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**Priya K**
Department of Veterinary Pharmacology and Toxicology, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala, India

**Preethy John**
Department of Veterinary Pharmacology and Toxicology, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala, India

**Saranya N**
Department of Veterinary Pathology, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala, India

**Corresponding Author:**
**Priya K**
Department of Veterinary Pharmacology and Toxicology, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala, India

# Effect of *Eclipta prostrata* L. (L.) leaf powder on gross and histopathology of liver of broiler chicken during aflatoxicosis

**Priya K, Preethy John and Saranya N**

**Abstract**
The study was aimed to investigate the protective effect of *Eclipta prostrata* leaf powder on experimentally induced aflatoxicosis in broiler chicken. Sixty Cobb400 day old broiler chicks were randomly divided into six groups comprising 10 birds in each group. Aflatoxicosis was experimentally induced in all groups except $T_1$ and $T_3$ by giving 500 ppb of aflatoxin $B_1$ ($AFB_1$) contaminated maize from eighth day of age onwards. The group $T_1$ was kept as normal control and $T_2$ as toxic control. $T_3$ was fed with *E. prostata* leaf powder at 0.2 per cent level. The leaf powder of *E. prostrata* was given to $T_4$, $T_5$ and $T_6$ at dose rates of 0.05, 0.1 and 0.2 per cent, respectively. Gross examination displayed enlarged, friable and discoloured liver and histopathological examination of liver showed bile duct proliferation, necrosis, fatty change and leucocyte infiltration indicative of liver damage in $T_2$. In $T_4$ and $T_5$ the similar lesions were noted but to a lesser extent. But in $T_6$ birds apparently normal liver was noted.

**Keywords:** Aflatoxicosis, broiler chicken, *Eclipta prostrata*, gross and histopathology, liver

## Introduction
Aflatoxicosis is a major devastating problem affecting poultry to a greater extend which imposes a huge economic burden upon the livestock farmers. Aflatoxins are secondary metabolites produced by the fungi *Aspergillus flavus* and *A. parasiticus.* Aflatoxins $B_1$, $B_2$, $G_1$, $G_2$, $M_1$, $M_2$ are the different types of aflatoxin and are differentiated by their fluorescence under ultraviolet light. Among these, aflatoxin $B_1$ is considered to be the most toxic and common contaminant in feed. It gets metabolised in liver by cytochrome P450 3A4 enzyme and gets converted to the toxic product aflatoxin $B_1$ - 8, 9 - epoxide. This metabolite intercalates with DNA forming DNA adducts which can be carcinogenic. Aflatoxin $B_1$ has been proven to be hepatotoxic, carcinogenic, mutagenic and to cause immunosuppression as well as oxidative stress in animals.

Aflatoxicosis occurs mainly through feed. About 25 per cent of grains and legumes, that form the vital sources of poultry feed, are estimated to be contaminated with mycotoxins. As per Biomin mycotoxin survey 2017, in South Asia, 81 per cent of feed samples were contaminated with aflatoxin and 64 per cent of samples had aflatoxin above threshold level.

In broiler chicken, the aflatoxin affects liver, kidney, gut morphology, spleen and thymus which lead to reduction in production performances as well as alteration in biochemical parameters. It also causes bruising of carcasses leading to discarding of poultry meat. The aflatoxicosis causes mortality either directly or by lowering the immunity against several other infectious diseases such as Newcastle disease and Infectious Bursal disease.

Since the globe is spanning towards organic livestock production, the use of the therapeutic potentials of plants and plant derived products to curb the toxic effects of aflatoxins is a widely accepted concept.

*Eclipta prostrata* (L.) L. previously called as *E. alba* (known as Kayyonni in Malayalam) belonging to Asteraceae family is reported to have many medicinal properties (Mithun *et al.*, 2011) [1]. Perusal of literature revealed few reports on the protective effect of *E. prostrata* against aflatoxicosis in broilers. Hence, the study was designed with the objective of protective effect of *Eclipta prostrata* (L.) L. leaf powder on gross and histopathology of liver of broiler chicken during aflatoxicosis.

## Materials and Methods

Aflatoxin was produced in maize using the culture *Aspergillus flavus* NRRL 6513 as per the method of Shotwell *et al*., (1966) [2]. The maize culture powder yielded 143.48 ppm of aflatoxin. This mouldy maize was incorporated in experimental feed to arrive 500 ppb of aflatoxin.

The fresh plants of *Eclipta prostrata* were procured locally from Thrissur district of Kerala and were authenticated by Botanical Survey of India (BSI), Coimbatore. The voucher specimen was deposited at the Department of Veterinary Pharmacology and Toxicology, College of Veterinary and Animal Sciences, Mannuthy. The leaves were collected; shade dried and pulverized using an electrical pulveriser. The powdered leaves powder was stored in air tight container at room temperature.

Sixty Cobb400 day old broiler chicks weighing 50 ± 5 g were randomly divided into six groups comprising 10 birds in each group. The birds were maintained under deep litter system and provided with *ad libitum* water and feed throughout the experimental period. All the birds were vaccinated as per the standard schedule. Aflatoxicosis was experimentally induced in all groups except $T_1$ and $T_3$ by giving 500 ppb of aflatoxin $B_1$ ($AFB_1$) from eighth day of age onwards. The group $T_1$ was kept as normal control and $T_2$ as toxic control. $T_3$ was fed with *E. prostata* leaf powder at 0.2 per cent level. The leaf powder of *E. prostrata* was given to $T_4$, $T_5$ and $T_6$ at dose rates of 0.05, 0.1 and 0.2 per cent respectively.

On 42nd day of the experiment all the birds were sacrificed and liver was subjected to gross and histopathological examinations. Liver was observed for gross lesions and recorded. For histopathological examination**,** representative tissue samples of liver were preserved in 10 per cent Neutral Buffered Formalin (NBF). The tissues were processed for routine paraffin embedding and 4-5 micron sections were stained with Mayer's Haematoxylin Eosin stain as per the staining techniques followed by Bancroft and Gamble (2002) [3] to study the histopathological changes.

## Results and Discussions

### Gross Pathology of Liver

On gross examination, no lesions were noted in normal control (Plate 1-A). In aflatoxin control group, enlarged, friable and discoloured liver with blackish spots (Plate 1- B) was seen. Enlarged kidney and spleen, thymus with haemorrhagic spots were also noticed. Enlargement and paleness were also observed in liver of birds treated with *E. prostrata* leaf powder at 0.05 per cent and 0.1 per cent (Plate 1- D, E). No gross pathological lesions were observed in $T_3$ and $T_6$ (Plate 1- C, F).

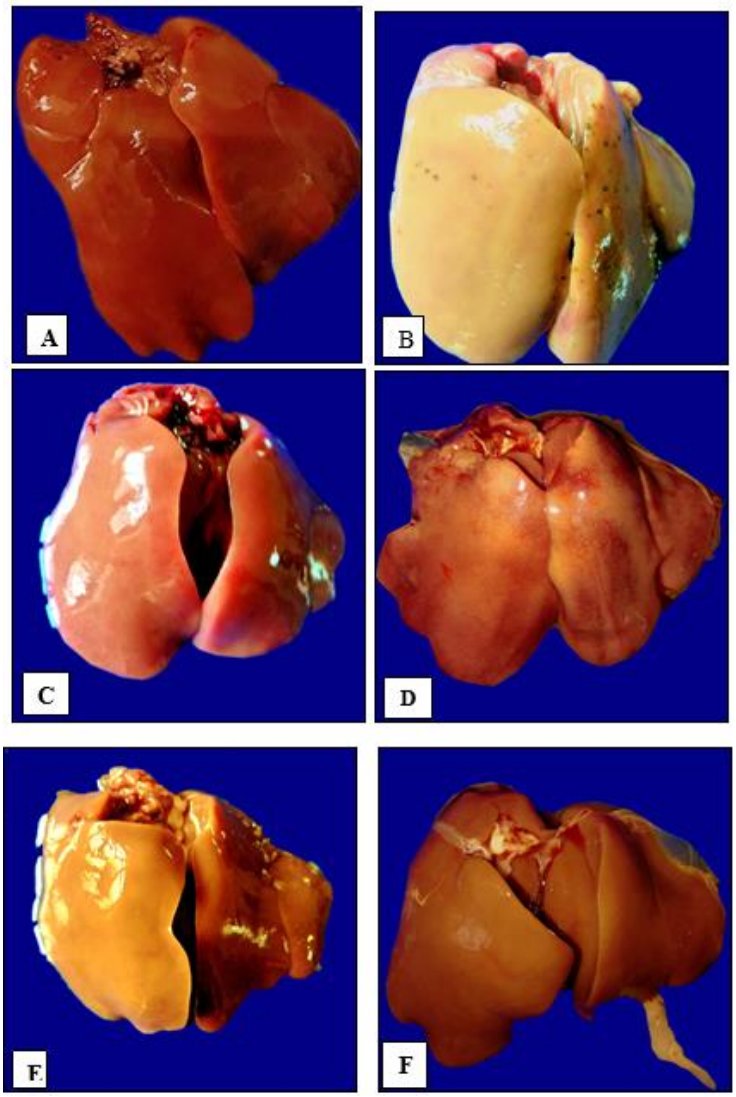


**Plate 1:** Gross pathology of liver (A), (C) and (F) $T_1$, $T_3$ and $T_6$ : Normal liver (B) $T_2$: Pale, enlarged and friable liver (D) $T_4$: Uneven paleness was noticed in liver (E) $T_5$: Mild paleness was noticed in liver

Gross pathological examination of liver from the groups $T_1$, $T_3$ and $T_6$ showed normal appearance and consistency of liver. The group $T_2$ birds displayed enlarged, friable and discoloured liver. Enlarged kidney, splenomegaly and haemorrhagic spots on the thymus were also seen. These lesions were corresponds with the reports of (Hussain *et al.*, 2008) [4]. They conveyed feeding of aflatoxin contaminated feed in broiler chicken exhibited enlarged, discoloured and friable liver and enlarged kidney. (Indresh *et al*., 2013) [5] Reported that enlarged friable and discoloured liver is due to accumulation of lipids in the liver.

Addition of *E. prostrata* powder at the dose rate of 0.05 per cent and 0.1 per cent showed similar lesions with mild intensity in a dose dependent manner. However no gross lesions on liver were found in birds with 0.2 per cent *E. prostrata* leaf powder. This effect is identical with the results of (Ma-ma *et al*., 1978) [6]. They reported that administration of fresh leaf juice of E. *alba* to guinea pig revealed slight pale and congested liver than the normal liver against $CCl_4$ induced hepatoxicity.

**Histopathology of Liver**

On histopathological examination of the liver, apparently normal architecture was noted in $T_1$, $T_3$ and $T_6$ (Plate 2- A, B and Plate 3- D). In toxic control group ($T_2$), liver showed extensive lesions such as fatty change (Plate 2- E), necrosis (Plate 2- D) especially in the periportal areas, bile duct proliferation with increase in number of bile ducts (Plate 2- C), proliferation of bile duct epithelial cells, infiltration of inflammatory cells (Plate 2- F) and apoptotic cells. All these lesions were suggestive of aflatoxicosis in birds. In group $T_4$, mild fatty change, bile duct proliferation, enlarged and degenerated bile duct epithelial cells occluding the lumen (Plate 3- B), congestion of central vein (Plate 3- A) and sinusoids were noticed. In $T_5$, bile duct proliferation (Plate 3- C), congestion and necrosis of hepatic cells were noted to a lesser extend compared with toxic group and $T_4$.

On histopathological examination, the aflatoxin control group showed extensive lesions in liver such as fatty change, bile duct proliferation, periportal hepatic necrosis, proliferation of bile duct epithelial cells, infiltration of inflammatory cells and apoptotic cells. These lesions indicated the definite onset of aflatoxicosis in birds. This result is in consonance with the findings of (Bailey *et al*., 2006) [7] who noticed the similar lesions in the aflatoxin (4 ppm) fed birds. (Hussain *et al*., 2008) [4] noticed bile duct proliferation, fatty change, hepatic necrosis, hemorrhages in hepatocytes and dilatation of sinusoids. Similar lesions was also obtained by (Gowda *et al*., 2008) [8]. In $T_4$ and $T_5$ also, liver showed similar lesions with lesser intensity. The alteration in the lipid transport could be the reason for fatty changes in liver during aflatoxicosis (McLean and Dutton, 1995) [9]. The overexpression of CYP isoform genes (*CYP1A1* and *CYP2H1*) could be responsible for proliferative changes in aflatoxicosis (Yarru *et al*., 2009) [11].

In groups $T_1$, $T_3$, $T_6$ the liver exhibited normal histology. This indicated that *E. prostrata* leaf powder at the dose rate of 0.2 per cent in the basal diet provided better protection when compared with low doses (0.05 and 0.1 per cent). This result is in line with (Lin *et al.*, 1996) [12] who observed that crude alcoholic extract of *E. prostrata* significantly improved the $CCl_4$ and galactosamine induced histopathological changes in liver but not the acetaminophen induced hepatic damage.

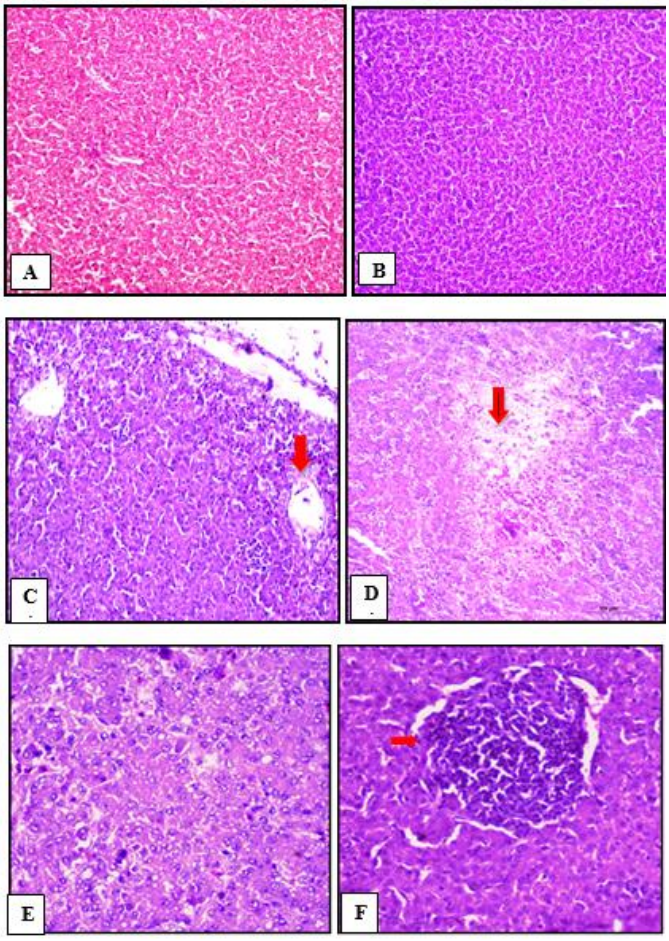


**Plate 2:** Photomicrographs of H & E stained liver sections (A) and (B) $T_1$ and $T_3$: Normal liver(H&E X100) (C) $T_2$: Bile duct proliferation (H&E x 400), (D) $T_2$: Hepatic necrosis (H&E x 200) (E) $T_2$: Fatty change (H&E x 100), (F) $T_2$: Leucocyte infiltration (H&E x 200)

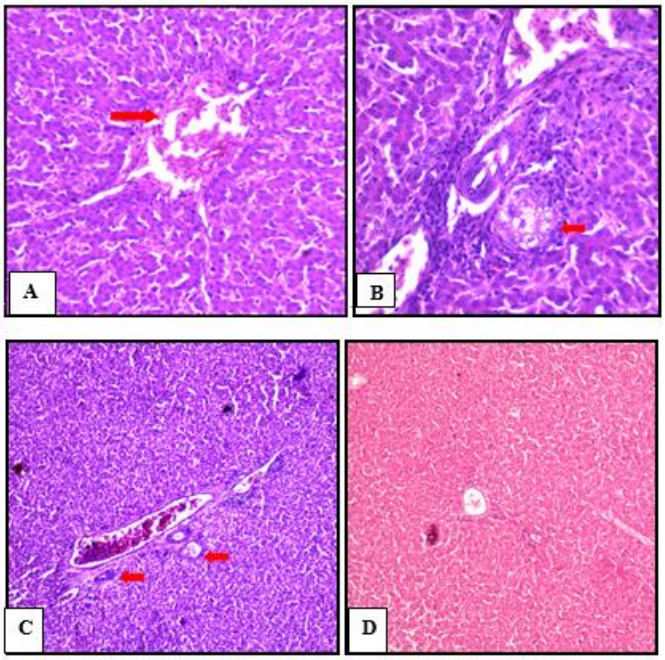


**Plate 3:** Photomicrographs of H & E stained liver sections (A)$T_4$: Central venous congestion (H&E x 200) (B) $T_4$: Bile duct occlusion (H&E x 400) (C) $T_5$: Bile duct proliferation (H&E x 100) (D) $T_6$: Normal liver (H&E x 100)

## Conclusion

The Gross and histopathological examination of liver revealed apparently normal hepatocytes in the birds fed with aflotoxin. The hepatoprotection of *Eclipta prostrata* L. (L.) leaf might be due to induction of hepatic regeneration mechanism and activation of the functions of reticuloendothelial system. Hence, dried leaf powder of *Eclipta prostrata* L. (L.) can be used as hepatoprotective agent in poultry feed.

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## References

1. Mithun NM, Shashidhara S, Vivek Kumar R. *Eclipta alba* (L.) A review on its phytochemical and pharmacological profile. Pharmacology Online. 2011; 1:345-357.
2. Shotwell OL, Hesseltine CW, Stubblefield RD, Sorenson WG. Production of aflatoxin on rice. Applied Microbiology. 1966; 14:425-428.
3. Bancroft JD, Gamble M. Theory and Practice of Histological Techniques. (5th Ed.). Harcout, London, 2002, 181-182.
4. Hussain Z, Khan MZ, Hassan ZU. Production of aflatoxins from Aspergillus flavus and acute aflatoxicosis in young broiler chicks. Pakistan Journal of Agricultural Science. 2008; 45:95-102.
5. Indresh H, Devegowda G, Ruban SW, Shivakumar M. Effects of high grade bentonite on performance, organ weights and serum biochemistry during aflatoxicosis in broilers. Veterinary World. 2013; 6:313-317.
6. Ma-Ma K, Nyunt N, Tin KM. The protective effect of *Eclipta alba* on carbon tetrachloride-induced acute liver damage. Toxicology and Applied Pharmacology. 1978; 45:723-728.
7. Bailey CA, Latimer GW, Barr AC, Wigle WL, Haq AU, Balthrop JE *et al*. Efficacy of montmorillonite clay (NovaSil PLUS) for protecting full-term broilers from aflatoxicosis. Journal of Applied Poultry Research. 2006; 15:198-206.
8. Gowda NKS, Ledoux DR, Rottinghaus GE, Bermudez AJ, Chen YC. Efficacy of turmeric (*Curcuma longa*) containing a known level of curcumin, and a hydrated sodium calcium aluminosilicate to ameliorate the adverse effects of aflatoxin in broiler chicks. Poultry Science. 2008; 87:1125-1130.
9. McLean M, Dutton MF. Cellular interactions and metabolism of aflatoxin: an update. Pharmacology & Therapeutics. 1995; 65:163-192.
10. Yarru LP, Settivari RS, Antoniou E, Ledoux DR, Rottinghaus GE. Toxicological and gene expression analysis of the impact of aflatoxin B1 on hepatic function of male broiler chicks. Poultry Science. 2009; 88:360-371.
11. Lin SC, Yao CJ, Lin CC, Lin YH. Hepatoprotective activity of Taiwan folk medicine: *Eclipta prostrata* Linn. against various hepatotoxins induced acute hepatotoxicity. Phototherapeutics Research. 1996; 10**:**483-490.