Journal of Pharmacognosy and Phytochemistry
Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2019; SP5: 387-389

Sanjiv Kumar
Ph. D. Scholar, Department of Veterinary Pathology, Ranchi Veterinary College, BAU, Ranchi, Jharkhand, India

MK Gupta
Department of Veterinary Pathology, Ranchi Veterinary College, BAU, Ranchi, Jharkhand, India

Sanjit Kumar
Department of Veterinary Pathology, Ranchi Veterinary College, BAU, Ranchi, Jharkhand, India

Brajesh Kumar
Department of Veterinary Pathology, Ranchi Veterinary College, BAU, Ranchi, Jharkhand, India

Correspondence
Sanjiv Kumar
Ph. D. Scholar, Department of Veterinary Pathology, Ranchi Veterinary College, BAU, Ranchi, Jharkhand, India

(Special Issue- 5)
International Conference on
"Food Security through Agriculture & Allied Sciences"
(May 27-29, 2019)

Studies on alteration of haemato-biochemical parameters by feeding higher doses of febuxostat in grower broilers

Sanjiv Kumar, MK Gupta, Sanjit Kumar and Brajesh Kumar

Abstract
The present study was conducted to explore any toxic effect of febuxostat on certain haemato-biochemical parameters by inducing sub-acute toxicity in broilers. For this purpose twelve grower broilers were housed and maintained in the department of veterinary pathology of Ranchi veterinary college, Ranchi. Six out of them was kept as controlled and rest as treatment group. Treatment group birds received febuxostat orally @ 50mg/kg. b. wt. for seven days. Blood samples were collected at end of experiment for haemato-biochemical studies. Lowered values of urea, creatinine and globulin were observed in birds of the treatment group in comparison to the birds of control group. Haematological studies showed no significant difference in the values of total erythrocyte count between the birds of control group and febuxostat treated group while the value of total leucocyte count increased significantly along with significant increase in the number of heterophil count and decrease in lymphocyte count. Since the effect caused was negligible and variations were in the normal range, it was concluded that febuxostat is a safe drug and might be used in its therapeutic dose.

Keywords: broilers, subacute toxicity, febuxostat

Introduction
Tremendous development of poultry industry resulted in major boost to economic resources and generation of employment in this country. The Poultry is considered as one of the most efficient biological mode of converting the raw food materials into "Productive food" in the form of egg and meat containing high quality animal protein, minerals and vitamins that is required for the maintaining good health of human being. Efficient poultry production greatly depends upon the high survival rate and good health of flocks. The high rate of morbidity and mortality among the fowl result in unsustainable losses to poultry farmer. Now a days poultry birds are genetically modified to meet the increased demands of meat and eggs. Due to lack of awareness the management of poultry flocks is also not upto the mark. This leads to some serious problems in poultry farming leading to decreased production and early death of flocks in affected flocks. Hyperuricemia and resultant gout is one those conditions causing an economic impact on poultry industry and suffering to the birds. Gout is a metabolic disorder where excessive uric acid accumulates in body fluids. Humans, other great apes and birds do not have the ability to convert uric acid into allantoin or ammonia, thus gout is common in them (Eggebeen AT, 2007) [5]. Gout is a systemic disease that results from the deposition of monosodium urate crystals (MSU) in tissues. Increased serum uric acid (SUA) above a specific threshold is a prerequisite for the precipitation of uric acid crystals and tophi formation. Gout occurs if plasma sodium urate concentration exceeds its solubility. As uric acid is excreted *via* tubular secretion, 70% of the kidney is supposed to be malfunctioning to cause hyperuricemia. Apart from renal failure, dietary protein, high calcium content above the bird's requirement might also cause hyperuricemia. In case of renal failure articular or visceral gout occurs. Increased production of UA is responsible for only 10% cases of gout, while the remaining 90% are caused by its renal under-excretion (Mandal AK, 2015). Uric acid production and metabolism are complex processes involving various factors that regulate hepatic production, as well as renal and gutexcretion of this compound. Uric acid is the end

Product of an exogenous pool of purines and endogenous purine metabolism. The physiological role of xanthine oxidase enzyme is to catalyze the terminal two reactions of purine catabolism. In particular XO catalyses the oxidation from hypoxanthine to xanthine and from xanthine to uric acid. Febuxostat [2-[3-cyano-4- (2-methylpropoxy) phenyl)-4-methylthiazole-5-carboxylic acid] sold under the brand names Uloric and Adenuric, is a medication used in the treatment of chronic gout and hyperuricemia. Febuxostat inhibits xanthine oxidase, therefore reducing production of uric acid. Febuxostat inhibits both oxidized as well as reduced form of xanthine oxidase because of which febuxostat cannot be easily displaced from the molybdenum pterin site (Love BL, 2010) [1]. Work on the effect of febuxostat in poultry has shown promising results, but, sometimes overdosing of drug is done by poultry farmers, in lack of proper knowledge. A dose rate of 4-6 mg/kg is found to be effective in broilers (M.K. Patel, 2017) [7]. Therefore, the present work was taken to explore the toxic effect, if any, caused by feeding persistent higher doses of febuxostat in birds.

Materials and methods

Twelve, day old broiler chicks of either sex were procured and housed in the cages kept in the well-ventilated and lighted rooms at the animal house of the Department of Pathology, Ranchi veterinary college. Prior to housing, the poultry room was thoroughly cleaned, white washed and disinfected. The cages of the birds were also washed with 2.5% phenolic solution and disinfected by blow lamping. The birds were vaccinated for combined IB and RD vaccine on 3rd day and IBD on 13th day by naso-ocular route followed by R_2B, vaccine on 25thday.At the age of 25 days they were randomly assigned into two groups. At the time of experiment all the birds were apparently healthy. One group was kept as control while the other group was administered febuxostat orally for seven days. The experimental plan for dose, route of administration of drug and duration of treatment is shown in table 1.

Table 1: Showing the experimental plan of work

Groups	No. of birds and age	Treatment	Route of administration	Duration
C	06 (25 day old)	No Treatment	Orally	7 days
T	06 (25 day old)	Febuxostat @ 50 mg/kg b.wt.	Orally	7 days

At the end of experiment 2.0 ml blood was collected from jugular vein of each birds from both the groups in a clean dry vial containing 2.0 mg ethylene diamine tetra acetic acid (EDTA) as an anti-coagulant for haematological studies. The haematological attributes *viz* Total Leukocyte Count (TLC), Differential Leukocyte Count (DLC), Total Erythrocyte Count (TEC), were carried out following the methodology described by Schalm *et al*., (1975) [8]. As described by Natt and Herick (1952) [10], TEC was estimated by counting 5 secondary squares (four corners and one central) and results were expressed in million/mm^3 while TLC was done by counting cells in 9 primary squares and results were expressed in thousand/mm^3. TLC and TEC were done simultaneously from the same sample charged in haemocytometer counting chamber using erythrocyte diluting pipette and Natt and Herick solution as a diluent.

Differential Leucocyte Count (DLC) was done on blood smears prepared from fresh blood samples and stained with May Grunwald-Giemsa stain following the method described by Lucas and Jamros (1974) [9] with slight modification. DLC was performed by counting a minimum of 100 cells under oil emersion.

For estimation of different biochemical parameters, blood samples were collected from each bird of both groups at the end of the experiment in a clean and dry test tube without using any anticoagulant. The serum samples were separated and centrifuged at 3000 rpm for 10 minutes at room temperature and stored at 0^0C till further analysis. Serobiochemical tests *viz* Serum Creatinine, Serum Urea and globulin were estimated by using tulip's diagnostic kits.

Results and Discussion

The details of biochemical findings have been shown in table 2. Lowered values of urea, creatinine and globulin were observed in birds of the treatment group in comparison to the birds of control group, though the variation was within normal range. Each kidney has millions of small blood-filtering units, called nephrons. The nephrons constantly filter blood through a very tiny cluster of blood vessels *i.e.* glomeruli. These structures filter waste products, excess water, and other impurities out of the blood. Creatinine is one of the substances that kidneys normally eliminate from the body. Because the breakdown of muscle produces creatinine, low muscle mass can result in low levels of creatinine. Similarly urea is synthesized in liver and eliminated through kidneys. Measuring the level of urea and creatinine in the blood provides information about kidney and liver function. High levels may indicate that the kidney is damaged and not working properly. However, reduced plasma level of urea and creatinine has less clinical significance. Globulins are a group of proteins in blood. They are made in liver and immune system. They play an important role in liver function, blood clotting, and fighting infection. Low globulin levels can be a sign of liver or kidney disease. The decrease in level of urea, creatinine and globulin was significant but still within the normal range, therefore it appears that negligible effect has occurred in liver or kidneys due to febuxostat overdose.

Table 2: Showing the values of different biochemical studies:

Parameters	C	T
Urea(mg/dl)	13.63 ± 1.08^{a}	12.66 ± 0.44^{a}
Creatinine (mg/dl)	0.35 ± 0.02^{b}	0.27 ± 0.02^{a}
Globulin (mg/dl)	2.71 ± 0.12^{a}	2.68 ± 0.09^{a}

Significant ($p < 0.05$)

The details of haematological findings have been shown in table 3. Haematological studies showed that there was no significant difference in the values of total erythrocyte count between the birds of control group and febuxostat treated group while the value of total leucocyte count increased significantly. This increase in number of total leucocyte might be due to irritating nature of febuxostat. There was highly significant increase in the number of heterophil count; however, there was significant decrease in number of lymphocyte. Decrease in lymphocyte count may be of relative nature.

Table 3: Showing the values of different haematological studies.

Parameters	C	T
TEC(m/mm3)	3.18±0.05 [a]	3.22±0.14 [a]
TLC (/mm3)	10383±218.20 [a]	12716±298.24 [b]
Differential leucocyte count (%)		
Heterophil	37.83±2.06 [a]	56.50±0.67 [b]
Lymphocyte	54.17±1.74 [b]	34.50±0.43 [a]
Monocyte	3.00±0.37 [a]	3.16±0.48 [a]
Eosinophil	5.00±0.45 [a]	5.83±0.48 [a]

Significant ($p < 0.05$)

Febuxostat was discovered by scientists at the Japanese pharmaceutical company Teijin in 1998. Febuxostat is a non-purine-selective inhibitor of xanthine oxidase. It works by non-competitively blocking the molybdenum pterincenter which is the active site on xanthine oxidase (Bruce SP, 2006 and Yu KH., 2007) [2, 3]. Previous workers also reported rare side effects of febuxostat in humans (Chou HY, 2015) [6]. M. A. Becker *et al.*, 2011 [4] in his experiment also concluded that febuxostat is a safe and potent hypouricemic agent in healthy humans, although not uncommon, adverse events were mild and self- limited, and no deaths or serious adverse events were observed.

The above findings suggest that the side effects of feeding febuxostat in poultry at dose rate of 50mg/b.wt.is negligible and can be recommended for treating gout at its therapeutic dose.

Acknowledgements

The authors feel highly obliged to the Dean, Ranchi Veterinary College, Ranchi for his continuous financial and technical support for conducting the experiment.

References

1. Love BL, Barrons R, Veverka A, Snider KM. Urate-lowering therapy for gout: focus on febuxostat. Pharmacotherapy. 2010; 30(6):594-608.
2. Bruce SP. Febuxostat: a selective xanthine oxidase inhibitor for the treatment of hyperuricemia and gout. Ann Pharmacother. 2006; 40:2187-94.
3. Yu KH. Febuxostat: a novel non-purine selective inhibitor of xanthine oxidase for the treatment of hyperuricemia in gout. Recent Pat Inflamm Allergy Drug Discov. 2007; 1:69-75.
4. Becker MA, MacDonald PA, Hunt B, Gunawardhana L. Treating hyperuricemia of gout: safety and efficacy of febuxostat and allopurinol in older versus younger subjects. Nucleosides Nucleotides Nucleic Acids. 2011; 30:1011-7.
5. Eggebeen AT. Gout: an update. Am Fam Physician. 2015-2007; 76(6):801-8.
6. Chou HY, Chen CB, Cheng CY, Chen YA, Ng CY, Kuo KL. Febuxostat associated drug reaction with eosinophilia and systemic symptoms (DRESS). J Clin Pharm Ther. 2015; 40(6):689-92.
7. Patel MK, Dave CJ, Rathod RC, Joshi BP, Ghodasara DJ. Study on ameliorative Effect of Febuxostat on Gout Induced Model in Broiler Chicks. The Indian Journal of Veterinary Sciences & Biotechnology. 2017; 13(1):46-50.
8. Schalm OW, Jain NC, Carrol EJ. Veterinary Haematology, 3rd edn. Lea and Febiger, Philadelphia, 1975.
9. Lucas AM, Jamros C. Atlas of avainhaematology. Oxford and IBH publishing Co., New Delhi, 1974, 228.
10. Natt MP, Herrick CA. A New blood diluent's for counting the erythrocytes and leucocytes of chicken. Poultry Sci. 1952; 31:735-738.