

Protective effects of *Erythrina variegata* and *Spirulina platensis* in imidacloprid intoxicated White Leg Horn cockerels

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ABSTRACT

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Forty two white leghorn cockerels were divided randomly and equally into six groups viz. I, II, III, IV, V and VI. Group I served as control and other groups were administered as SP @ 0.2% in II, IMI @ 50 PPM in III, SP @ 0.2% plus IMI @ 50 PPM in group IV, EVLP (1%) in group V and EVLP (1%) plus IMI @ 50 PPM in VI, respectively, for 60 days. Grossly, only kidneys and testes showed lesions in group III only. Histopathological changes such as vascular changes, swelling of the glomeruli and hypercellularity of the glomerular capillaries in kidney; vascular changes, vacuolar degeneration of the hepatocytes throughout the parenchyma and presence of mononuclear cells around congested blood vessels in liver; reduction in size of the seminiferous tubules with a single layer of germinal epithelium in testes and congestion of blood vessels, elongation of neurons, gliosis and satellitosis in brain were observed in IMI treated group. Group VI showed mild pathological lesions while there were no pathological changes in liver, kidney, testes and brain from cockerels of group IV. It was concluded that simultaneous administration of *Spirulina platensis* (0.2%) and *Erythrina variegata* (1%) in imidacloprid intoxicated (@50 ppm) white leghorn cockerels in feed for 60 days produced ameliorating effects on the toxic pathology.

Keywords: Cockerels, *Erythrina variegata*, imidacloprid, Pathology, *Spirulina platensis*, white leg horn

Extensive, inadvertent and indiscriminate use of pesticides has imposed serious health hazards due to their presence in food, water and soil. Imidacloprid, 1((6-chloro-3-pyridinyl) methyl)-N-nitro-2-imidazolidinyl-mine, a neonicotinoid is globally used insecticide for crop protection worldwide from the last decade due to its low soil persistence and high insecticidal activity at low application rate. It is fastest growing in sales as insecticide globally because of its low selectivity for insects and apparent safety for humans¹. Recently imidacloprid has raised concern because of its ability to cause egg shell thinning, reduced egg production and hatching time, which are considered as signs of possible endocrine disrupters^{1,2}. *Spirulina*, a blue-green algae from the sea, is one of the high quality natural feed additives that can be used in animal and poultry nutrition. *Spirulina platensis* has been used for hundreds of years as a sea food for mans and animals due to its excellent

nutritional profile and high carotenoid content. *Spirulina* is relatively high in protein ranging between 55-65% and includes all of the essential amino acids. Phytochemical studies on *Erythrina variegata* species have demonstrated alkaloids and flavonoids as major constituents³. Different parts of *E. Variegata* have been used in traditional medicine as nervine sedative, febrifuge, anti-asthmatic and antiepileptic. This is an excellent herbal therapy for convulsion, fever, inflammation, bacterial infection, insomnia, helminthiasis, cough, cuts and wounds. Keeping in view the inadvertent use of imidacloprid as insecticide in various agricultural and animal husbandry practices, the present experiment was planned to study the toxic effects of imidacloprid and the ameliorating potential of *Spirulina platensis* and *Erythrina variegata* on tissue changes in various organs in imidacloprid induced toxicity in male white leghorn cockerels.

Table 1. Experimental design for the study.

Groups	Treatment	No. of birds
I	Control	7
II	<i>Spirulina platensis</i> @ 0.2% in feed	7
III	Imidacloprid @ 50ppm in feed	7
IV	<i>Spirulina platensis</i> @ 0.2% +Imidacloprid @ 50ppm in feed	7
V	<i>Erythrina variegata</i> @ 1 % in feed	7
VI	<i>Erythrina variegata</i> @ 1 % +Imidacloprid @ 50ppm in feed	7

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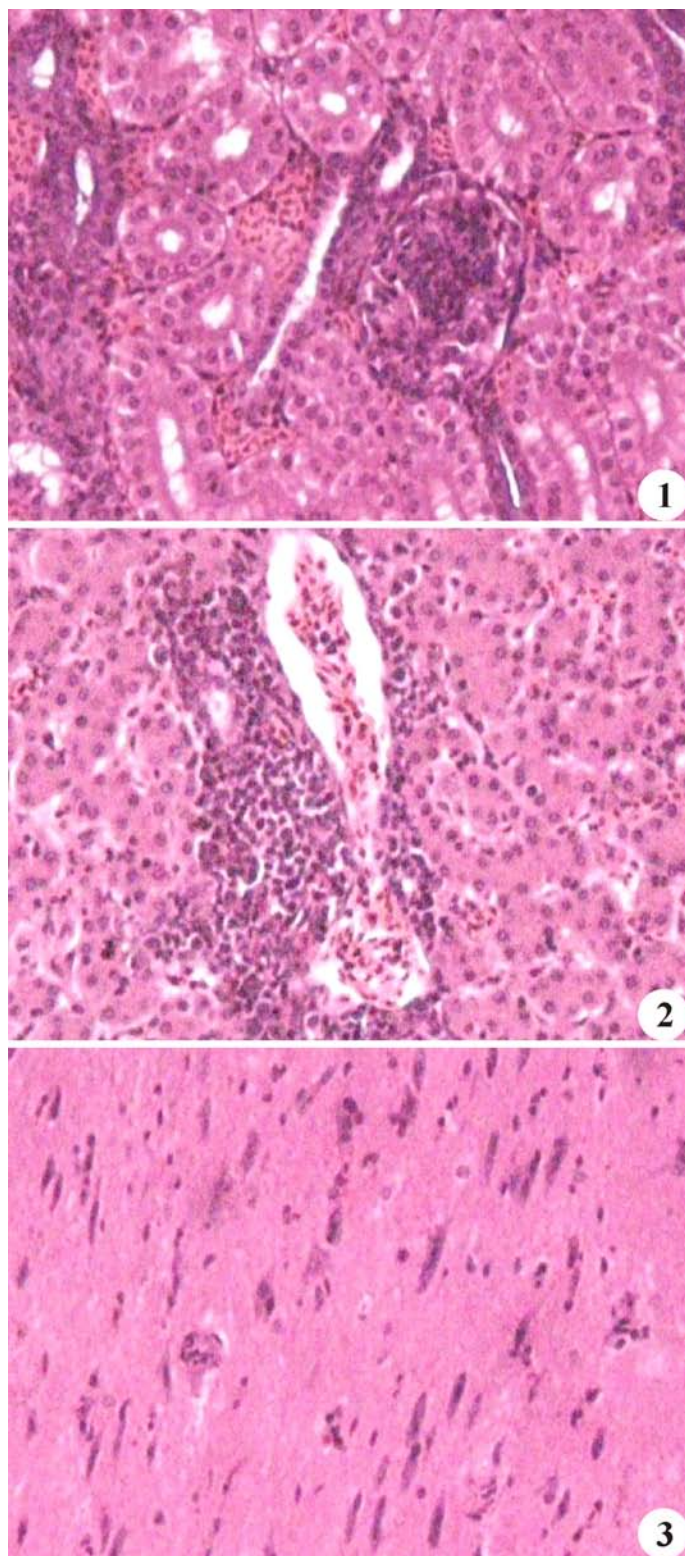


Fig.1. Kidney of group III bird showing severe congestion of large blood vessels, interstitial haemorrhages in between tubules and vacuolar degeneration in many of the kidney tubular epithelial cells. H&E x200. **Fig.2.** Liver of group III bird showing congestion of large blood vessel along with mononuclear cell infiltration around it. H&E x100. **Fig.3.** Brain of group III bird showing elongation of neurons and gliosis. H&E x200.

Forty two male white leghorn cockerels weighing between 250-275 gm of 4-6 months age were procured from Instructional Poultry Farm, Pantnagar. Birds were divided randomly and equally into six groups viz. I, II, III, IV, V and VI. The birds were maintained on the standard poultry feed and water *ad libitum*. The experiment was conducted as per the schedule mentioned in Table 1. The study was carried out for 60 days. After 60 days post treatment, all the birds were sacrificed and subjected to detailed postmortem examination. Gross lesions were duly recorded and the representative tissue samples of liver, kidney, brain and testes were collected in 10% buffered formalin and processed as per standard protocol for the histopathological examination⁴. *Erythrina variegata* leaves used in the study were collected from GBPUAT Pantnagar campus. *Spirulina platensis* (Spirulina DXN) manufactured by Twenty First Century Pharmaceuticals, Chennai was used in the study.

Grossly, kidneys in group III birds showed redness and swelling, while liver in the birds of this group (group III) was pale in colour and enlarged in size as compared to that of birds of other groups. Testes of cockerels in group III were severely reduced in size compared to other groups. Brain showed no gross lesion in cockerels of group III. No gross changes were observed in any of these organs in any other groups.

Histopathologically, no microscopic changes were observed in any of the organs of birds of control group I. Kidneys in the birds of group III showed severe congestion of blood vessel, interstitial haemorrhages and swelling of the glomeruli with hypercellularity of the glomerular capillaries (Fig. 1). There was mononuclear cell infiltration at various places. Degeneration and vacuolation of few kidney tubular cells in many of the tubules were also noticed. No microscopic changes were seen in the kidney of birds of groups I, II, IV and V, whereas these lesions were of very mild to moderate intensity in kidney of birds of group VI. Liver in birds of group III exhibited severe congestion of large blood vessels, sinusoidal congestion at places, vacuolar degeneration of the hepatocytes throughout the parenchyma, presence of mononuclear cells around congested blood vessels at many places (Fig. 2), and a few of the hepatocytes showing pyknosis. No microscopic changes were seen in birds of groups I, II, IV and V. These lesions were of very mild intensity in the liver of birds of group VI as compared to that of group III birds. Testes of group III birds showed reduction in size of the seminiferous tubules and many of the tubules were having only a single layer of germinal epithelium. No microscopic

changes were seen in testes of birds of groups I, II, IV and V. These lesions were of mild intensity in the birds of group VI. Brain of the birds in group III exhibited congestion of blood vessels, elongation of neurons, gliosis and satellitosis at few places (Fig. 3). No microscopic changes were observed in the brain of cockerels of groups I, II, IV and V. These lesions were of mild intensity in the birds of group VI.

The lesions recorded in present study in case of liver and kidney are in accordance to that reported by earlier workers in the liver and kidney of layer chickens given imidacloprid @ 131 mg/kg b wt. orally⁵ and in liver of rats given imidacloprid @ 0.21 mg/kg b wt orally for 4 weeks⁶. Lesions found in testes in our study have also been reported earlier in testes of rats given imidacloprid @ 80 mg/kg b wt⁷. The lesions recorded in brain in present study are similar to those reported earlier in brain of rats given 20mg/kg b wt imidacloprid for 90 days⁸. Protective effects of *Spirulina platensis* on liver and kidney as observed in present study are similar to those reported in liver and kidney by earlier workers when *Spirulina platensis* was administered @ 500 and 1000 mg/kg b wt against deltamethrin (30 mg/kg) toxicity in rats⁹. Nephroprotective effect of *Spirulina platensis* administered @ 1000 mg/kg b wt orally in gentamicin sulphate (100 mg/kg, i.p.) induced nephrotoxicity in rats have been reported earlier¹⁰. The lesions observed in liver, kidney, testes and brain in our study, which were produced by imidacloprid might be due to oxidative damage caused by increased production of highly reactive oxygen and nitrogen species. The ameliorative effect shown by *Erythrina variegata* and *Spirulina platensis* might be due to the presence of phycocyanins and flavonoids (antioxidants) respectively.

It is concluded from the present investigation that imidacloprid @ 50ppm in feed for 60 days produced toxic pathological effects. Feeding of *Spirulina platensis* (0.2%)

and *Erythrina variegata* leaf powder (1%) in diet, respectively for 60 days significantly ameliorated the imidacloprid induced toxic effect in the tissues of white leghorn cockerels.

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