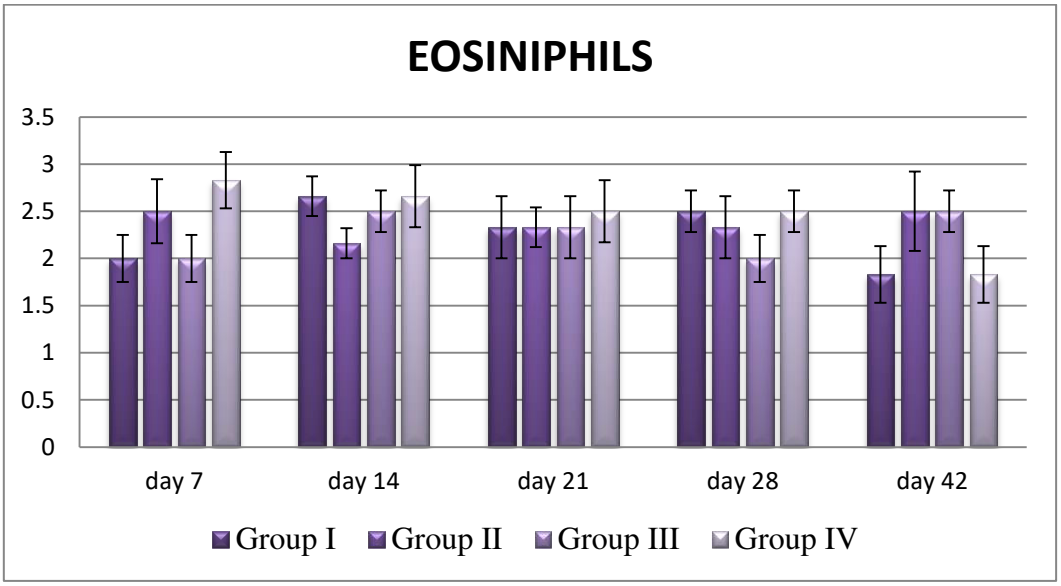


Fig. 9: Histogram showing Mean±S.E. of Eosinophil's (%) of Vanaraja in different groups at various time intervals

Table.11: Mean ± S.E. values of Monocytes (%) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group1	8.66±0.66[a]	9.16±0.40[a]	9.00±0.25[a]	8.33±0.42[a]	9.50±0.34[a]
Group II	9.33±0.33[a]	8.66±0.33[a]	8.83±0.30[a]	8.00±0.00[a]	9.66±0.33[a]
Group III	8.83±0.47[a]	9.16±0.54[a]	8.50±0.34[a]	8.33±0.33[a]	9.16±0.30[a]
Group IV	8.50±0.42[a]	8.33±0.33[a]	8.50±0.22[a]	8.00±0.36[a]	9.00±0.36[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

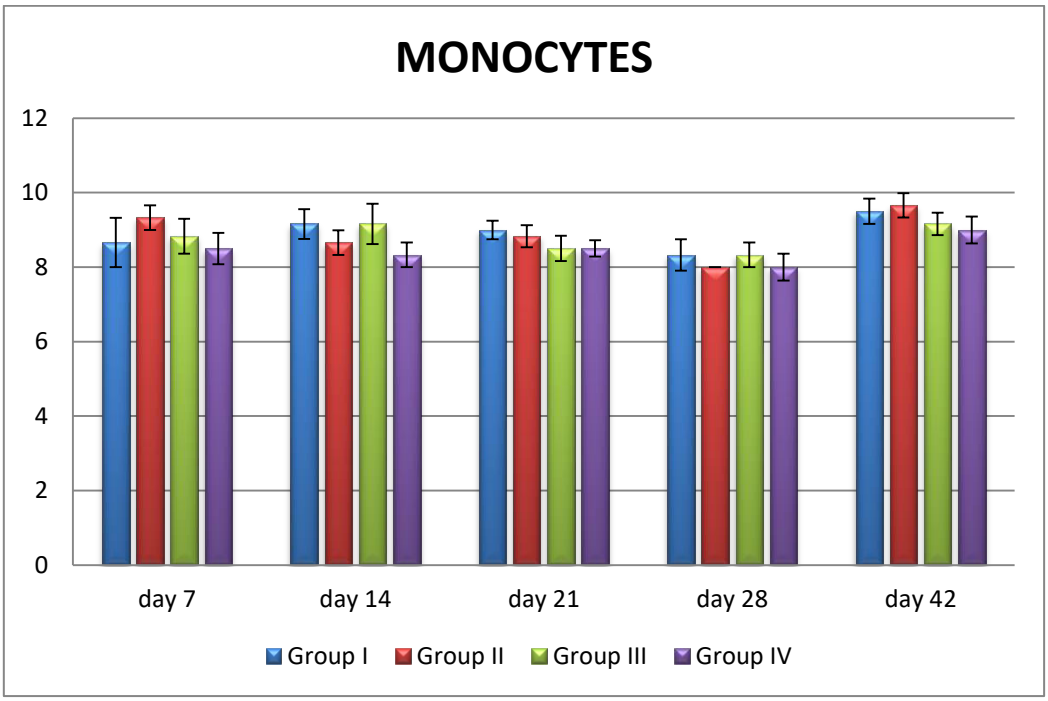


Fig. 11: Histogram showing Mean±S.E. of Monocyte (%) of Vanaraja chickens in different groups at various time intervals

Basophils

The Mean ± S.E value of basophils of vanaraja chicks of Group I, II, III and IV at the end of 7, 14, 21, 28 and 42 days. There was No significant alteration were observed in value of basophils in all groups at the end 7, 14, 21, 28, and 42 days.

Normal basophils indicated that did not have any allergic reaction and infection in vanaraja chickens. Our finding agreement with Christopher (1977) and Koutsos *et al.* (2001), and Singh *et al.,* (2013), reported by there was no significant changes observed in basophils.

Biochemical parameters

All the birds were maintained for 42 days and blood samples was collected at end of the 7^{th}, 14^{th}, 21^{st}, 28^{th} and 42 day. Blood was collected from vanaraja chicken about 2ml of blood. The sample was held at room temperature for 20 min till complete clot formation and reaction. Sample centrifuged at 2000 rpm for 10 min and serum was finally separated in a clean tube and refrigerated at 4^{o}C till analyzed.

The Mean and SE value of serum biochemical parameters for different groups have been summarized in Table.12, 13, 14, 15,16,17,18 and chart. 12, 13, 14, 15,16,17,18

Serum Uric acid

The Mean ± S.E value of serum uric acid of Vanaraja chicks of Group I, II, III and IV at the end of 7 days were 6.24 ± .26, 6.21 ± .26, 6.07 ± 31 and 6.14 ±0.25 respectively, No significant differences were observed in groups I, II, III and IV at the end of 7 days.

The Mean ± S.E value of serum uric acid of Vanaraja chicks of Group I, II, III and IV at the end of 14 days were 6.40 ±.30, 14.80 ± 0.33, 5.95 ± 0.26 and 15.50 ± 0.39 respectively, Aceclofenac induce group II and IV showed significant higher serum uric acid value as compared to group I and III. No significant difference was observed between group II and III. Vanaraja chickens in group III induced with aceclofenac + febuxostat. There was no significant mean value of serum uric acid observed in group III compared to control Group I at the end of 14 days.

The Mean and S.E value of serum uric acid of chicks of Group I, II, III and IV at the end of 21 days were 6.21 ± 0.43, 18.34 ± 0.21, 6.32 ± 0.20 and 18.64 ± 0.29 respectively, Aceclofenac administered Group II and Group IV revealed significantly increase value than

Table 10: Mean ± S.E. values of Basophil's (%) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	1.16±0.30[a]	0.83±0.30[a]	0.83±0.30[a]	1.00±0.25[a]	0.83±0.16[a]
Group II	0.66±0.33[a]	0.50±0.22[a]	0.83±0.16[a]	0.66±0.33[a]	0.50±0.22[a]
Group III	1.00±0.36[a]	0.66±0.21[a]	1.16±0.30[a]	0.83±0.30[a]	0.66±0.21[a]
Group IV	0.66±0.21[a]	0.66±0.21[a]	0.66±0.21[a]	0.83±0.16[a]	0.50±0.22[a]

Superscript a, b, c, d in column indicates significant difference between the groups Within days

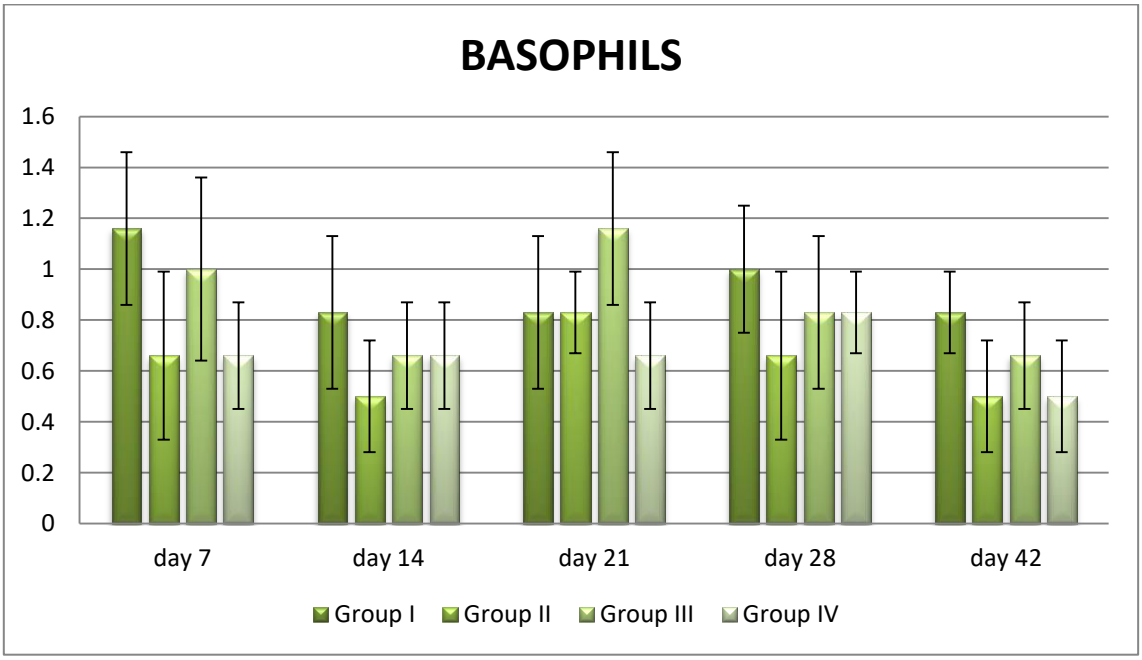


Fig. 10: Histogram showing Mean±S.E. of Basophil's (%) of Vanaraja chickens in different groups at various time intervals

groups II, III. Vanaraja chickens in group III induced with aceclofenac + Febuxostat. There was no significant mean value of serum uric acid observed in group III compared to control Group I at the end of end of 21days.

The Mean and S.E value of Serum uric acid of Vanaraja chicks of Group I, II, III and IV at the end of 28 days were 5.16 ± 0.17, 24.07 ± 0.61, 6.39 ± 0.30 and 24.21 ± .54 respectively, Aceclofenac induced Group II and Group IV revealed significantly higher values than groups II, III. No significant difference was observed between group II and III. Vanaraja chickens in group III administered aceclofenac + Febuxostat. There was no significant mean value of serum uric acid observed in group III compared to control Group I at the end of end of 28 days.

The Mean ± S.E value of serum uric acid of Vanaraja chicks of Group I, II, III and IV at the end of 42 days were 6.12 ± 0.25, 6.41 ± 0.23, 5.81 ± 0.38 and 5.63 ± 0.15 respectively. Visceral gout affected vanaraja chickens belong to group II treated with bottle gourd juice and Group IV treated with Febuxostat drug revealed no significant changes in mean value of serum uric acid compared to control Group I and group III. Vanaraja chickens in group III induced with aceclofenac + Febuxostat. There was no significant mean value of serum uric acid observed in group III compared to control Group I at the end of end at the end of 42 days.

Plasma uric acid analysis is an important biochemical diagnostic tool to know the uric acid metabolism and functional status of kidney. Damaged to the kidney affect uric acid excretion leading visceral gout and hyperurecaemic.

In present study increased value of uric acid in group II and Group IV at end of 14 days, 21 days 28 days during the present study revealed that aceclofenac negatively affect kidney functions which was outward terms of serum uric acid level similar finding also reported by Gajera (2006), Shinde (2007), Patel *et al.* (2007), Sharma *et al.,* (2012), Ghodasara *et al.,* (2014), and Ramjan *et al.,* (2015) also observed increased uric acid level in diclofenac treated group as compare to control group. Uric acid level increased in gout affected birds were also reported by Chandra et al (1985), Mert(1991), Uma et al., (1997), Oaks et al., (2004), Irtaza et al., (2008), Hedaoo et al., (2008), Rahul (2010) and Sharma and vegad (2010).

It was apparent that aceclofenac caused renal failure and hyperuricaemia. Kidney functions were compromised. It failed to excrete out uric acid properly. Later on, Blood became supersaturated with uric acid and it was precipitated in kidney, liver, spleen, and many mucosal and serosal surfaces in the form of urate crystals. Cellular reaction to these crystals further activated inflammatory changes in the kidney finally resulted in gout.

Decreased serum uric acid in Febuxostat treated group in vanaraja birds. The decreased level of uric acid in other species of animal affected with gout were also reported by Horiuchi *et al.* (1999) revealing dose dependent reduction of serum uric acid for Febuxostat was 1.6 mg/kg vs.9.1 mg/kg for allopurinol at uricase inhibition by oxonate over 6 days. Khosravan et al., (2006) noticed Febuxostat treatment in human resulted in significant decreases in serum and urinary urate concentration and increased in serum urate concentrations ranged from 27% to 76%. Komoriya *et al.* (2004) found serum uric acid level in human were 8.60, 8.56, 8.37, 8.53, 8.46, 8.62 and 9.11 mg/100 ml at24, 22, 20,18,16,12 h and just before the first administration of febuxostat. After 4 week treatment phase, the levels were 5.96, 5.82, 5.61, 5.53, 5.51, 5.56 and 6.40 mg /100 ml just before and 2, 4, 6, 8, 12 and 24 hours after the final dose. These results indicate that Febuxostat reduces Uric Acid level by 33.3%.

No any other systemic study or classified approach was made in relation to decreased serum uric acid in bottle gourd treated group in vanaraja birds decreased level of uric acid in other species of animal affected with gout were also reported by Kausar and Waris (2016) revealed that treatment was done by bottle gourd which not only helped to remove excessive uric acid in blood, but also other organs remained unaffected. In current study, uric acid increased by direct oral dose of uric acid. Rats were divided into four groups; control, Zyloric treated, bottle gourd orally treated and bottle gourd extract treated.

In present study aceclofenac induce group II treated with bottle gourd juice and it is obvious that bottle gourd juice have good effect on aceclofenac induced visceral gout in vanaraja birds. Experimental study in vanaraja birds shown that significant reduce serum uric acid after the end of 28 days and 42 days treated with bottle gourd juice. Group II showed significant decreases as compared to control group I as well as Preventive group III (aceclofenac + Febuxostat) the finding suggested that effective eradication of uric acid by 20ml bottle gourd juice in vanaraja birds.

In present study aceclofenac induce group IV treated with febuxostat. It is obvious that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja birds.

Table. 12: Mean ± S.E. values of uric acid of (mg/dl) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14day	21days	28days	42days
Group I	6.24 ± .26 [a]	6.40 ±.30[a]	6.21 ± 0.43 [a]	5.16 ± 0.17 [a]	6.12 ± 0.25 [a]
Group II	6.21 ±0.26[a]	14.80 ± .33[b]	18.34 ± 0.21[b]	24.07 ± 0.61[b]	6.41 ±0.23[a]
Group II	6.07 ±0.31[a]	5.95 ± 0.26 [a]	6.32±0.20[a]	6.39 ± 0.30 [a]	5.81± 0.38[a]
Group IV	6.14±0.25[a]	15.50 ± .39[b]	18.64±0.2[b]	24.21±0.5[b]	5.63 ± 0.15 [a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

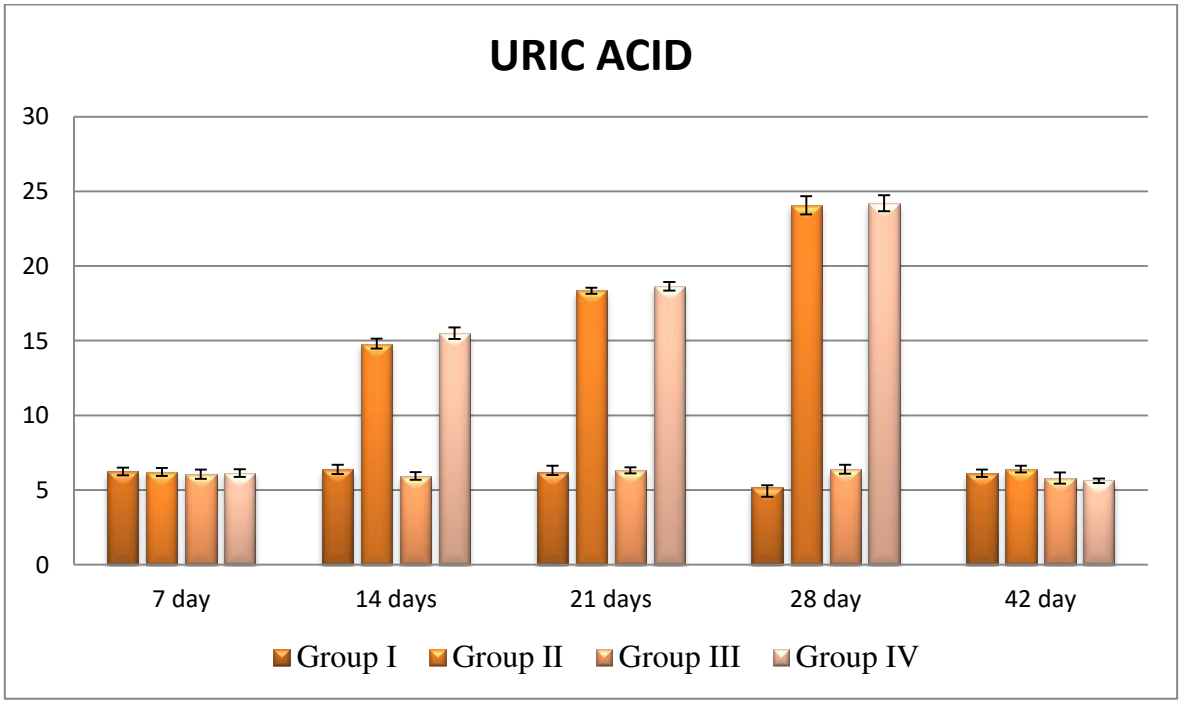


Fig. 12: Histogram showing Mean±S.E. of uric acid of (mg/dl) of Vanaraja chickens in different groups at various time intervals

Experimental study in vanaraja birds shown that significant reduce serum uric acid after the end of 28 week and 42 days treated with Febuxostat. Group IV showed significant decreases as compared to control group I as well as Preventive group III (aceclofenac + Febuxostat) the finding suggested that effective elimination of uric acid by 4mg/kg febuxostat drug in vanaraja birds.

Serum Creatinine

Creatinine analysis is an important biochemical diagnostic tool to know the functional status of kidneys. Creatinine is primarily cleaned out of the blood by the kidneys glomerular filtration and proximal tubular secretion.

The Mean ± S.E value of serum creatinine of Vanaraja chicks of Group I, II, III and IV at the end of 7 days were 0.51 ± 0.059, 0.49 ± 0.083, 0.50 ± 0.028, and 0.50 ± 0.061 respectively. No significant differences were observed in groups I, II, III and IV at the end of 7 days.

The Mean ± S.E value of serum Creatinine of Vanaraja chicks of Group I, II, III and IV at the end of 14 days were 0.52 ± .076, 1.52 ± 0.206, 0.52 ± 0.115 and 1.53 ± 0.196 respectively, The vanaraja chickens of aceclofenac induced Group II and Group IV showed higher significant changes in value of serum creatinine as compared to Group I and Group III. there was no significant mean value of serum creatinine observed in group III as compared to control Group I. Group III administered Aceclofenac + Febuxostat, there was no significant mean value of serum creatinine observed in group III compared to control Group I at the end of 14 days.

The Mean ± S.E value of serum creatinine of Vanaraja chickens of Group I, II, III and IV at the end of 21 days were 0.51±0.035, 1.84 ± 0.32, 0.54±0.085, and 1.80±0.246 respectively Aceclofenac induced Group II and Group IV revealed significantly higher values than groups II, III. No significant difference was observed between group II and III. Vanaraja chickens in group III administered aceclofenac + Febuxostat. There was no significant mean value of serum creatinine observed in group III compared to control Group I at the end of end of 21 days.

The Mean ± S.E value of serum creatinine of Vanaraja chickens of Group I, II, III and IV at the end of 28 days were 0.50 ± 0.027, 1.98 ± 0.117, 0.56 ± 0.179 and 2.15 ± 0.162 respectively. The vanaraja chickens of aceclofenac induced Group II and Group IV showed

higher significant changes in value of serum creatinine as compared to Group I and Group III. There was no significant mean value of serum creatinine observed in group III as compared to control Group I. Vanaraja chickens in group III administered aceclofenac + Febuxostat. There was no significant mean value of serum creatinine observed in group III compared to control Group I at the end of 28 days.

The Mean ± S.E value of serum creatinine of Vanaraja chickens of Group I, II, III and IV at the end of 42 days were 0.54 ± 0.096, 0.58 ± 0.088, 0.50 ± 0.044 and 0.53 ± 0.122 respectively, Visceral gout affected vanaraja chickens belong to group II treated with bottle gourd juice and Group IV treated with Febuxostat drug revealed no significant changes in mean value of serum creatinine compared to control Group I and group III at the end of 42 days.

In present study increased value of creatinine in group II and Group IV at end of 14 days, 21 days 28 days during the present study revealed that aceclofenac induce affect kidney function in term of increase in creatinine levels. Similar finding is also reported by Feizi *et al.* (2012). Irtaza *et al.,* (2008), Shinde (2007), Madhuri *et al*., (2008) , Hedaoo *et al.,*(2008), Jain *et al* (2009), Awan *et al* (2011), and Undhad *et al.,* (2012), Ghanvat *et al.,* (2012), Sharma *et al.,* (2012), Ghodasara *et al* (2014).

No any other systemic study or classified approach was made in relation to decreased serum creatinine in bottle gourd treated group in vanaraja chickens decreased level of serum creatinine in other species of animal affected with gout were also reported by Kausar and Waris (2016) revealed that treatment was done by bottle gourd which not only helped to remove excessive uric acid in blood, but also other organs remained unaffected. In current study, uric acid increased by direct oral dose of uric acid. Rats were divided into four groups; control, Zyloric treated, bottle gourd orally treated and bottle gourd extract treated.

Aceclofenac induces caused necrosis of proximal convoluted tubules lead to reduction in uric acid and creatinine excretion causing rapid rise of uric acid and creatinine in blood. Increased Serum creatinine value were revealing of kidney damage due to aceclofenac elevation of serum uric acid and creatinine levels are indicators of nephrotoxicity.

In present experimental study aceclofenac induce group II treated with bottle gourd juice and it is obvious that bottle gourd juice have good effect on aceclofenac induced visceral gout in vanaraja birds. Experimental study in vanaraja birds shown that significant

Table.13 : Mean ± S.E. values of creatinine of (mg/dl) of Vanaraja chickens in different groups at various time intervals

Groups	7day	14day	21days	28days	42days
Group I	0.51±0.059[a]	0.52 ± .076 [a]	0.51±0.035[a]	0.50± 0.027[a]	0.54±0.096 [a]
Group II	0.49±0.083[a]	1.52 ± 0.206 [b]	1.84 ± 0.32[b]	1.98± 0.117 [b]	0.58± 0.088[a]
Group III	0.50±0.028[a]	0.52 ± 0.115 [a]	0.54±0.085[a]	0.56±0.179[a]	0.50± 0.044 [a]
Group IV	0.50±0.061[a]	1.53 ± 0.196[b]	1.80±0.246[b]	2.15±0.162[b]	0.53± 0.122 [a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

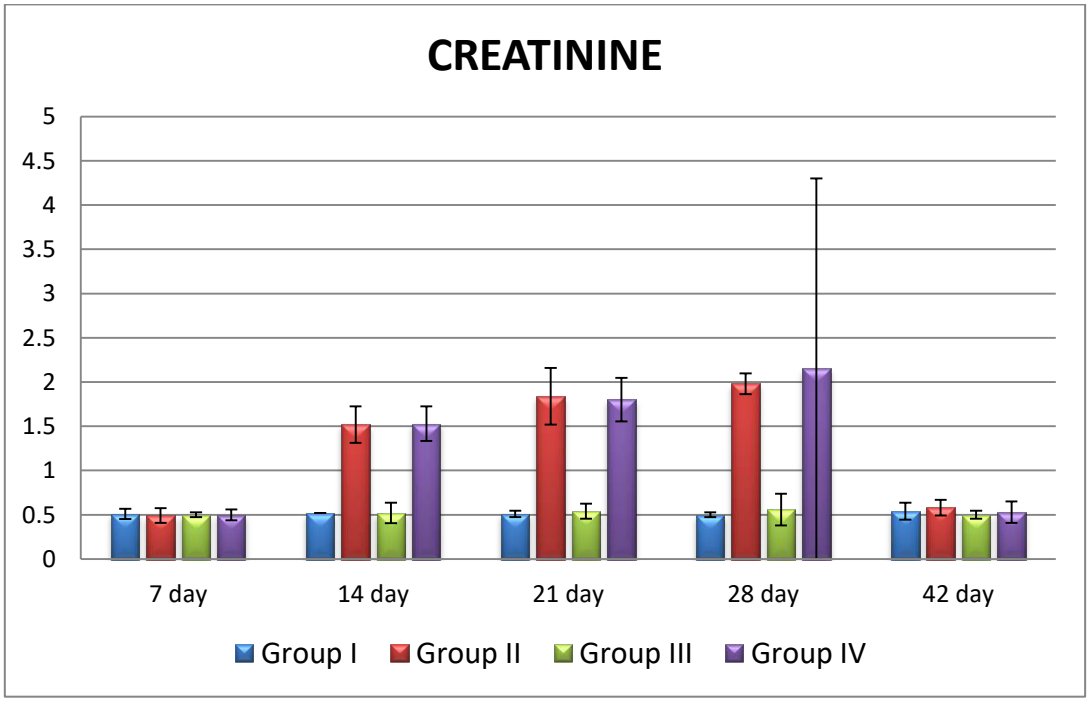


Fig.13: Histogram showing Mean±S.E. of creatinine of (mg/dl) of Vanaraja chickens in different groups at various time intervals

reduce serum creatinine after the end of 28 week and 42 days treated with bottle gourd juice. Group II showed significant decreases serum creatinine level as compared to control group I as well as Preventive group III (Aceclofenac + Febuxostat). It was apparent that bottle gourd juice have protective effect on aceclofenac induced visceral gout in vanaraja chicken.

In present study aceclofenac induce group IV treated with febuxostat. It was evident that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja birds. Experimental study in vanaraja birds shown that significant reduce serum creatinine level after the end of 28 week and 42 treated with Febuxostat. Group IV showed significant decreases as compared to group I (control group) as well as Preventive group III (Aceclofenac + Febuxostat). in the absence of available literature the value of creatinine further indicate that the both the dose of febuxostat through decreased the serum creatinine concentration which was elevated due to aceclofenac toxicity., their efficacy in reducing creatinine levels was almost similar. It was apparent that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja chicken.

Serum total protein

The Mean ± S.E value of Serum total protein of vanaraja chicks of Group I, II, III and IV at the end of 7, 14, 21, 28 and 42 days. there was No significant alteration were observed in value of Serum total protein in all groups at the end 7, 14,21,28, and 42 days.

Normal TSP indicated that did not have any infection in vanaraja chickens. Our findings agreements with Singh *et al.,* (2013), Chandra *et al.,* (1985) reported by there was no significant changes observed in Serum total protein.

Serum Albumin

The Mean ± S.E value of Albumin of Vanaraja chicks of Group I, II, III and IV at the end of 7 days were1.82 ± 0.10, 1.98 ± 0.16, 1.69 ± 0.11, and 1.96 ± 0.10 respectively. No significant differences were observed in groups I, II, III and IV at the end of 7 days.

The Mean ± S.E value of Albumin of Vanaraja chicks of Group I, II, III and IV at the end of 14days were 2.08 ± 0.19, 3.83 ± 0.02, 2.07 ± 0.14 and 3.80 ± 0.09 respectively. Aceclofenac induced affected Group II and IV were showed significantly increased values than Groups I and Group III. Aceclofenac + Febuxostat administered in vanaraja chickens in no significant

mean value observed in group III as at end of compared to control Group I at the end of 14 days.

The Mean ± S.E value of Albumin of Vanaraja chicks of Group I, II, III and IV at the end of 21 days were 2.06 ± 0.11, 4.43 ± 0.10, 2.01 ± 0.20 and 4.48 ± 0.13 respectively. The vanaraja chickens of aceclofenac induced Group II and Group IV showed higher significant changes in value of serum Albumin as compared to Group I and Group III. there was no significant mean value of serum albumin observed in group III as compared to control Group I. Group III administered aceclofenac + Febuxostat .there was no significant mean value of Serum Albumin observed in group III compared to control Group I at the end of 21 days.

The Mean ± S.E value of Albumin of Vanaraja chicks of Group I, II, III and IV at the end of 28 days were 1.48 ± 0.11, 4.69 ± 0.11, 1.62 ± 0.18 and 4.69 ± 0.08 respectively. The vanaraja chickens of aceclofenac induced Group II and Group IV showed higher significant changes in value of serum Albumin as compared to Group I and Group III. there was no significant mean value of serum albumin observed in group III as compared to control Group I. Group III administered aceclofenac + Febuxostat .there was no significant mean value of Serum Albumin observed in group III compared to control Group I at the end of 28 days.

The Mean ± S.E value of Serum Albumin of Vanaraja chickens of Group I, II, III and IV at the end of 42 days were 1.98 ± 0.13, 2.13 ± 0.33, 2.29 ± 0.22 and 2.52 ± 0.33 respectively. Bottle gourd treated group II and febuxostat treated group IV No significant differences were observed in groups I, II, III and IV at the end of 42 day.

In present study increased value of albumin in group II and Group IV at end of 14 days, 21 days 28 days during the present study revealed that aceclofenac induced the level of serum albumin was significantly increased as compared group I and Group III. This finding agreement with Chandra *et al.* (1985), Singh *et al.* (2013).

Increased serum albumin value might be due to dehydration.

In present experimental study aceclofenac induced group II treated with bottle gourd juice, it is observed that bottle gourd juice have good effect on aceclofenac induced visceral gout produced in vanaraja chickens. Experimental study on vanaraja birds shown that significant decreased albumin level after the end of 28 days to 42 days treated with bottle gourd juice. Group II showed significant decreased serum albumin level as compared to control group I as well as Preventive group III (aceclofenac + Febuxostat). It was apparent

Table. 14: Mean ± S.E. values of albumin of (g/dl) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	1.82±0.10[a]	2.08±0.19[a]	2.06±0.11[a]	1.48 ± 0.11[a]	1.98 ± 0.13[a]
Group II	1.98±0.16[a]	3.83±0.02[b]	4.43±0.10[b]	4.69 ± 0.11[b]	2.13 ± 0.33[a]
Group III	1.69±0.11[a]	2.07±0.14[a]	2.01±0.20[a]	1.62 ± 0.18	2.29 ± 0.22[a]
Group IV	1.96±0.10[a]	3.80±0.09[b]	4.48±0.13 [b]	4.69 ± 0.08[b]	2.52 ± 0.33[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

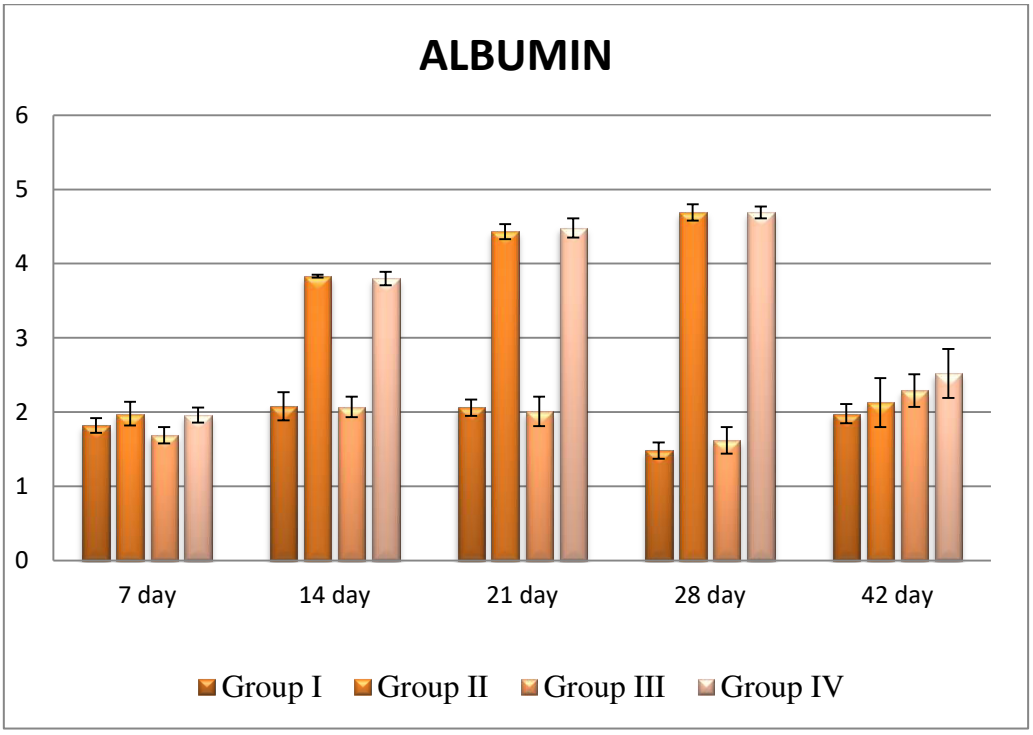


Fig.14: Histogram showing Mean±S.E. of Albumin of (g/dl) of Vanaraja in different groups at various time intervals

Table. 15: Mean ± S.E. values of TSP of (g/dl) of Vanaraja in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	4.78± 0.18[a]	4.53±0.28[a]	4.29± 0.10[a]	4.46± 0.18[a]	4.15±0.24[a]
Group II	4.61± 0.17[a]	4.50±0.13[a]	4.45± 0.18[a]	4.63± 0.19[a]	4.56±0.22[a]
Group III	4.50±0.22[a]	4.80±0.19[a]	4.61±0.29[a]	3.73±0.66[a]	4.13±0.21[a]
Group IV	4.70±0.22[a]	4.63±0.19[a]	4.73± 0.28[a]	4.68± 0.22[a]	4.53± 0.21[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

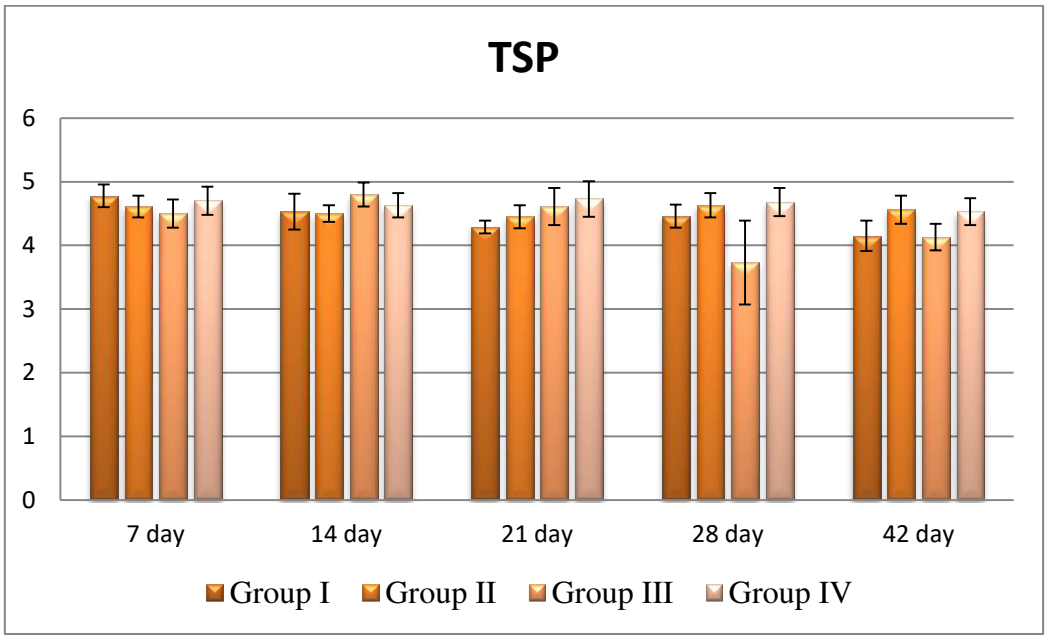


Fig. 15: Histogram showing Mean±S.E. of TSP of (g/dl) of Vanaraja chickens in different groups at various time intervals

that bottle gourd juice have protective effect on aceclofenac induced visceral gout in vanaraja chicken. In the absence of available literature the value of serum Albumin further indicate that bottle gourd juice normal Albumin concentration which was decreased due to aceclofenac toxicity.

In present study aceclofenac induce group IV treated with febuxostat. It was evident that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja birds. Experimental study in vanaraja birds showed that significant decreased albumin level after the end of 28 days and 42days treated with Febuxostat. Group IV showed no significant as compared to group I (control group) as well as Preventive group III (aceclofenac + Febuxostat).

Serum Globulin

The Mean ± S.E value of Globulin of Vanaraja chicks of Group I, II, III and IV at the end of 7 days were 2.96 ± 0.23, 2.63 ± 0.19, 2.80 ± 0.28, and 2.74 ± 0.21 respectively, No significant differences were observed in groups I, II, III and IV at the end of 7 days.

The Mean ± S.E value of Globulin of Vanaraja chicks of Group I, II, III and IV at the end of 14 days were 2.44± 0.33, 0.67±0.13, 2.72±0.27 and 0.82±0.24 respectively, Aceclofenac induced affected Group II and IV were showed significantly lower values than Groups I and Group III. Aceclofenac + Febuxostat administered in vanaraja chickens in no significant mean value observed in group III as at end of compared to control Group I at the end of 14 days.

The Mean ± S.E value of Globulin of Vanaraja chicks of Group I, II, III and IV at the end of 21 were 2.22 ± 0.14, 0.49 ± 0.12, 2.42 ± 0.42 and 0.48 ± 0.17 respectively. The vanaraja chickens of aceclofenac induced Group II and Group IV showed decreased significant changes in value of serum Globulin as compared to Group I and Group III. there was no significant mean value of serum Globulin observed in group III as compared to control Group I. Group III administered aceclofenac + Febuxostat .there was no significant mean value of Serum Globulin observed in group III compared to control Group I at the end of 21 days.

The Mean ± S.E value of Globulin of Vanaraja chicks of Group I, II, III and IV at the end of 28 were 2.97 ± 0.12, 0.42 ± 0.13, 2.42 ± 0.42 and 0.50 ± 0.10 respectively. The vanaraja chickens of aceclofenac induced Group II and Group IV showed decreased

significant changes in value of serum Globulin as compared to Group I and Group III. there was no significant mean value of serum Globulin observed in group III as compared to control Group I. Group III administered aceclofenac + Febuxostat .there was no significant mean value of Serum Globulin observed in group III compared to control Group I at the end of 28 days.

The Mean ± S.E value of serum Globulin of Vanaraja chickens of Group I, II, III and IV at the end of 42 days were 2.15 ± 0.19, 2.43 ± 0.37, 1.85 ± 0.36 and 2.01 ± 0.21 respectively, Bottle gourd treated group II and febuxostat treated group IV. No significant differences were observed in groups I, II, III and IV at the end of 42 days.

In present study decreased value of Globulin in group II and Group IV at end of 14 days, 21 days 28 days during the present study revealed that aceclofenac induced the level of serum globulin was significantly lower as compare group I and Group III. This finding agreement with Chandra *et al.,* (1985), Singh *et al.* (2013) might be due to emaciation and dehydration.

In present experimental study aceclofenac induced group II treated with bottle gourd juice, it is observed that bottle gourd juice have good effect on aceclofenac induced visceral gout produced in vanaraja chickens. Experimental study on vanaraja birds shown that significant normal serum Globulin after the end of 28 days to 42 days treated with bottle gourd juice. Group II showed significant normal serum globulin level as compared to control group I as well as Preventive group III (aceclofenac + Febuxostat). It was apparent that bottle gourd juice have protective effect on aceclofenac induced visceral gout in vanaraja chicken. In the absence of available literature the value of serum globulin further indicate that bottle gourd juice normal globulin concentration which was decreased due to aceclofenac toxicity.

In present study aceclofenac induce group IV treated with febuxostat. It was evident that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja birds. Experimental study in vanaraja birds shown that significant normal serum globulin level after the end of 28 days and 42.treated with Febuxostat. Group IV showed no significant as compared to group I (control group) as well as Preventive group III (aceclofenac + Febuxostat).

Table. 16: Mean ± S.E. values of Globulin of (g/dl) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	2.96±0.23[a]	2.44± 0.33[a]	2.22±0.14[a]	2.97± 0.12[a]	2.15± 0.19[a]
Group II	2.63±0.19[a]	0.67±0.13[b]	0.49± 0.12[b]	0.42± 0.13[b]	2.43± 0.37[a]
Group III	2.80±0.28[a]	2.72±0.27[a]	2.42± 0.42[a]	2.42± 0.42[a]	1.85±0.36[a]
Group IV	2.74±0.21[a]	0.82±0.24[b]	0.48± 0.17[b]	0.50± 0.10[b]	2.01± 0.21[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

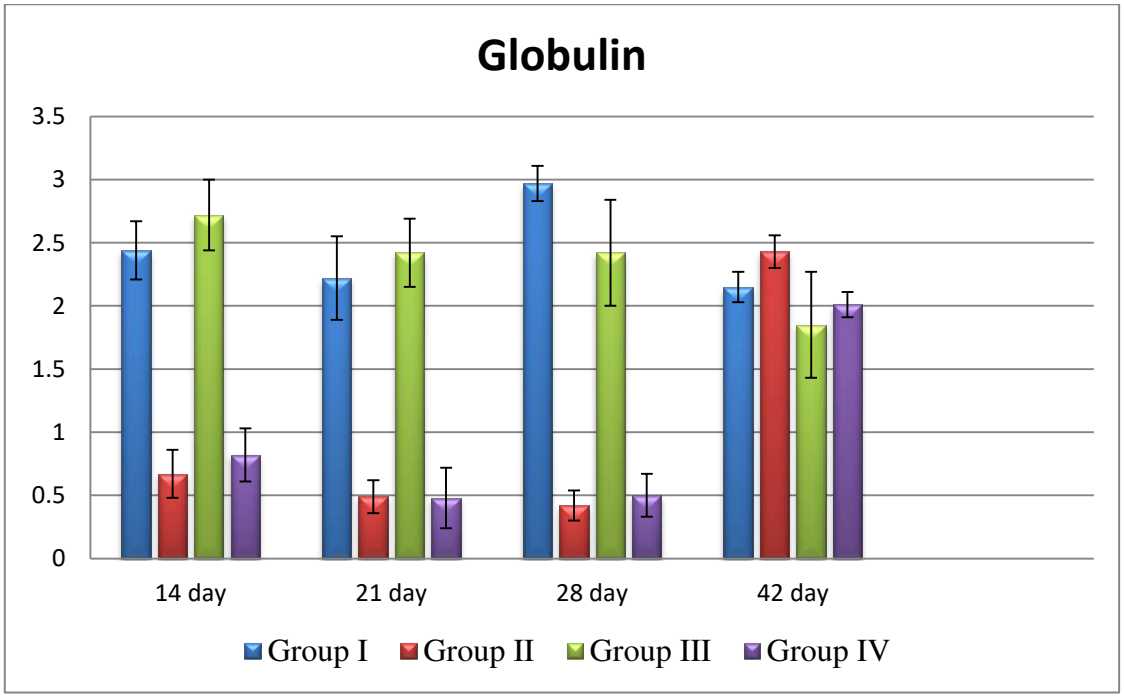


Fig. 16: Histogram showing Mean±S.E. of Globulin of (g/dl) of Vanaraja chickens in different groups at various time intervals

Serum Glutamic Pyruvic transaminase (U/L)

Serum glutamic pyruvic transaminase is also known as Alanine transaminase. SGPT is an enzyme mostly found in the cell of liver but also smaller amount in the kidney, heart, muscles etc. Serum SGPT mainly used for the detection for the functional status of liver and development of hepatocellular injury.

The Mean ± S.E value of serum SGPT of Vanaraja chicks of Group I, II, III and IV at the end of 7 days were 33.23 ± 1.00, 33.89 ± 0.93, 33.75 ± 1.28 and 33.65 ± 1.16 respectively, No significant differences were observed in groups I, II, III and IV at the end of 7 days.

The Mean ± S.E value of Serum SGPT of Vanaraja chicks of Group I, II, III and IV at the end of 14 days were 33.40 ±0.71, 48.47 ± 2.56, 33.83 ± 1.15 and 48.41 ± 0.92 respectively. Aceclofenac administered vanaraja chickens Group II and Group IV showed higher significant changes in value of serum SGPT as compared to Group I and Group III. There was no significant mean value of serum SGPT observed in group III. Vanaraja chickens in group III induced with aceclofenac + Febuxostat .There was no significant mean value of serum SGPT observed in group III compared to control Group I at the end of 14 days.

The Mean ± S.E value of Serum SGPT of Vanaraja chickens of Group I, II, III and IV at the end of 21 days were 33.03 ± 0.78, 51.75 ± 2.16, 33.65 ± 0.74 and 51.48 ± 1.74 respectively. 2 The vanaraja chickens of aceclofenac induced Group II and Group IV showed higher significant changes in value of serum SGPT as compared to Group I and Group III. There was no significant mean value of serum SGPT observed in group III as compared to control Group I. Group III revealed no any significant alteration in SGPT value as compared group I. Vanaraja chickens in group III induced with aceclofenac + Febuxostat . There was no significant mean value of serum SGPT observed in group III compared to control Group I. At the end of 21 days.

The Mean ± S.E value of Serum SGPT of Vanaraja chickens of Group I, II, III and IV at the end of 28 days were 33.49 ± 0.53, 56.36 ± 0.88, 33.68 ± 0.85 and 57.27 ± 2.21 respectively, Aceclofenac administered vanaraja chickens Group II and Group IV showed higher significant changes in value of serum SGPT as compared to Group I and Group III. There was no significant mean value of serum SGPT observed in group III. Vanaraja

chickens in group III induced with aceclofenac + Febuxostat .There was no significant mean value of serum SGPT observed in group III compared to control Group I at end of 28 days.

The Mean ± S.E value of SGPT of Vanaraja chickens of Group I, II, III and IV at the end of 42 days were 33.65 ± 0.69, 32.75 ± 1.38, 33.93 ± 0.97, and 32.23±2.81 respectively, Bottle gourd treated group II and febuxostat treated group IV. There were no significant differences were observed in groups I, II, III and IV at the end of 42 day.

Aceclofenac administered vanaraja chickens Group II and Group IV showed higher significant difference in value of serum SGPT as compared to Group I and Group III. There was no significant mean value of serum SGPT observed in group III as compared to control Group I. Vanaraja chickens in group III induced with Aceclofenac + Febuxostat. There was no significant mean value of serum SGPT observed in group III compared to control Group I at the end of 28 days.

In present experimental study, increased value of Serum SGPT in group II and Group IV at end of 14 days, 21 days 28 days during the present study revealed that aceclofenac induce affect liver function in term of increase in serum SGPT levels. Increased in the activity of SGPT in aceclofenac induce group II and group IV may be due severe nephritis are the result of leakage from various organs such as liver, heart and kidney mainly, as uric acid granulomas formation replace partial/whole parenchymatous tissues.

Our finding agreement with Chandra et al., (1984), Similar finding is also reported by Garcia (1998) and Jain *et al.,* (2009). Darbar *et al*, (2010), Kausar and Waris, (2016) reported that found increased level of ALT in producing gout by given oral dose of uric acid in rat.

Decreased serum SGPT in Febuxostat treated group in vanaraja chickens .the decreased level of serum SGPT in other species of animal affected with gout were also reported by Patel (2017) reported that treated groups with febuxostat medium and high dose respectively along with diclofenac revealed significant decrease in serum ALT.

In present experimental study aceclofenac induce group II treated with bottle gourd juice, it is observed that bottle gourd juice have good effect on aceclofenac induced visceral gout produced in vanaraja chickens. Experimental study on vanaraja chickens shown that significant reduce serum SGPT after the end of 28 says to 42 days treated with bottle gourd juice. Group II showed significant decreases serum SGPT level as compared to control group

Table. 17: Mean ± S.E. values of SGPT (U/L) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14day	21days	28days	42days
Group I	33.23± 1.00[a]	33.40±0.71[a]	33.03±0.78[a]	33.49±0.53[a]	33.65±0.69[a]
Group II	33.89±0.93[a]	48.47±2.56[b]	51.75±2.16[b]	56.36±0.88 [b]	32.75±1.38[a]
Group III	33.75±1.28 [a]	33.83±1.15 [a]	33.65±0.74[a]	33.68±0.85[a]	33.93±0.97[a]
Group IV	33.65±1.16 [a]	48.41±0.92 [b]	51.48±1.74[b]	57.27±2.21[b]	32.23±2.81[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

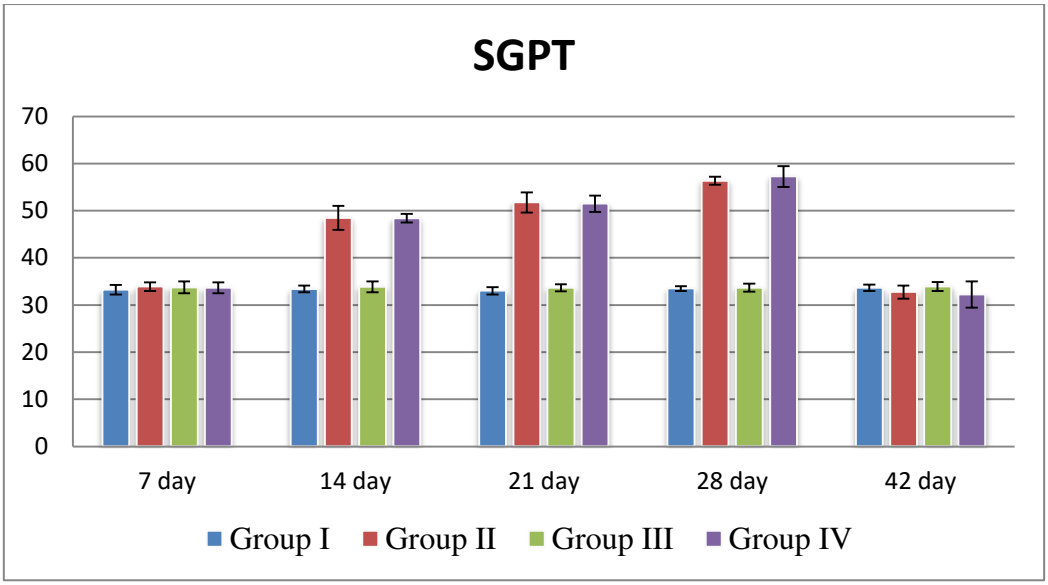


Fig. 17: Histogram showing Mean±S.E. of SGPT of (g/dl) of Vanaraja in different groups at various time intervals

I as well as Preventive group III (aceclofenac + Febuxostat). It was apparent that bottle gourd juice have protective effect on aceclofenac induced visceral gout in vanaraja chicken.

In present study aceclofenac induce group IV treated with febuxostat. It was evident that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja birds. Experimental study in vanaraja chickens shown that significant reduce serum SGPT level after the end of 28 week and 42 treated with Febuxostat. Group IV showed significant decreases as compared to group I (control group) as well as Preventive group III (aceclofenac + Febuxostat). Febuxostat drug decrease serum SGPT in this Group.

Serum glutamic oxaloacetic transaminase (SGOT)

SGOT analysis is an important biochemical diagnostic tool to know the functional status of liver, heart, kidney, brain and skeletal muscle and RBC. It is commonly measured clinically as a marker for liver health. The effect of 28 days administration of aceclofenac drug on AST level to vanaraja chickens present in table.

The Mean ± S.E value of serum SGOT of Vanaraja chicks of Group I, II, III and IV at the end of 7 days were 149.89 ± 6.53, 149.81 ± 2.81, 150.21 ± 7.21 and 149.55 ± 8.72 respectively. No significant differences were observed in groups I, II, III and IV at the end of 7 days.

The Mean ± S.E value of SGOT of Vanaraja chicks of Group I, II, III and IV at the end of 14 days were 150.49 ± 6.08, 216.04±1.72, 151.75±4.87 and 216.07±1.19 respectively. Aceclofenac administered vanaraja chickens Group II and Group IV showed higher significant changes in value of serum SGOT as compared to Group I and Group III. there was no significant mean value of serum SGOT observed in group III as compared to control Group I. administered aceclofenac + Febuxostat in vanaraja chickens group III revealed no significant mean value observed in group III compared to control Group I at the end of 14 days.

The Mean ± S.E value of SGOT of Vanaraja chickens of Group I, II, III and IV at the end of 21 days were151.09±5.14, 220.38±1.91, 151.51±5.37 and 221.02±0.77 respectively. Aceclofenac administered Group II and IV revealed significantly increased values than groups I and III. Febuxostat with Aceclofenac induced Group III revealed no any significant alteration in SGOT value as compared group I. aceclofenac + Febuxostat administered in

vanaraja chickens in group III revealed no significant mean value observed in group III compared to control Group I at the 21 days.

The Mean ± S.E value of serum SGOT of Vanaraja chickens of Group I, II, III and IV at the end of 28 days were146.10 ± 2.56, 242.31 ± 2.27, 151.66 ± 4.39, and 242.81 ± 1.17 respectively, Aceclofenac induced Group II and IV revealed significantly increased values than groups I and III. Febuxostat with aceclofenac induced Group III revealed no any significant alteration in SGOT value as compared group I. aceclofenac + Febuxostat administered in vanaraja chickens in group III revealed no significant mean value observed in group III compared to control Group I at the end of 28 days.

The Mean ± S.E value of serum SGOT of Vanaraja chickens of Group I, II, III and IV at the end of 42 days were 148.29±4.26, 151.33±4.12, 150.11±4.82, and 149.49±2.83 respectively. Bottle gourd treated group II and febuxostat treated group IV No significant differences were observed in groups I, II, III and IV at the end of 42 days.

Aceclofenac administered vanaraja chickens Group II and Group IV showed higher significant changes in value of serum SGOT as compared to Group I and Group III. there was no significant mean value of serum SGOT observed in group III as compared to control Group I. Aceclofenac + Febuxostat administered in vanaraja chickens in group III revealed no significant mean value observed in group III compared to control Group I at the end of 21 days.

In present experimental study, increased value of Serum SGOT in group II and Group IV at end of 14 days, 21 days 28 days during the present study revealed that aceclofenac induce affect liver function in term of increase in serum SGOT levels. Increased in the activity of SGOT in aceclofenac induce group II and group IV may be due acute and chronic liver injury. Our finding agreement with Chandra *et al.,* (1984), Similar finding is also reported by Garcia 1998 and Alarcon *et al.* (2002). Jain *et al.* (2009), Singh *et al.* (2013), Kausar and Waris (2016) reported that found increased level of ALT in producing gout by given oral dose of uric acid in rat.

In present experimental study aceclofenac induce group II treated with bottle gourd juice, it is observed that bottle gourd juice have good effect on aceclofenac induced visceral gout produced in vanaraja chickens. Experimental study on vanaraja birds shown that significant reduce serum SGOT after the end of 28 says to 42 days treated with bottle gourd

Table. 18: Mean ± S.E. values of SGOT (U/L) of Vanaraja chickens in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	149.89±6.3[a]	150.49±6.08[a]	151.09±5.14[a]	146.10±2.56[a]	148.29±4.26[a]
Group II	149.81±2.1[a]	216.04±1.72[b]	220.38±1.91[b]	242.31±2.27[b]	151.33±4.12[a]
Group III	150.21±7.1[a]	151.75±4.87[a]	151.51±5.37[a]	151.66±4.39[a]	150.11±4.82[a]
Group IV	149.55±8.2[a]	216.07±1.19[b]	221.02±0.77[b]	242.81±1.17[b]	149.49±2.83[a]

Superscript a, b, c, d in column indicates significant difference between the groups within days

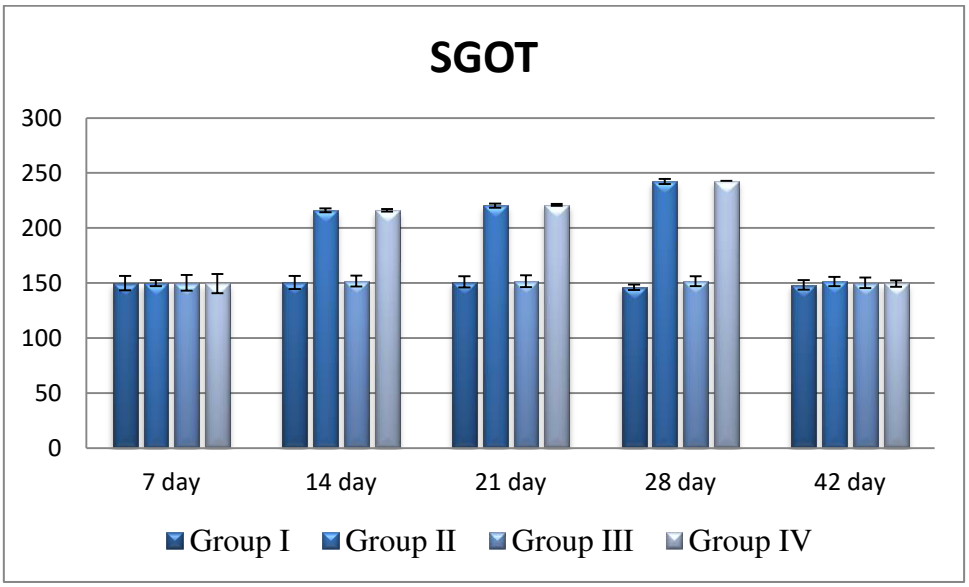


Fig.18: Histogram showing Mean±S.E. of SGOT of (U/L) of Vanaraja in different groups at various time intervals

juice. Group II showed significant decreases serum SG0Tlevel as compared to control group I as well as Preventive group III (aceclofenac + Febuxostat). It was apparent that bottle gourd juice have protective effect on aceclofenac induced visceral gout in vanaraja chicken. Decreased serum SGOT in Bottle gourd juice treated group in vanaraja birds. The decreased level of serum SGOT in other species of animal affected with gout were also reported by Kausar and Waris (2016).

In present study aceclofenac induce group IV treated with febuxostat. It was evident that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja birds. Experimental study in vanaraja birds shown that significant reduce serum SGOT level after the end of 28 days and 42. Treated with Febuxostat. Group IV showed significant decreases as compared to group I (control group) as well as Preventive group III (aceclofenac + Febuxostat). Febuxostat drug decrease serum SGOT in this Group. Decreased serum SGOT in Febuxostat treated group in vanaraja birds. The decreased level of serum SGOT in other species of animal affected with gout were also reported by Patel (2017) reported that treated groups with febuxostat medium and high dose respectively along with diclofenac revealed significant decrease in serum SGOT.

Alkaline phosphatase (U/L)

Alkaline phosphatase is an enzyme of the hydrolase class of enzyme and act in alkaline medium. It is mainly found in liver, biliary tract epithelium and in the bone. The primary importance of measuring alkaline phosphatase is to check the possibility of liver damage or bone disease. When liver, bile ducts and gall bladder system are not functioning properly. This enzyme is not excreted through the bile and alkaline phosphatase is released into the blood stream.

The Mean ± S.E value of alkaline phosphatase of Vanaraja chicks of Group I, II, III and IV at the end of 7 days were 233.01±10.91, 359.37±64.50, 322.43±47.23, and 303.60±21.94 respectively. No significant differences were observed in groups I, II, III and IV at the end of 7 days.

The Mean ± S.E value of alkaline phosphatase of Vanaraja chicks of Group I, II, III and IV at the end of 14 days were 223.14±5.01, 933.20±22.98, 267.96±7.26, and 939.39±20.32 respectively. Aceclofenac induced affected Group II and IV were showed significantly increased values than Groups I and Group III. Aceclofenac + Febuxostat

administered in vanaraja chickens in group III revealed no significant mean value observed in group III as at end of compared to control Group I at the end of 14 days.

The Mean ± S.E value of alkaline phosphatase of Vanaraja chickens of Group I, II, III and IV at the end of 21 days were 268.23 ± 44.40, 1115.50 ± 18.79, 378.06 ± 96.47, and 1179.66 ±19.01 respectively. Aceclofenac induced affected Group II and IV were showed significantly higher values than Groups I and Group III. Aceclofenac + Febuxostat administered in vanaraja chickens in group III revealed no significant mean value observed in group III as compared to control Group I at the end of at the end of 21 days.

The Mean ± S.E value of Alkaline phosphatase of Vanaraja chickens of Group I, II, III and IV at the end of 28 days were 232.27±11.26, 1204.30±28.01, 273.81 ± 21.46 and 1200.75±26.91 respectively. Aceclofenac induced affected Group II and IV were showed significantly higher values as compared Groups I and Group III. Aceclofenac + Febuxostat administered in vanaraja chickens in group III revealed no significant mean value observed in group III as compared to control Group I at the end of at the end of 28 days.

The Mean ± S.E value of alkaline phosphatase of Vanaraja chickens of Group I, II, III and IV at the end of 42 days were 259.29±19.85, 336.25±44.12, 317.81±33.17 and 312.07±43.75 respectively. Visceral gout affected vanaraja chickens belong to group II treated with bottle gourd juice and Group IV treated with Febuxostat drug revealed no significant changes in mean value of compared to control Group I and group III at the end of 42 days No significant differences were observed in groups I, II, III and IV at the end of 42 days.

In present experimental study, increased value of Alkaline phosphatase in group II and Group IV at end of 14 days, 21 days 28 days during the present study revealed that aceclofenac induce affect liver function in term of increase in serum alkaline phosphatase levels. Increased in the activity of Alkaline phosphatase in aceclofenac induce group II and group IV might be due to acute hepatocellular damage and enhance activity revealed renal damage. Our finding agreement with Zimmerman and henby, (1969), Ghodasara *et al.* (2014) Ramzan *et al.* (2015).

In present experimental study aceclofenac induced group II treated with bottle gourd juice, it is observed that bottle gourd juice have good effect on aceclofenac induced visceral gout produced in vanaraja chickens. Experimental study on vanaraja birds shown that

Table. 19: Mean ± S.E. values of AKP of (U/L) of Vanaraja chicken in different groups at various time intervals

Groups	7days	14days	21days	28days	42days
Group I	233.01±10.9[a]	223.14±5.01[a]	268.23±44.40[a]	232.27±11.26[a]	259.29±19.85[a]
Group II	359.37±64.5[a]	933.20±22.98[b]	1115.50±18.7[b]	1204.30±28.0[b]	336.25±44.12[a]
Group III	322.43±47.2[a]	267.96± 7.26 [a]	378..06±96.47[a]	273.81 ±21.46[a]	317.81±33.17[a]
Group IV	303.60±21.9[a]	939.39±20.32[b]	1179.66±19.1[b]	1200.75±26.9[b]	312.07±43.75[a]

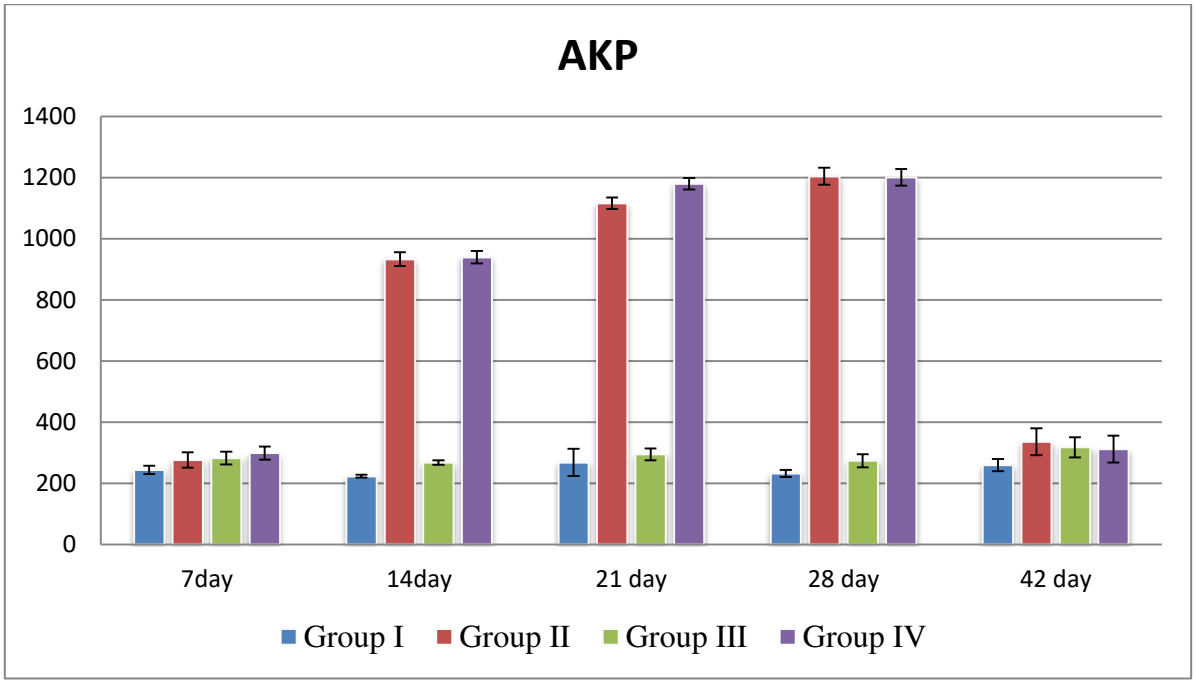


Fig. 19: Histogram showing Mean±S.E. of AKP of (g/dl) of Vanaraja chickens in different groups at various time intervals

significant reduce serum alkaline phosphatase after the end of 28 to 42 days treated with bottle gourd juice. Group II showed significant decreases serum alkaline phosphatase level as compared to control group I as well as Preventive group III (aceclofenac + Febuxostat). It was apparent that bottle gourd juice have protective effect on aceclofenac induced visceral gout in vanaraja chicken. Decreased serum alkaline phosphatase in Bottle gourd juice treated group in vanaraja birds. In the absence of available literature the value of alkaline phosphatase further indicate that bottle gourd juice decreased alkaline phosphatase concentration which was increased due to aceclofenac toxicity, their efficacy in reducing alkaline phosphatase level was almost similar.

In present study aceclofenac induce group IV treated with febuxostat. It was evident that febuxostat have protective effect on aceclofenac induced visceral gout in vanaraja birds. Experimental study in vanaraja birds shown that significant reduces serum alkaline phosphatase level after the end of 28 days and 42. Treated with Febuxostat. Group IV showed significant decreases as compared to group I (control group) as well as Preventive group III (aceclofenac + Febuxostat). Febuxostat drug decrease serum alkaline phosphatase in this Group. Decreased serum alkaline phosphatase in Febuxostat treated group in vanaraja birds. The decreased level of serum alkaline phosphatase in other species of animal affected with gout were also reported by Patel (2017) reported that treated groups with febuxostat medium and high dose respectively along with diclofenac revealed significant decrease in serum AKP.

Pathological changes

Visceral gout affected vanaraja chicken were died during the experiment and dead birds were necropsied and subject to pathological observation. Similarly surviving birds were sacrificed at the end of experiment and necropsied for pathological investigation.

Gross pathology

Vanaraja chickens in group I (control) and group III (preventive Group) which were sacrificed at the end of 42 days did not show any gross pathological changes. The vanaraja chickens administered aceclofenac in Group II Showed on 21 day haemorrhage, chalky white urate deposition on serous surface all organs (fig.4). Group IV were administered aceclofenac showed vanaraja chicken on 21 day showed deposition of urate crystal and congestion in kidney and liver (fig. 5), In Group II, on 28 day showed deposition of urate crystal in

pericardium, congestion in chalky white urate deposition were also deposition all over the visceral organ in kidney (fig.6), In Group II, on 28 day showed deposition of urate crystal in pericardium, congestion in chalky white urate deposition were also deposition all over the visceral organ in kidney, liver and heart. (fig.7) Gross pathological lesions of affected vanaraja chickens of the present study, our finding agreement with Gilbert *et al.* (2002), Seema (2008), Irtaza *et al.* (2008), Undhad (2012), and Patel *et al. (*2013),

At the end of experiment 28 days, decreased severity of lesions was observed in group II and group IV. On 42th day onwards improvement observed in the gross lesions in visceral gout affected group II and group IV, (fig. 12, fig. 13) the improvement due to bottle gourd juice and Febuxostat drug. That has both anti-inflammatory and antioxidant properties.

Kidney

Gross pathology and histopathology

In group I and group II, there were no notifiable changes throughout the experimental period. And Vanaraja chickens from group II and group IV died during the experiment noticed typical gross lesions of visceral gout in the kidney. The affected kidney vanaraja chicken(Group IV) on 14 day showed enlarged frosted and white chalky urate deposition on kidneysurface congestion and pin point haemorrhage .the interstitial tissues showed congestion, oedema, haemorrhage, glomerular changes included thickening of Bowman's capsule and proliferative glomerulonephritis (fig.9) Microscopical lesions in the tissue section of kidney from vanaraja chickens died during experiment in group II and group IV revealed varying degree of haemorrhage, congestion, necrotic and degenerative changes along with urate deposition in renal parenchyma. The kidney section s from vanaraja chicken died in group II and group IV observed lesions like congestion, inter tubular haemorrhage and degeneration, enlarged and frosted chalky white urate deposition with haemorrhage on surface cystic dilation of tubules and marked presence of deposition urate crystal in radiating pattern.Affected vanaraja chicken (group II) kidney showing, severe haemorrhage, congestion, and deposition of urate (14day) H&EX100 (fig.11). In some kidney section mild to moderate infiltration of mononuclear cells was also observed. In group Group II on day 14 vanaraja chicken kidney showed, severe haemorrhage, congestion, and deposition of radial pattern urate crystals H&E X 400.(fig.13) and in Group IV on day 14 vanaraja chicken kidney showed, severe haemorrhage, congestion, and deposition of radial pattern urate

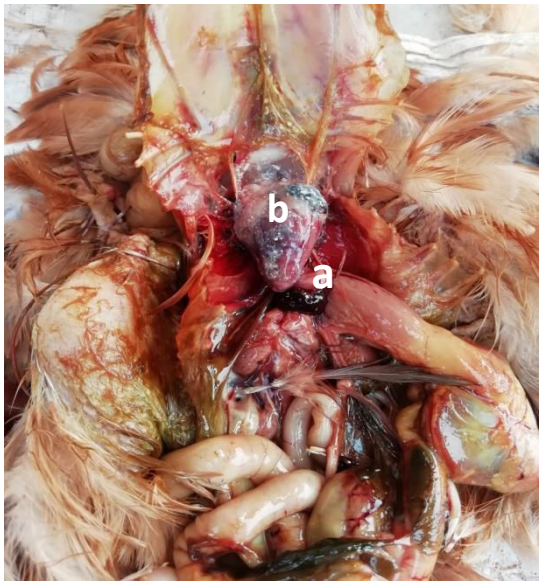


Fig.4.Photograph of vanaraja chicken (Group II, 21 day) showing (a) haemorrhage, (b) chalky white urate deposition on serous surface all organs

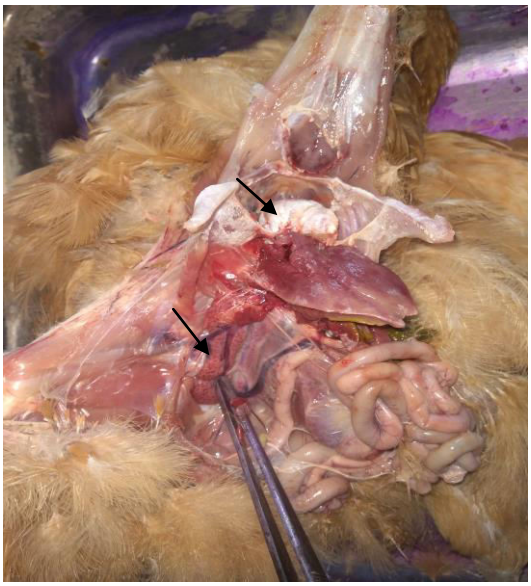


Fig..5. Photograph of vanaraja chicken (Group IV 21 day) showing congestion and haemorrhage in kidney and liver

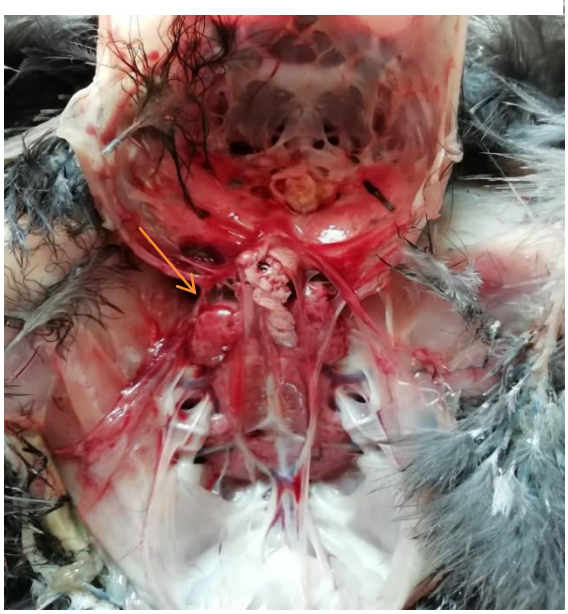


Fig .6 Photograph of vanaraja chicken (Group II, 28 day) showing enlarged frosted and white chalky urate deposition on kidney

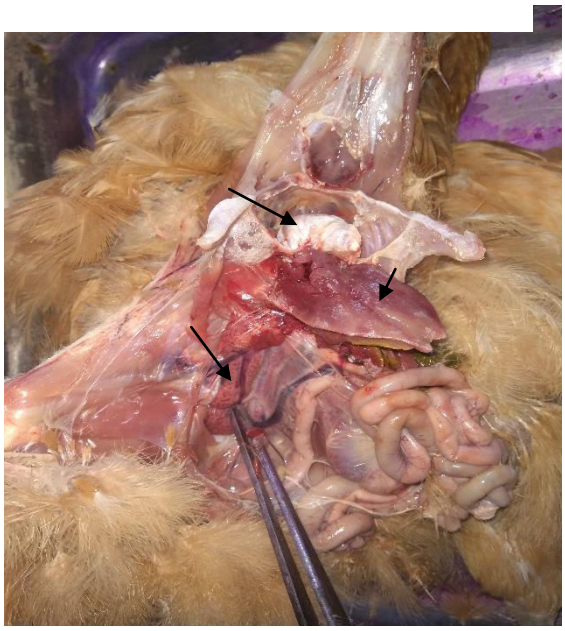


Fig..7.Photograph of vanaraja chickens (Group IV, 28 day) showing deposition of urate crystal in pericardium congestion in kidney, heart and liver

crystals H&EX400. (fig.14), in Group IV vanaraja chicken heart showed urate crystal deposition, severe haemorrhage and congestion.Deposition of urate crystal in renal parenchyma. Which appeared black with De Galantha'stain X100s (fig.19).in group IV on 21 day vanaraja chicken kidney showed black needle shape crystal De Galantha's stain X100 (fig.20). The vanaraja chickens both from group I and group III which were sacrificed at the end of the experiment did not showed lesions.

Vascular lesions of haemorrhage and hyperemia along with tubular degeneration in the cases of visceral gout were similarly described by Chandra and Singh (1980), Uma *et al.* (1999), Hedaoo *et al.* (2008), Ghodasara *et al.* (2014) during their study.

Rahamathulla and Mohiyuddeen (1973), Nayak *et al.* (1988), Meteyer *et al*., (2005), Shinde (2007) and Seema (2008) during their study in case of visceral gout reported degeneration, congestion, hemorrhages, and necrotic changes of kidney.

Deposition of urate crystal in renal parenchyma which appeared black with De Galantha's staining reported by Mulcay *et al.* (2003) Meteyers *et al.* (2005) and Madhuri *et al. (*2008)

The kidney lesions in birds affected with visceral gout showed deposition of homogeneous amorphous urate crystals and showing radiating crystalline pattern (tophi) in the parenchyma and in interstitial tissues of kidney associated with inflammatory reaction were also reported by Iyer (1941), Hurd (1965), Christopher (1977), Kaushik and Kalra (1979), Shultz *et al.,* (2004), Meteyer *et al.,* (2005), Mir *et al*., (2005), Patel*et al*., (2007), Irtaza *et al*., (2008), Seema (2008), Jana *et al*., (2009), Naidoo *et al.,* (2009), Rahul (2010) Muhammad *et al.,* (2012), Ghodasara *et al.,* (2014) and Akhter and Sarker (2015) during their study.

Patel *et al.,* (2014) carried out experiment with aceclofenac on layer chicks and observed lesions congestion, infiltration, enlarged and frosted kidney with chalky white urate deposition, and pin point hemorrhage With De Galantha's stain, the urate deposits give positive reaction which appeared as black needle shaped urate crystal.

The microscopic lesions observed in the kidney in Aceclofenac treated groups II and group IV was specific of visceral gout with deposition of uric acid crystals. The lesions confirmed that the compound aceclofenac can cause visceral gout in vanaraja chicken when given in dose of 20 mg/kg vanaraja chickens through oral gavage. The lesions confirmed that the compound aceclofenac at dose rate of 20 mg/kg bird through oral gavage can cause

visceral gout in vanaraja chickens. This experimental result suggested that high dose aceclofenac produced toxicity in vanaraja chickens

The vanaraja chickens were sacrificed at the end of 42 days. In Group II and Group IV post treatment with bottle gourd and Febuxostat respectively did not revealed any specific microscopic lesions in kidney. The experimental result confirmed that the compound febuxostat did not have any toxicity in vanaraja chickens when given in dose of 4 mg/kg bird through oral gavage. The experimental result confirmed that the compound febuxostat protect the kidney against the visceral gout induced by aceclofenac in vanaraja chickens when given in dose of 20 mg/kg feed.

Liver

Gross pathology and histopathology

Aceclofenac induced vanaraja chickens from group II and group IV died during the experiment noticed typical gross lesions of visceral gout in the liver. The liver section of vanaraja chickens in group II and group IV sacrificed on 14^{th} day, 21^{st} days showed severe haemorrhages, congestion, and necrosis of hepatocytes.The vanaraja chicks (Group II, 14 day) showed, haemorrhage and deposition of urate crystals on liver (fig.8) .In most of the cases, varying degree of chalky white urate deposits covered the serosal surface of liver in chicks died during experiments. There were fatty changes in the periportal area which may be due to increased concentration of uric acid in the blood. Microscopically, Microscopical lesions in the tissue section of liver from vanaraja chickens died during experiment in group II and group IV revealed varying degree necrosis of hepatic cells and infiltration of mononuclear cells with focal deposition of urate crystals. The liver section from vanaraja chicken died in group II observed lesions like congestion, inter tubular haemorrhage and degeneration (fig.15). Group IV the liver section of vanaraja chickens revealed haemorrhage, congestion, degeneration and deposition of urate crystal (fig.16) Deposition of urate crystal in hepatic parenchyma. Which appeared black with De Galantha's stain X100, (fig25, 27, 28), and also observed lesion like urate crystals in radial patterns (fig 26, 29, 30) with De Galantha's stain X400.

Deposition of chalky white urate on the liver in visceral gout were in agreement with findings of Shultz *et al.* (2004), Meteyer *et al.* (2005), Senthil kumar and Thirumurugan (2005), Mir *et al.* (2005), Shinde (2007), Patel *et al.* (2007), Seema (2008), Jana*et al.* (2009), Sharma and Vegad (2010), Ghodasara *et al.* (2014) and Akhter and Sarker (2015)

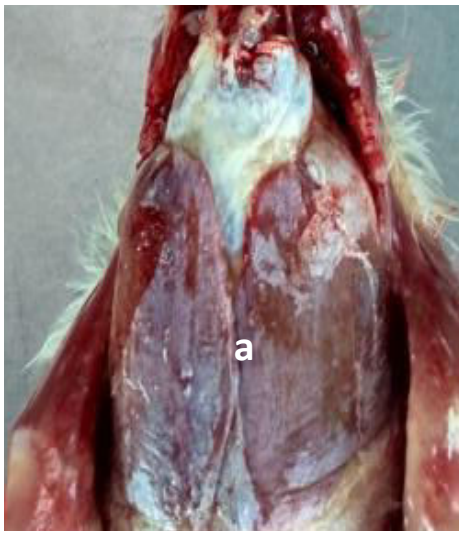


Fig.8 Photograph of vanaraja chicken (Group II, 14 day) showing, haemorrhage deposition of urate crystals on live

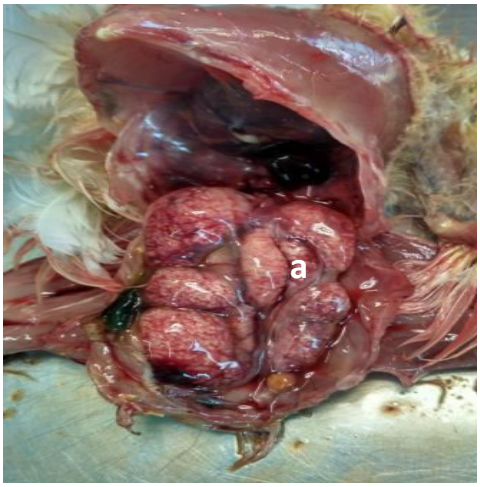


Fig.9 Photograph of vanaraja chicken (Group II, 14 day) showing enlarged and petechial haemorrhage and urate crystal in kidneys

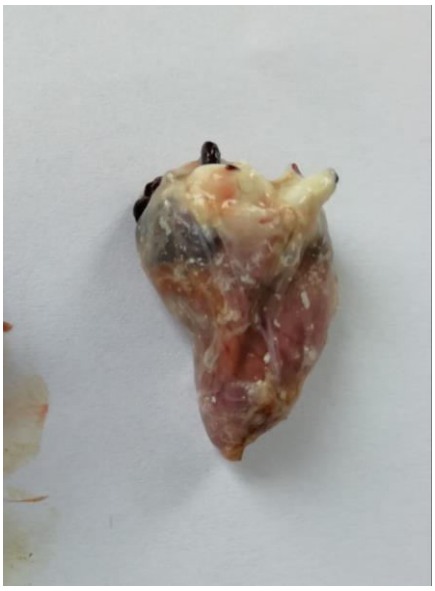


Fig. 10.. Photograph of vanaraja chicken (Group II, 14 day) showing deposition of urate crystals on heart

Zaragoza *et al*. (1995) reported NSAID induced aceclofenac and diclofenac responsible for hepatitis. The liver sections showed mild hydropic degeneration, periportal fibrosis and focal aggregation of lymphocytes.

Similar lesions were also reported by Uma *et al.,* (1997), Seema (2006), Hedaoo *et al.*, (2008), Shinde (2008), Jana *et al.* (2009) and Muhammad *et al.,* (2012) in their studies.

Deposition of urate crystal in renal parenchyma which appeared black with De Galantha's staining reported by Madhuri *et al.,*(2008), Feizi *et al.,*(2011), Auda, (2013), Sathiyaseelan *et al.* (2018), Patel *et al.* (2014).

Patel *et al.* (2014) carried out experiment with aceclofenac on layer chicks and observed lesions congestion, infiltration, with chalky white urate deposition, and pin point hemorrhage in liver With De Galantha's stain, the urate deposits give positive reaction which appeared as black needle shaped urate crystal.

The microscopic lesions observed in the liver in Aceclofenac treated groups II and group IV was specific of visceral gout with deposition of uric acid crystals. The lesions confirmed that the compound aceclofenac can cause visceral gout in vanaraja chicken when given in dose of 20 mg/kg vanaraja chickens through oral gavage. The lesions confirmed that the compound aceclofenac at dose rate of 20 mg/kg bird through oral gavage can cause visceral gout in vanaraja chickens.

This experimental result suggested that high dose aceclofenac produced toxicity in vanaraja chickens. The vanaraja chickens were sacrificed at the end of 42 days. In Group II and Group IV post treatment with bottle gourd and Febuxostat respectively did not revealed any specific microscopic lesions in liver. The experimental result confirmed that the compound febuxostat did not have any toxicity in vanaraja chickens when given in dose of 4 mg/kg bird through oral gavage. The experimental result confirmed that the compound febuxostat protect the liver against the visceral gout induced by aceclofenac in vanaraja chickens when given in dose of 20 mg/kg feed.

Heart

Gross pathology and histopathology

In group I and group II, there were no notifiable changes throughout the experimental period. Aceclofenac induced vanaraja chickens from group II and group IV died during the

experiment noticed typical gross lesions of visceral gout in the heart. Gross lesion of heart showed chalky white urate deposition varying degree along with infiltration mononuclear cells and destruction of myocardial cells. Lesions of heart included congestion, hemorrhage, disruption of myocardial fibers, and edema.(fig.10)

Microscopical lesions in the tissue section of kidney from vanaraja chickens died during experiment in group II revealed vanaraja chicks heart showing mild interfibrillar haemorrhage and congestion (Group II, 14, days) H&E X100 (fig.17) and on 21 days in group IV vanaraja chicken showed urate crystal deposition, severe haemorrhage and congestion in heart H&E X100 (fig.18), vanaraja chickens died during experiment from group II and IV reveled vascular changes comprised of congestion and focal to diffuse hemorrhages between muscle fibers. Urate crystals have been dissolved out of cardiac tissue during processing, the crystalline pattern of their deposition were recognized by destruction of myocytes and infiltration of inflammatory cells mainly mononuclear cells in heart of group II and IV chicks died during experiment and also revealed severe infiltration of inflammatory cells along with urate deposition in pericardium, myocardium and were black in color by De Galantha's stain (fig.22,23,24), The chicks sacrificed at the end of experiment from all groups group did not reveal appreciable changes in heart.

Jana *et al.,* (2009) also observed vascular changes along with urate deposition and cellular infiltration in heart of broiler manifesting visceral gout. Lesions like edema, epicardial thickness and necrosed muscle fibers of heart were also noticed by Jain *et al.,* (2009) during their study on diclofenac in white leghorn birds.

Patel *et al.,* (2014) carried out experiment with aceclofenac on layer chicks and observed lesions congestion, infiltration, severe engorgement of blood vessel in cardiac parenchyma along with chalky white urate deposition, and pin point hemorrhage in heart With De Galantha's stain, the urate deposits give positive reaction which appeared as black needle shaped urate crystal.

Deposition of chalky white urate on the pericardium in visceral gout were in agreement with findings of Shultz *et al.* (2004), Mir *et al.* (2005), Senthil kumar and Thirumurugan (2005), Meteyer *et al.* (2005), Gajera (2006), Naidoo *et al.* (2007), Patel (2007), Shinde (2007), Seema (2008), Jana *et al.,* (2009), Sharma and Vegad (2010), Muhammad *et al.* (2012) and Ghodasara *et al.* (2014). However, Akhter and Sarker (2015), Sndhyarani *et al.* (2019)

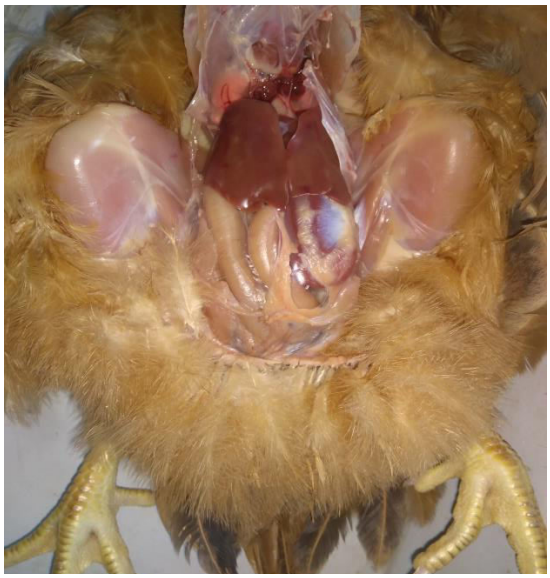


Fig.11. Photograph of Sacrificed vanaraja chicken on 42 day (Group I, 42 day) showing normal appearance of all organs

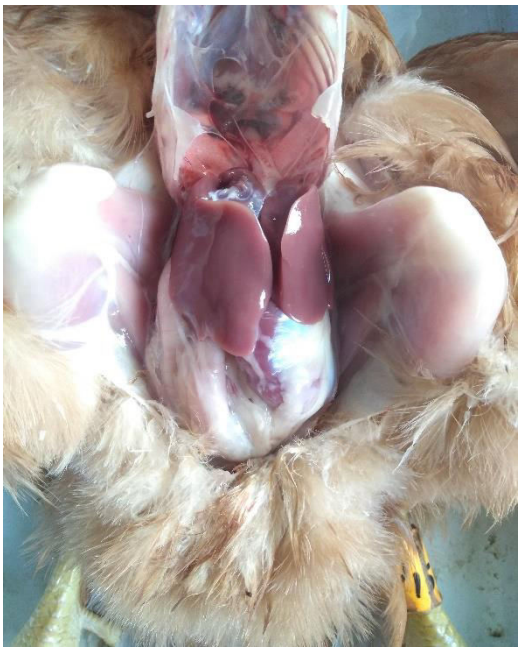


Fig.12.Photograph of Sacrificed vanaraja chicken on 42 day (Group II, 42day) showing normal appearance of all organs

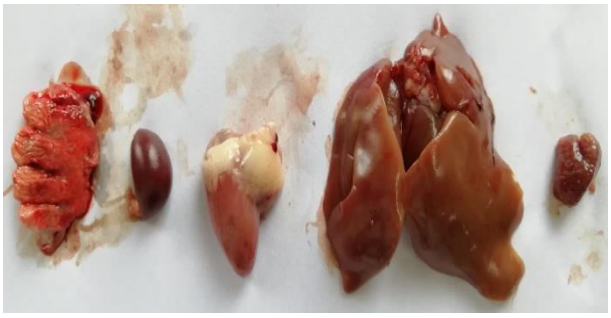


Fig. 13. Photograph of vanaraja chicken (Group III, 42 day) showing normal appearance of all organs

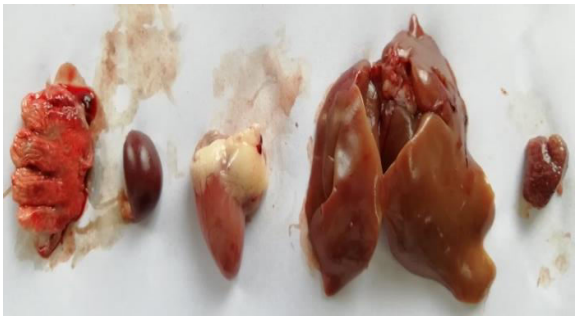


Fig.14. Photograph of vanaraja chicken (Group IV, 42 day,) showing normal appearance of all organs

The microscopic lesions observed in the heart in Aceclofenac treated groups II and group IV was specific of visceral gout with deposition of uric acid crystals. The lesions confirmed that the compound aceclofenac can cause visceral gout in vanaraja chicken when given in dose of 20 mg/kg vanaraja chickens through oral gavage. The lesions confirmed that the compound aceclofenac at dose rate of 20 mg/kg bird through oral gavage can cause visceral gout in vanaraja chickens.

This experimental result suggested that high dose aceclofenac produced toxicity in vanaraja chickens. The vanaraja chickens were sacrificed at the end of 42 days. In Group II and Group IV post treatment with bottle gourd and Febuxostat respectively did not revealed any specific microscopic lesions in liver. The experimental result confirmed that the compound febuxostat did not have any toxicity in vanaraja chickens when given in dose of 4 mg/kg bird through oral gavage. The experimental result confirmed that the compound febuxostat protect the heart against the visceral gout induced by aceclofenac in vanaraja chickens when given in dose of 20 mg/kg feed.

Plates

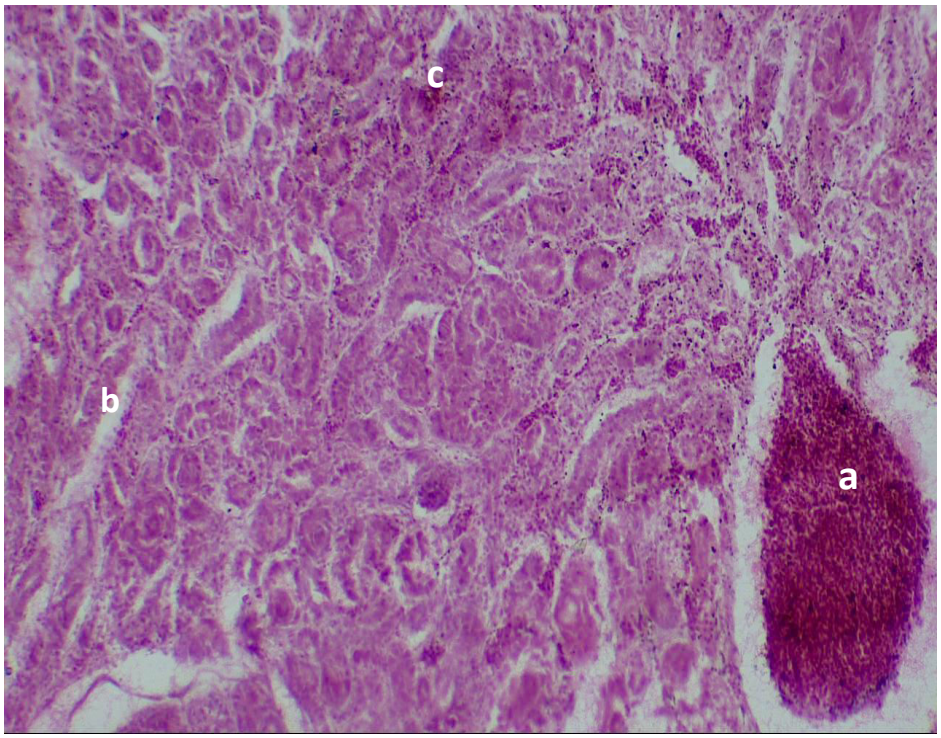


Fig.15..Photograph of vanaraja chicken (group II) kidney showing, severe (a) haemorrhage, (b) congestion, and (c) deposition of urate (14day) H&EX100

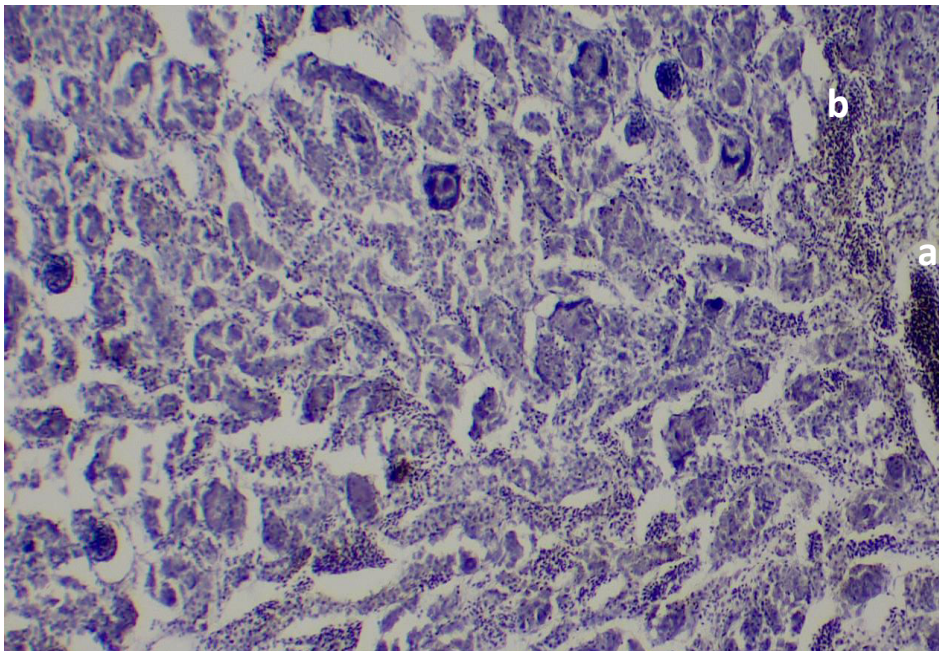


Fig.16. Photograph of vanaraja chicken (group IV) kidney showing (a) haemorrhage and (b) deposition of urate crystal (Group IV, 21days) H&E X100

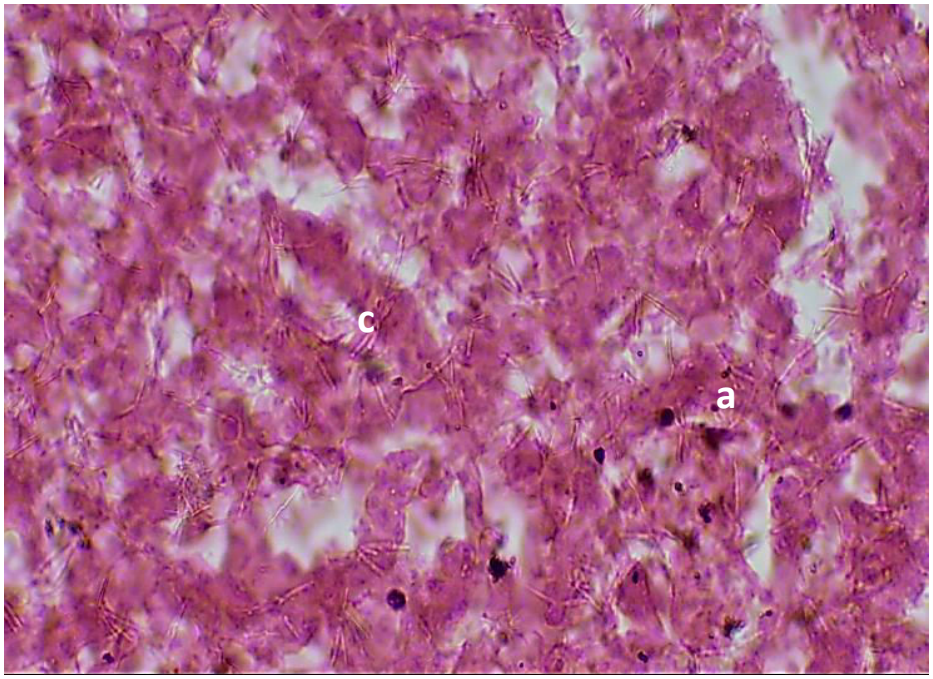


Fig.17 Photograph of vanaraja chicken kidney showing, (a) severe haemorrhage, (b) congestion, and(c) deposition of radial pattern urate crystals (Group II 14day) H&E X 400

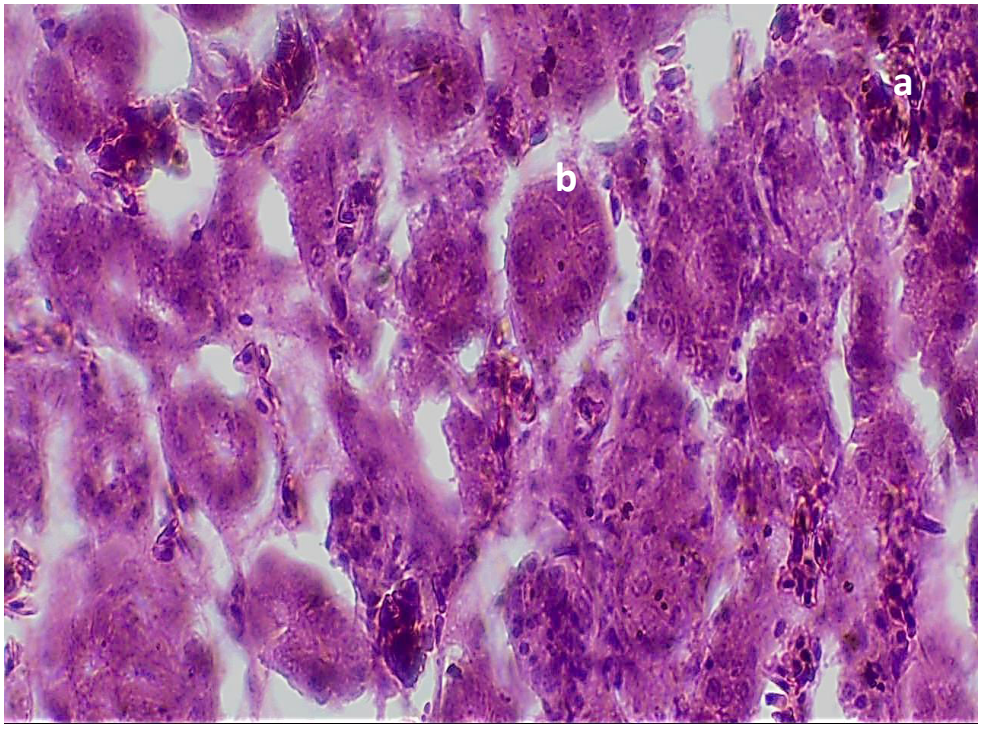


Fig.. 18. Photograph of vanaraja chicken kidney showing (a) haemorrhage and (b) deposition o radial pattern urate crystal (Group IV, 14 days) H&E X 400

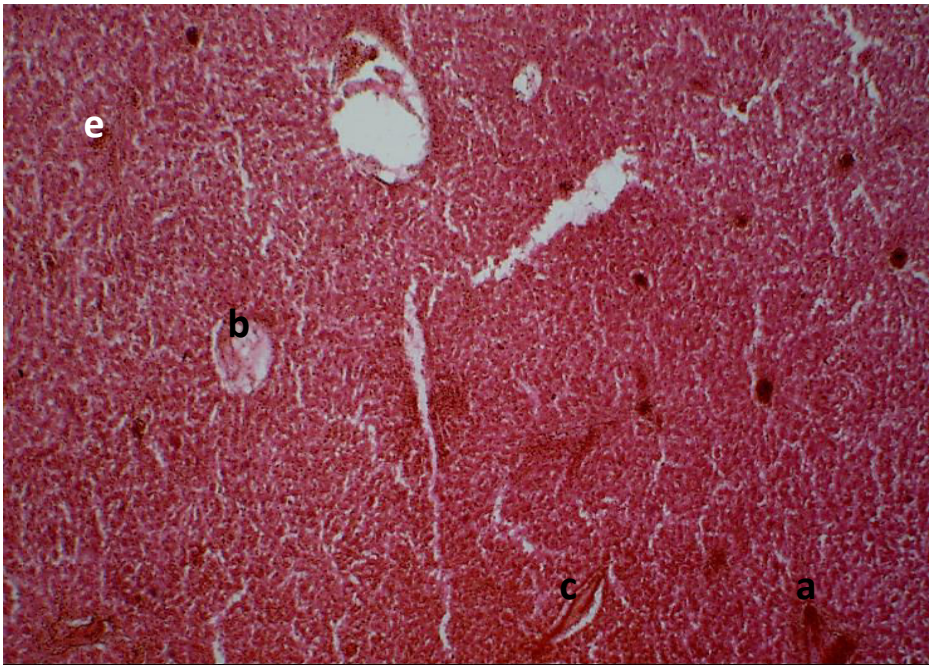


Fig .19 Photograph of vanaraja chicken liver showing, (a) severe haemorrhage, (b) congestion, (c) degeneration and (e) deposition of urate crystal (Group II, 14day) H&EX100

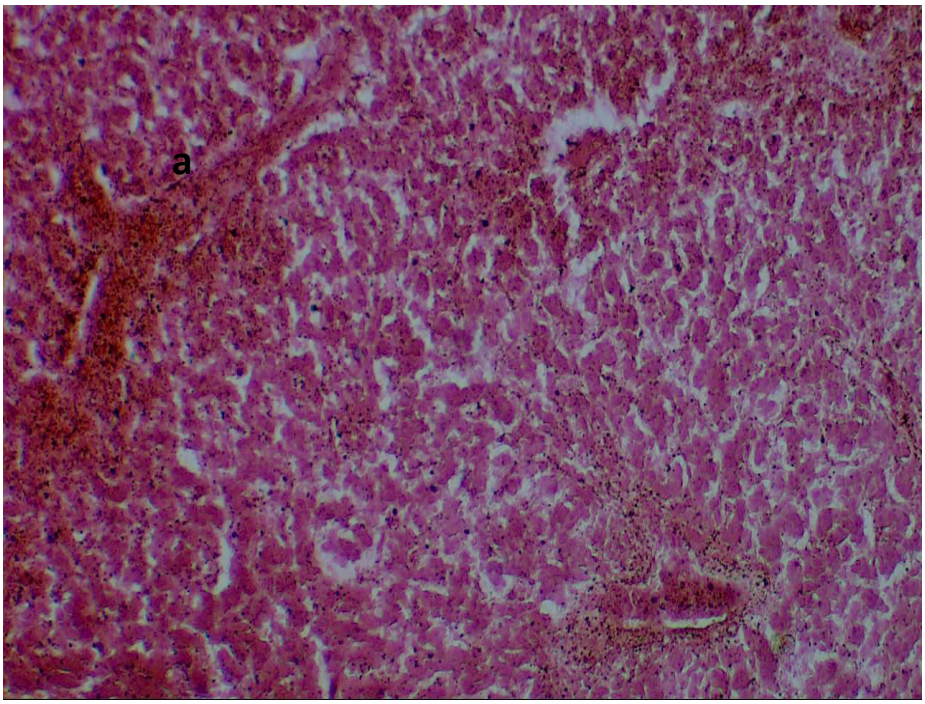


Fig. 20. Photograph of vanaraja chicken liver showing, (a) deposition of urate crystal in hepatic parenchyma (Group IV,21day) H&EX100

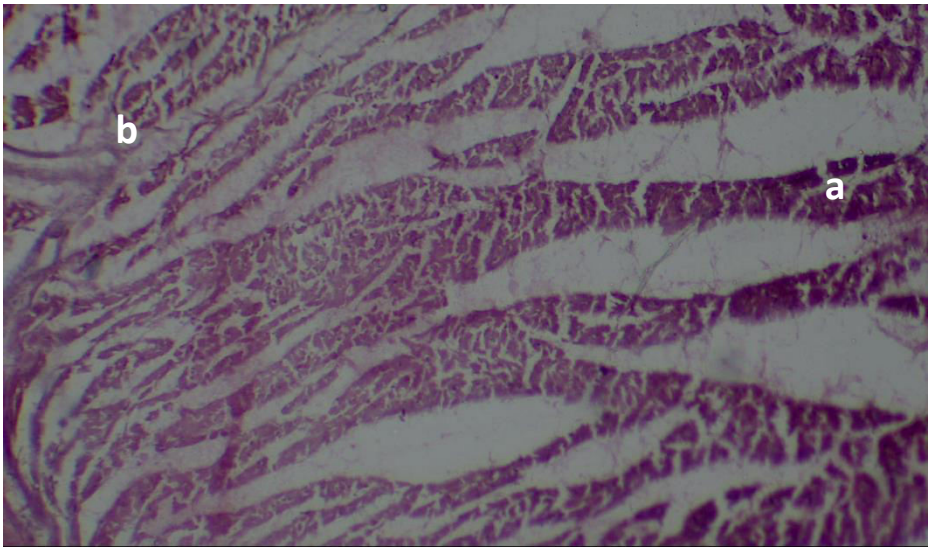


Fig. 21 Photograph of vanaraja chicks heart showing mild interfribllar (a) haemorrhage and (b) congestion (Group II, 14, days) H&E X100

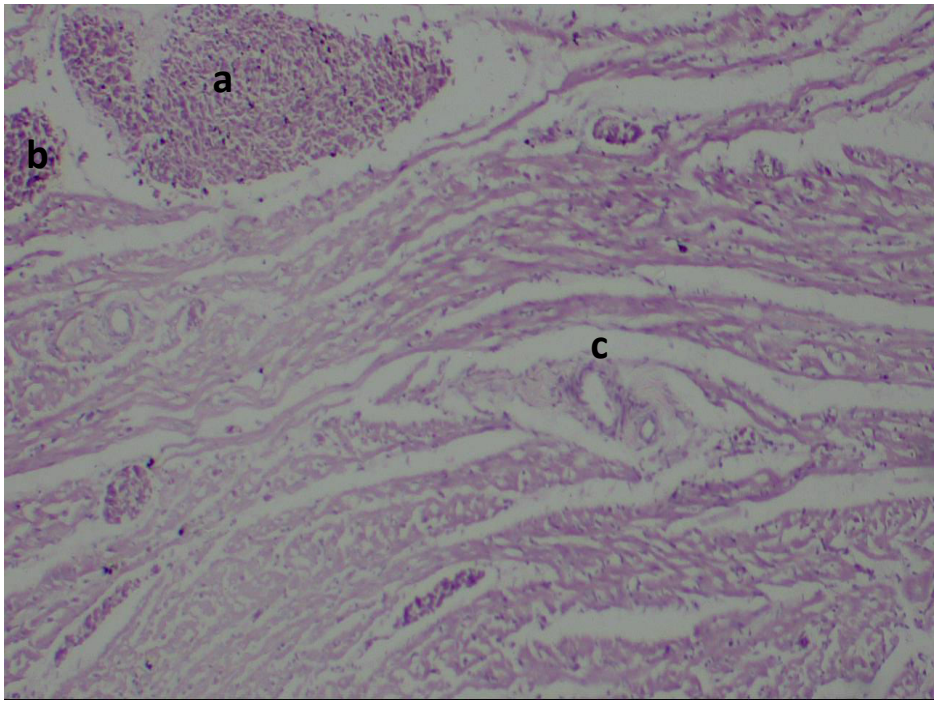
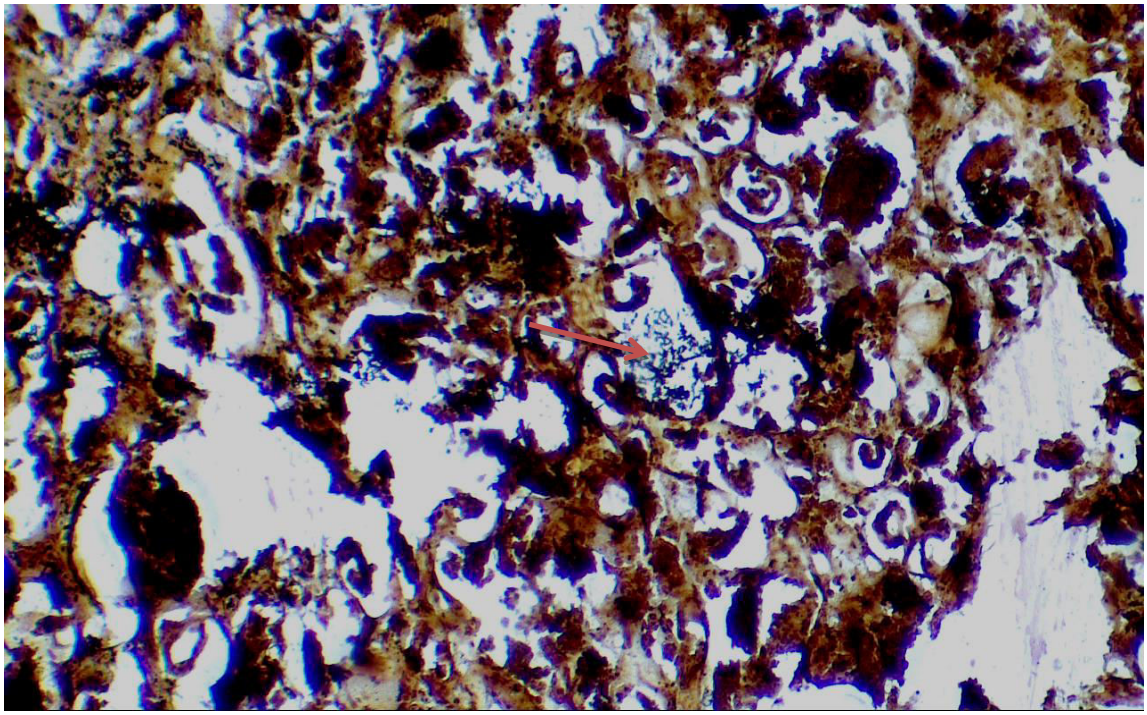


Fig. 22. Photograph of vanaraja chicken heart (a) urate crystal deposition, (b) severe haemorrhage and (c) congestion. Group IV, 21days



Fog.23. Photograph of vanaraja chicken kidney showing black urate crystal in parenchyma (Group II, 14 day) De Galantha's stain X100

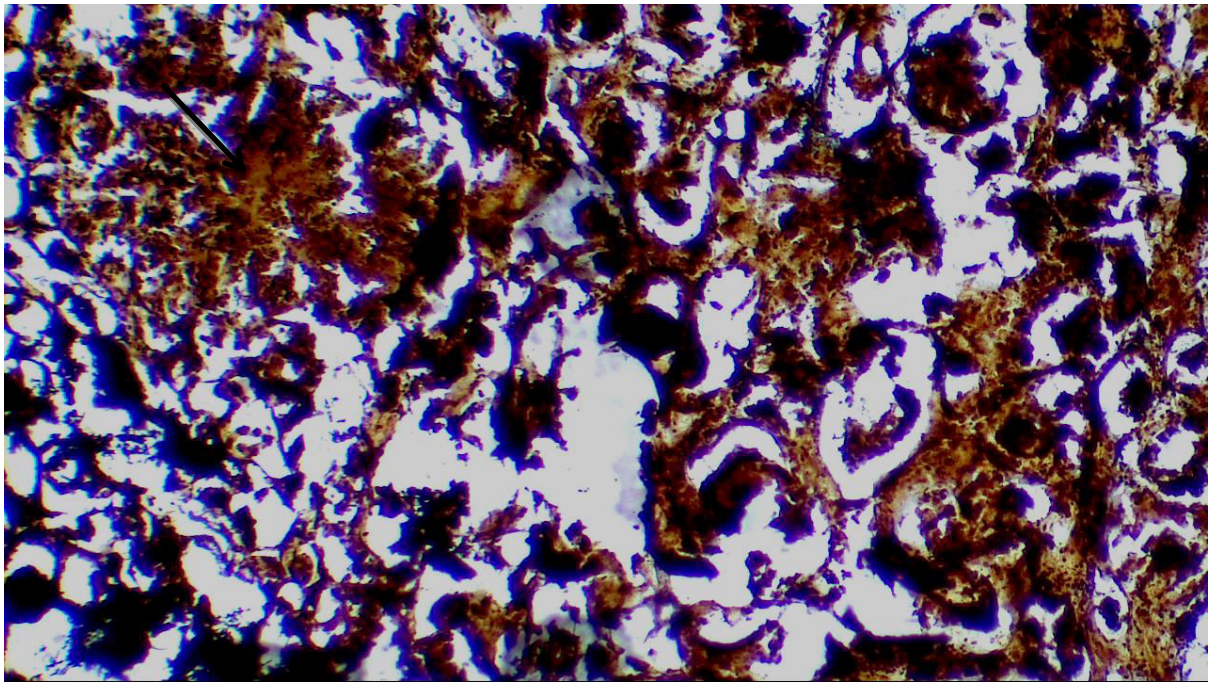


Fig. 24 Photograph of vanaraja chicken kidney showing black needle shape crystal (Group IV, 21day) De Galantha's stain X100

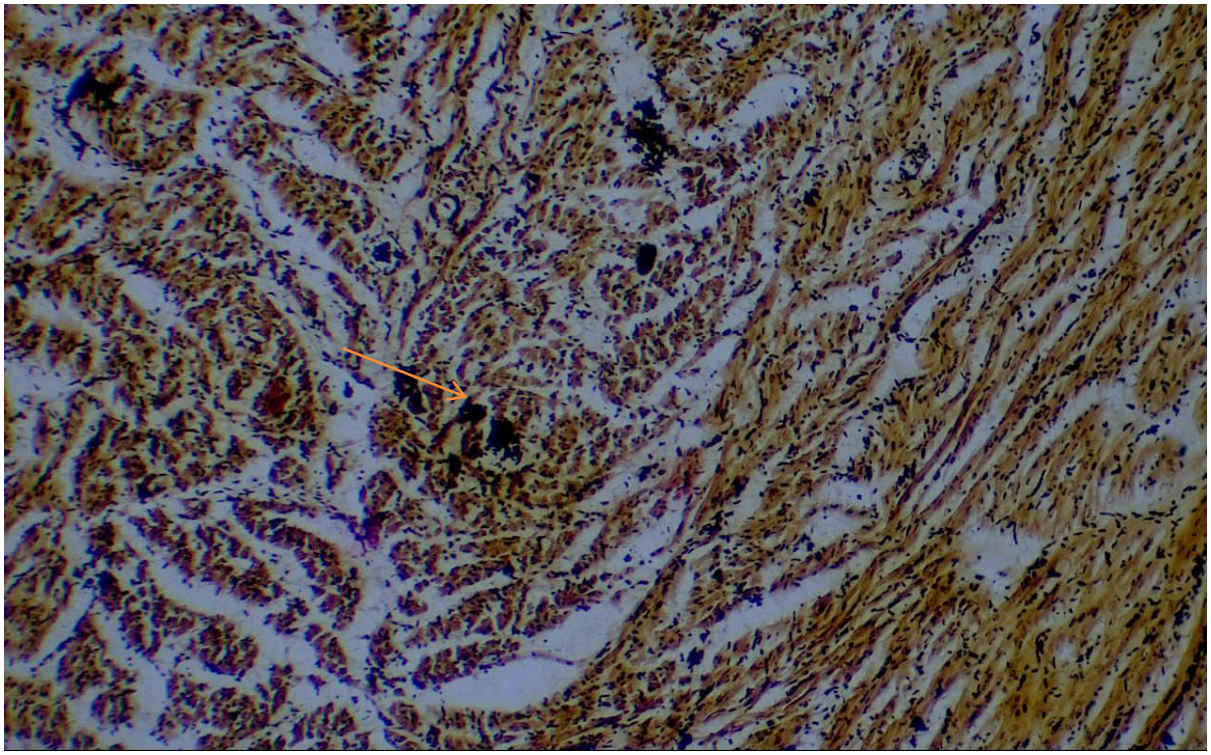


Fig.25. Fig Photograph of vanaraja chicken showing black needle shape urate crystals in heart (Group IV, 21day) De Galantha's stain X100

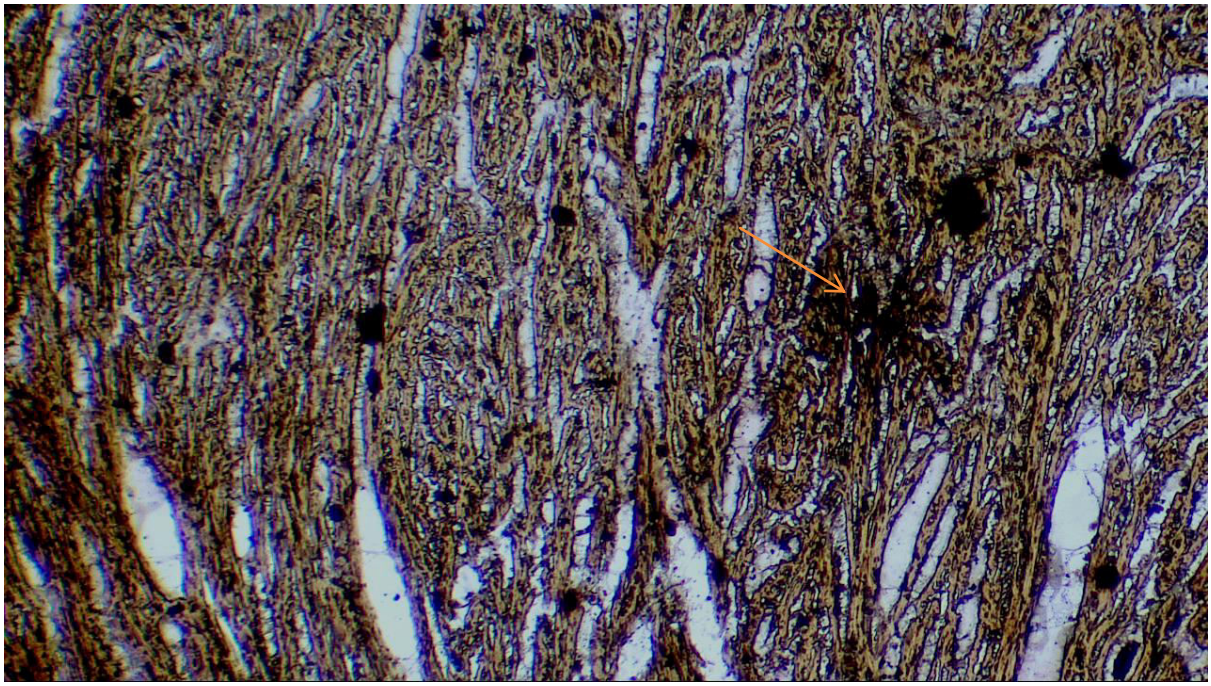


Fig.26. Photograph of vanaraja chicken showing black needle shape crystal in heart (Group IV, 21day) De Galantha's stain X100

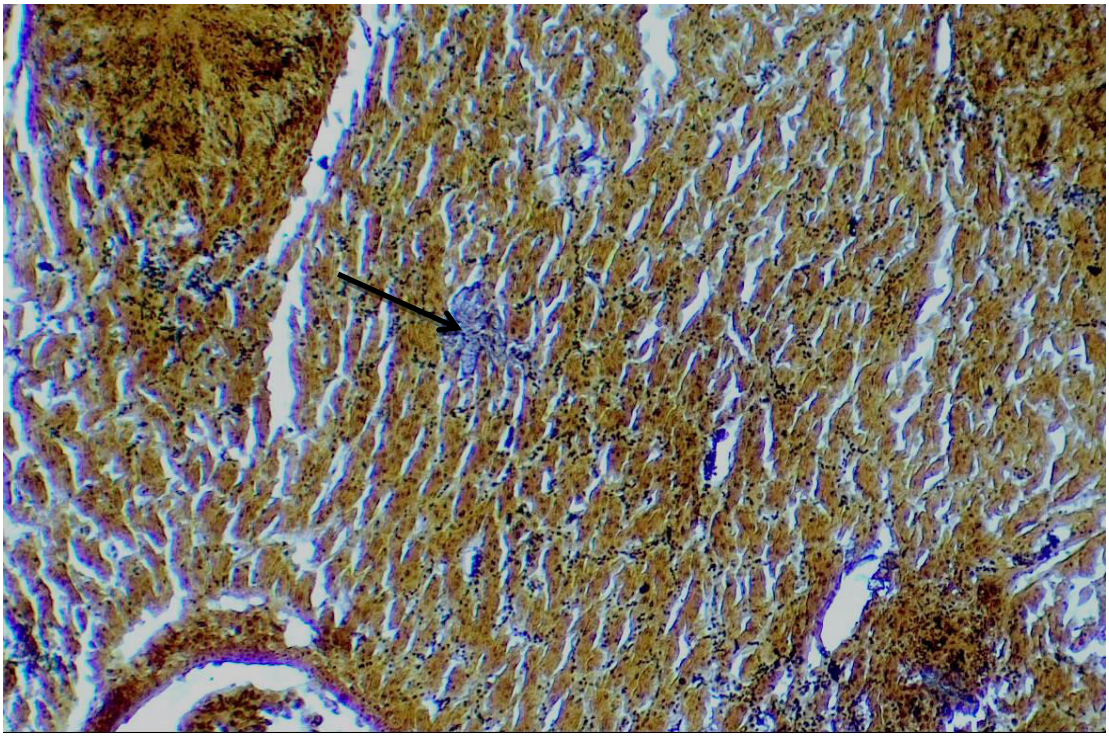


Fig.27. photograph of vanaraja chicken showing black needle shape crystals in liver (Group II, 14day)De Galantha's stain X100

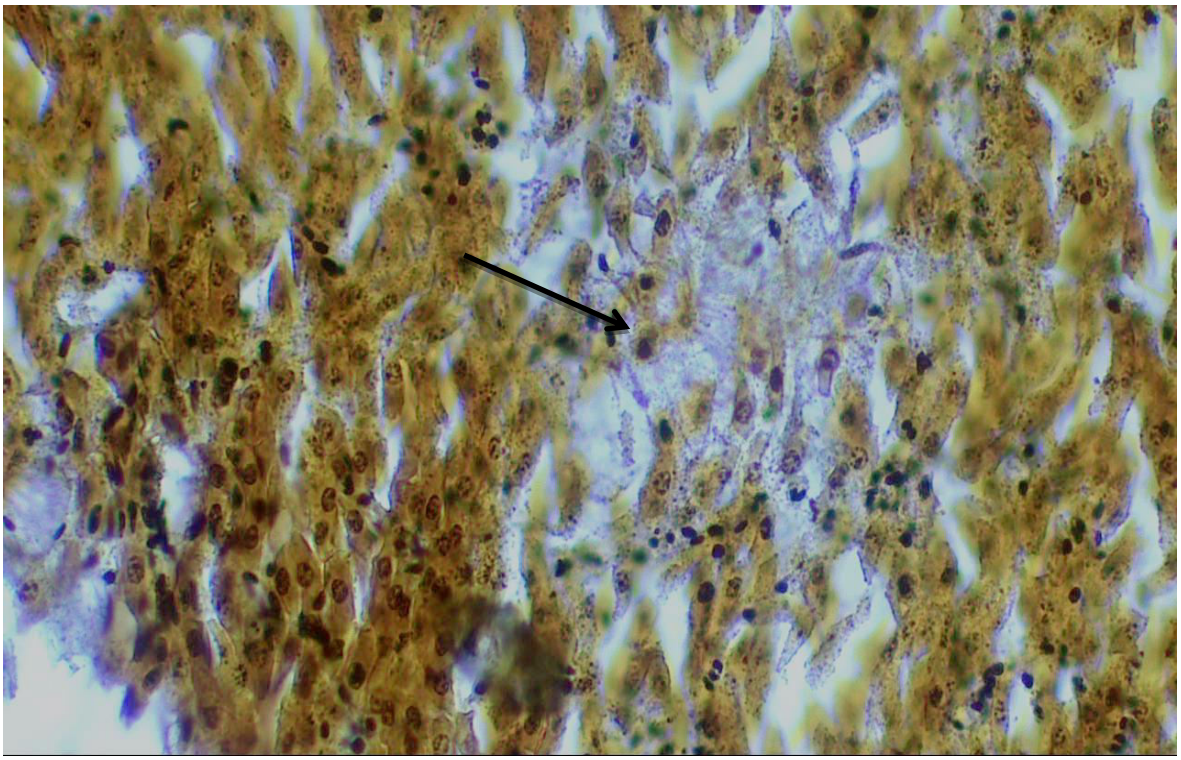


Fig. 28. Photograph of vanaraja chicken showing black needle shape Crystal in liver(Group II, 14 day) De Galantha's stain X400

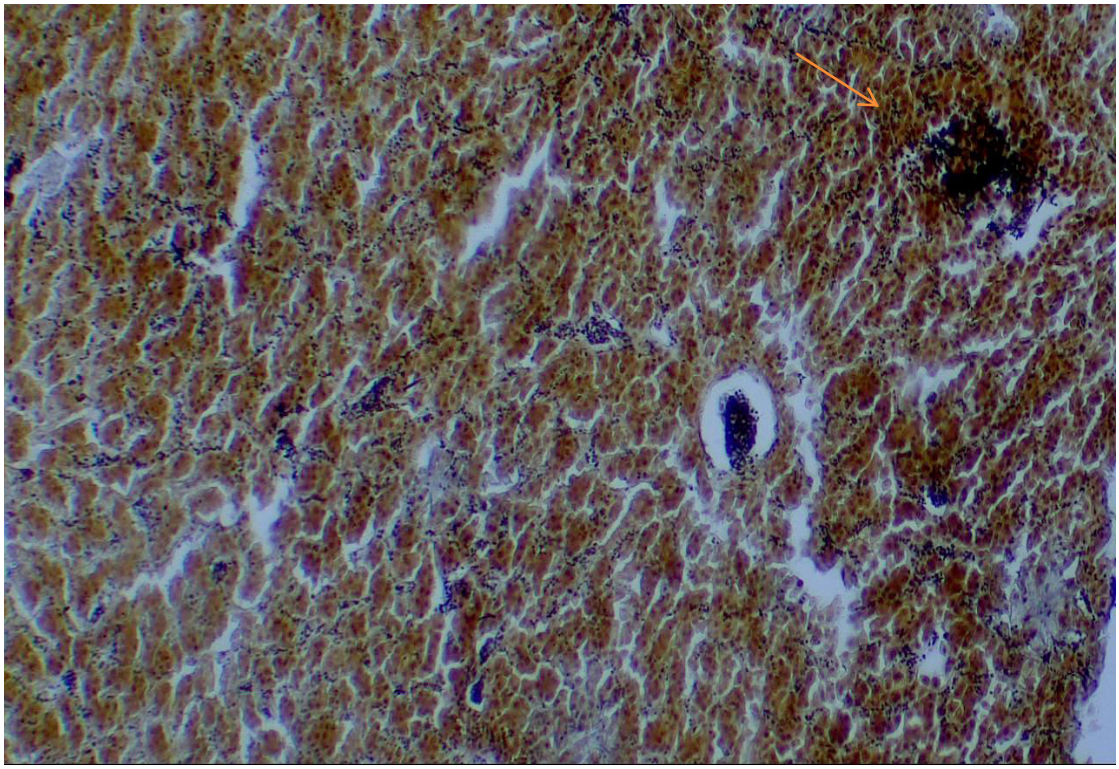


Fig.29 Photograph of vanaraja chicken showing black needle shape in liver

(Group IV, 14 days) De Galantha's stain X100

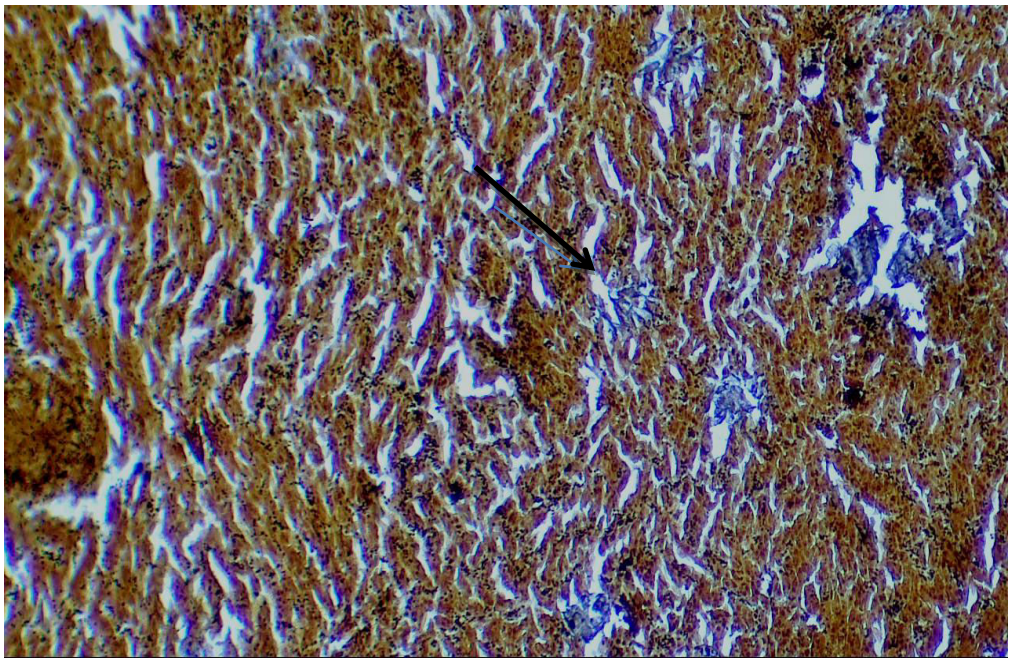
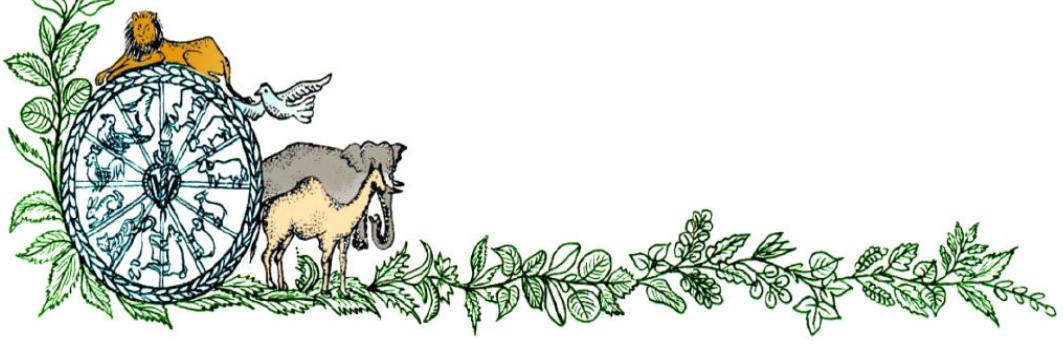


Fig.30 Photograph of vanaraja chicken showing black needle shape crystal in liver

(Group IV, 21day) De Galantha's stain X100

Summary & Conclusion



5 Summary and Conclusion

The present study was carried out to know the "Clinico pathological studies and therapeutic effects of Febuxostat on visceral gout in vanaraja chickens". To undertake these objectives patho-morphological, serum biochemical and hematological were carried out in vanaraja chickens.

Gout is a common metabolic disorder that results in abnormal accumulation of urates in domestic birds. It occurs as two distinct forms, namely, visceral and articular gout. These two syndromes differ in age of onset, frequency, sex predilection, causes, pathogenesis, gross and microscopic lesions.

Visceral gout is a common metabolic disorder characterized by high level of uric acid in the blood lead to deposition of urates on the surface of various visceral organ, in some cases, surface of muscle and synovial sheath of tendons and deposition may also occur on the surface of heart, liver, spleen, kidney and peritoneum appear as white chalky coating. This condition can occur as an individual problem at any age but outbreaks are seen in young chicks in the first and second week of life. Clinical sign such as Chalk-like urate deposits on pericardial sac and liver capsule, Increased thirst (polydipsia), and decreased appetite, abnormal droppings. Chalky urates in their stool, feather plucking, plumage. visceral gout have prime economic important in poultry due to increased incidence causing production loss, regular mortally and lack of availability of specific treatment.

Present Study was conducted on total no of seventy two (72), Vanaraja chickens, Birds were divided into four groups, and each group contain 18 chicks. Experimental trial was started after completions of one week observation period. The experimental study was conducted in 7 day old vanaraja chicks to know the effect of feeding aceclofenac drug with the special interest in causation of visceral gout and as well as role of Febuxostat and bottle gourd juice for treatment of visceral gout. Vanaraja chickens of group I was control group given normal feed, Group II and group IV induced with aceclofenac at the dose rate 20 mg/kg vanaraja chicken through feeding continuously for 7 to 28 days. Group III administered aceclofenac (20 mg/kg in feed) with febuxostat (4 mg/kg) vanaraja chickens through oral gavage continuously for 7 to 28 days, The effect of induced aceclofenac in group II and

group IV was assessed by studying the various parameters viz. clinical signs, mortality, weekly body weight, haematological, biochemical and patho-morphological changes.

Control Group I and group III appeared clinically healthy and did not observed typical clinical signs during all the experimental period. Induced aceclofenac group II and group IV showed clinical signs like viz, depression, anorexia, dehydration, ruffled feather, emaciation, lethargy, dooling of the wings, lameness with shrunken eyes, anorexia, and reduced feed intake. During the experiment mortality was observed in induced aceclofenac group II and Group IV 27.77 % and 27.77% respectively. .mortality started from 14 to 28 days. Visceral gout affected vanaraja chickens showed typical lesions and indicate that @ 20 mg/kg through feed continuously for 7 to 28 days can cause mortality and reduce body weight indicate that aceclofenac produced toxicity. In present study group I and group III did not revealed significant difference at end of 14 days, 21 days 28 days. During experiment observed increased value of haematological parameters like Hb, PCV, TEC, TLC and lymphocyte in aceclofenac induced group II and Group IV at end of 14 days, 21 days 28 days causing dehydration, haemoconcentration, metabolic acidosis and uraemia.

The mean value of biochemical parameters like Uric acid, Creatinine, Albumin, Alkaline phosphatase, SGPT, SGOT except TSP were significantly increased in aceclofenac induced group II and group IV at end of 14 days, 21 days 28 days, causing damaging effect on functioning of kidney, liver and heart and other organs and there was significant decreased mean value of Globulin on the 14, 21, and 28 day in group II and group IV in aceclofenac induced group II and group IV due to dehydration. At end of 28 to 42 days visceral gout affected vanaraja chickens in group II was treated with bottle gourd juice (20ml/l) and group IV treated with Febuxostat (4mg/kg), this suggested beneficial effect of bottle gourd juice and Febuxostat in visceral gout affected vanaraja chicken and elimination of urate deposition from the kidney and thus preventing the kidney damage and other organs. Febuxostat and bottle gourd juice treated groups II and VI suggested did not have negative impact on body weight, feed intake and well as safe to use without any side effects in Vanaraja chickens.

The patho-morphological lesions (both gross and microscopic) were mainly observed in vanaraja chickens that died during the experiment from aceclofenac group II and group IV. The vanaraja chickens which were sacrificed at the end of 42 days period of experiment did not reveal any specific gross and microscopic lesions in any of the treatment groups. Grossly, on surface of visceral organs white chalky urate deposits of varying degrees were observed in chicks which died during experiment from aceclofenac induced group II and IV.

Histopathologically, the lesions were characterized by congestion, degeneration, haemorrhage and deposition of uric acid crystals. Kidney was the main target organ affected. Febuxostat and bottle gourd juice treated groups II and VI did not revealed any patho-morphological changes. It could be stated that protective efficacy of febuxostat and bottle gourd juice.

Conclusion

- Aceclofenac administration at the dose rate of 20mg/kg and above for period of 28 days, induced visceral gout in vanaraja chickens.
- Aceclofenac drug, a preferential cyclooxygenase inhibitor, has nephrotoxicity and hepatotoxicity potential inducing visceral gout in vanaraja chickens.
- The severity of aceclofenac appeared to be less than other similar compound like diclofenac and ketoprofen in birds.
- Visceral gout affected vanaraja chicken showed symptom like dehydration, dullness, lethargy, emaciation, anorexia, feather plucking, reduce feed intake and shrunken eyes. Some birds also shows shifting leg lameness and inability to stand.
- Febuxostat can be used for treatment as well as preventive measure for visceral gout.
- Bottle gourd juice can be used for treatment of visceral gout.
- Aceclofenac treatment leads to increase in level of uric acid. Significant difference was observed in SGPT, SGOT, ALP, Serum Creatinine, serum albumin in serum sample of visceral gout affected vanaraja chickens as compared to normal birds.
- The globulin values were observed significant lowered in visceral gout affected vanaraja chickens.
- No significant alteration observed in total serum protein level in visceral gout affected vanaraja chickens.
- Visceral gout affected vanaraja chicken showed pathological lesions like deposition of
- Urate crystal, congestion and haemorrhage in kidney, heart and liver and other organs.

Suggestion of future research work

- Molecular work should be employed demonstrated urate crystal in different tissues.
- Role of fungal toxin in the production of gout should need to be investigate.
- Infectious bronchitis (IB virus) appear to cause gout in poultry.
- There for role of IB virus in gout in poultry required further attentions.
- The mechanism production of gout should be study at the sub cellular level.

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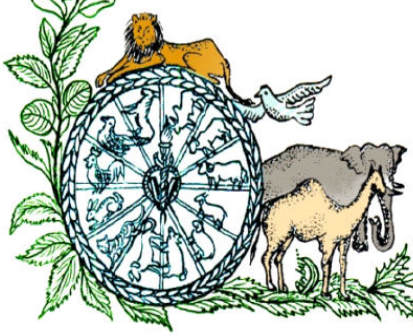

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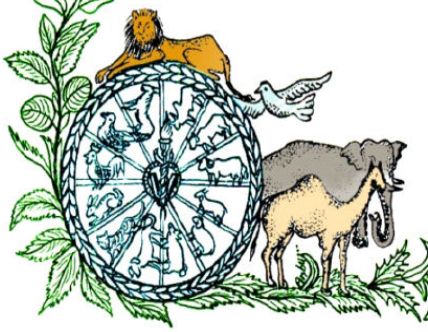
Appendix



7

Appendix

Preparation of commonly used stock solution

Solution	Method of Preparation
1% Acid Alcohol	Hydrochloric acid 1ml, 70% ethanol.-100ml, Mix well
0.2 % Ammonia water	Ammonium Hydroxide-2ml , Distilled water -100ml Mix well
Eosin stain	Pre formed stain purchase from (Nice Chemicals Pvt.Ltd.,)
Haematoxylin	Pre formed stain purchase from market (Nice Chemicals Pvt. Ltd.)
De Galantha's stain	10% Silver nitrate 3.0 ml, 2% hydroquinone 2.0ml, 3% Gelatin10.0 ml, mix well ,

Brief Bio Data

Name Kanchan Rawal

Father's Name late sh. Sita Ram Rawal

Mother's Name Smt. Sita Devi

Date of Birth 15th October, 1991

Permanent Address: Kanchan Rawal D/O late sh. Sita Ram Rawal, 61 housing board distt. Jodhpur, Rajasthan Pin code: 342001

Academic	**Qualification**			
	Month, Year	**School/College**	**Board/University**	**Marks(%)**
10th class	June, 2006	Board of secondary education, Rajasthan	RBSE	49.55
12th class	May, 2010	Board of secondary education, Rajasthan	RBSE	54.00
B.V.Sc.& AH	Jan, 2016	M.B. veterinary college, Dungarpur	RAJUVAS, Bikaner	7.19 (OGPA)