CLINICO-PATHOLOGICAL STUDIES OF THIACLOPRID TOXICITY IN BROILER CHICKENS

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ABSTRACT

The maximum tolerated dose (MTD) and clinico-pathological studies on administration of thiacloprid were undertaken in 40 broiler chicks of 14 days of age. Different groups having five birds each were given thiacloprid @ 50, 45, 40 39, 38, 37 and 35 mg/kg body weight (BW) by oral route as single dose. MTD value of thiacloprid was found to be 39 mg/kg body weight based on clinical signs and mortality. The birds administered thiacloprid above MTD levels exhibited clinical signs of toxicity such as nervous signs as head tremors, paralysis of one or both the legs along with posture abnormalities like legs abducted from body and resting on belly on the ground and ataxia. Apart from these signs, the affected birds also exhibited loss of appetite, respiratory distress, excessive salivation, dropping of wings, diarrhoea and closing of eye. Gross pathological lesions in groups showing thiacloprid toxicity revealed severe congestion in various organs such as liver, kidney, lungs and spleen and mild hemorrhages in heart, liver and lungs. Histopathologically, there was severe congestion, hemorrhages, and focal area of necrosis accompanied with infiltration of heterophils in liver, mild pericarditis and myocarditis and congestion and hemorrhages in other organs such as spleen, kidney and thymus. The study revealed that the MTD of thiacloprid in chickens was 39 mg/kg body weight by oral route and the doses higher than MTD caused severe toxicity in chicken.

Key words: Thiacloprid, maximum tolerated dose, toxicity

Thiacloprid [3-(6-chloro-3-pyridylmethyl)-1,3thiazolidin-2-ylidenecyanamide] is a new neonicotinoid insecticide that belongs to a new group of active ingredients, the cyanoamidines. Birds exposure to this insecticide occurs through the use of insecticide contaminated poultry litter (Amure and Stuart, 1978) and feed mixtures having insecticide residues used in poultry ration (Naber, 1977). Experimental studies are usually required to study the toxicity and for determination of maximum tolerated dose (MTD) or median lethal dose (LD₅₀) of such chemicals before they can be safely used for control of insects at farms. For thiacloprid, oral LD₅₀ values were found to be 640 mg/kg in rats and 49 mg/ kg body weight in Japanese and bobwhite quails (Tomizawa and Casida, 2005). However, no report could be traced out in the literature regarding the MTD of thiacloprid in broilers chicken. MTD values of thiacloprid in broilers chicken will play crucial role in conducting further experimental toxicity studies as this parameter is considered better than LD₅₀. Thus, in present study, MTD of thiacloprid was calculated and clinico-pathological effects of its toxicity were studied in broiler chickens following oral administration of thiacloprid.

MATERIALS AND METHODS

Experimental Birds: Forty, day-old broiler chicks were procured from a local hatchery and were reared in the

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departmental animal house under strict hygienic conditions. The birds were provided feed and water *ad libitum*. The experiment was conducted after the approval of Institutional Animal Ethical Committee of this university.

Determination of MTD: Thiacloprid (Alanto 21.7% SC-Bayer Crop Science Limited) was used in the present study. MTD of thiacloprid was determined after screening different doses of thiacloprid, following standard methods as described by Moser and Padilla (1998). Out of 40 chicks used for MTD determination, 20 chicks of 14 days old were divided into seven groups (A, B, C, D, E, F and G) with five chicks in each. All the birds in these groups were given thiacloprid @ 50, 45, 40, 39, 38, 37 and 35 mg/kg body weight, respectively by oral route as single dose (Table 1). To verify MTD, additional group of five chicks was administered orally with MTD of thiacloprid and was tested to define more closely the time course of the effects.

Pathological Studies: The toxico-pathological study was conducted in different groups (A to G) after giving the different doses of thiacloprid. The clinical signs observed in the birds and mortality in each group was recorded.. A detailed necropsy examination of all dead or sacrificed birds from each group was conducted for study of gross pathological lesions. Representative pieces of liver, heart, lung, kidney, spleen and thymus were collected in 10% buffered formalin for histopathological studies (Luna, 1968).

Table 1
Oral doses of thiacloprid used for determination of maximum tolerated dose (MTD) in broiler chicken

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Groups	Number of chicks/group	Different doses of thiacloprid (mg/ kg body weight.; orally)	
	C 1		
A	5	50	
В	5	45	
C	5	40	
D	5	39	
Е	5	38	
F	5	37	
G	5	35	

RESULTS AND DISCUSSION

Determination of MTD: MTD of thiacloprid was determined after screening different doses of thiacloprid. All the birds in different groups (groups A, B, C and G) were given thiacloprid @ 50, 45, 40 and 35 mg/kg body weight, respectively by oral route as single dose) were observed for mortality or clinical signs until death or up to 24 h whichever was earlier. Mortality was noticed only in the groups given thiacloprid @ 50, 45, and 40 mg/kg body weight (Table 2). No mortality was observed in the chicks given thiacloprid @ 35 mg/kg body weight. Again to verify MTD, three more doses of thiacloprid @ 39, 38 and 37 mg/kg body weight were given orally as single dose to three different groups (D, E and F each containing five chicks). No mortality was observed in these groups also. MTD value was found to be 39 mg/ kg body weight based on clinical signs and mortality. To verify MTD, additional group of five chicks was given orally with 39 mg/kg body weight of thiacloprid and was tested to define more closely the time course of the effects. In these birds also, no mortality was observed but they exhibited clinical signs. Therefore, MTD of thiacloprid in broilers chicken was found to be 39mg/kg body weight by oral route. According to the literature,

Table 2

Mortality pattern due to oral administration of thiacloprid in different groups of broiler chickens

Group	Dose of (mg/kg thiacloprid body weight; orally)	Number of chicks died/number of chicks administered	Percent mortality
A	50	2/5	40
В	45	2/5	40
C	40	1/5	20
D	39	0/5	0
E	38	0/5	0
F	37	0/5	0
G	35	0/5	0

oral LD50 values in Japanese and bobwhite quails has been reported to be 49 mg/kg body weight but no report could be traced in the literature on MTD determination in broilers chicken.

Clinical Signs of Thiacloprid Toxicity: The birds of groups A, B and C exhibited nervous clinical signs such as tremors of head, which started episodically after 15-20 min of the administration (lasting for 10 to 20 sec), paralysis of one or both the legs along with posture abnormality showing legs abducted from body and belly of bird touching the ground. Other clinical signs observed were loss of appetite, respiratory distress, excessive salivation, dropping of wings, diarrhoea, ataxia and closing of eye (Fig. 1). These clinical signs were more severe in groups A and B i.e. with higher thiacloprid doses as compared to groups C and D. However, such clinical signs were not noticed in groups E, F and G. The severity of these clinical signs decreased progressively and completely disappeared within 24 h of administration of thiacloprid in the birds which remained alive. The birds with higher doses than MTD had slow respiration rate, however, the rate of respiration was high just before death. The nervous signs and high respiratory rate could be correlated with the agonist action of thiacloprid at nicotinic acetylcholine receptors (nAChR) which has been reported to induce neuromuscular paralysis (Tomizawa and Casida, 2005; EPA, 2005). The early appearance (within 10-15 min of administration) and completely disappearance (within 24 h of administration) of clinical signs might be associated with the prompt absorption and excretion of thiacloprid.

Pathological Lesions of Thiacloprid Toxicity: Necropsy examination of the birds died or sacrificed in different thiacloprid treated groups revealed severe congestion in various organs such as liver, intestine (Fig. 2), kidney, lungs and spleen. The pathological changes were less severe in birds of groups D, E and F as compared to the groups A, B and C.

Histopathological examination revealed congestion, hemorrhages, fatty changes and focal area of necrosis accompanied with infiltration of mononuclear cells in the liver of group A and group B birds (Figs. 3 and 4). Heart revealed the congestion, mild hemorrhages and infiltration of few heterophils in myocardium along with pericarditis characterized by hemorrhages and infiltration of heterophils and infiltration of few mononuclear cells in myocardium.



Fig. 1: (Group A): Chicks showing dull, depressed paralyzed leg and belly of bird touched the ground

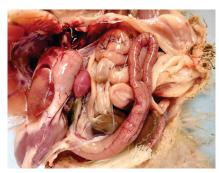


Fig. 2: (Group B): Chick showing congestion in intestine

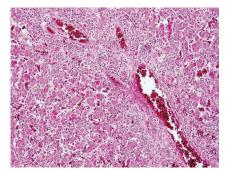


Fig. 3: (Group A): Liver revealed congestion and mononuclear cell infiltration (H&E X 200)

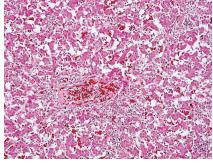


Fig. 4: (Group B): Liver revealed congestion and leucocytic infiltration in parenchyma (H&E X 200)

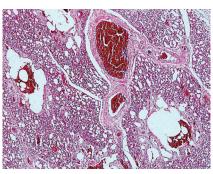


Fig. 5 : (Group A): Lung revealed severe congestion and haemorrhages (H&E 200X)

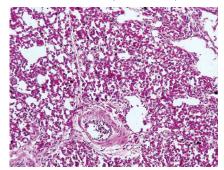


Fig. 6: (Group B): Lung revealed congestion of alveolar capillaries (H&E 200X)

In lungs, there was severe congestion along with serous pneumonia (Figs. 5 and 6) and mononuclear cell infiltration in bronchiolar mucosa. Other organs such as kidney, spleen, and thymus revealed mild to moderate congestion. The liver has been reported to be the principal target organ of thiacloprid toxicity (Goyal *et al.*, 2010; Hendawi *et al.*, 2016). The results of pathological studies are in accordance with findings of other workers in chickens (Goyal *et al.*, 2010) and rats (Roy, 2013; Hendawi *et al.*, 2016) who have also reported degeneration, necrosis and circulatory lesions due to administration of thiacloprid.

Thus it may be concluded from the present study that the MTD of thiacloprid in broiler chicken is 39mg/kg body weight and the toxicopathological lesions of its toxicity in chicken includes vascular changes in visceral organs with necrotic and inflammatory changes in liver, heart and lungs.

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