

Effect of Monocrotophos on Development of the Chick Embryo

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ABSTRACT

The avian embryo is a long- standing model for developmental biology and toxicology research both in ovo and in explants culture. Fertile eggs are inexpensive and the embryo develops rapidly, hence most extensively used. The absence of a placenta permits the direct study of teratogenic effects. The growing reliance on chemicals in agriculture and the indiscriminate use of pesticides leading to environmental pollution is posing a potential health hazard. In order to have a compromised formula between the use of pesticides and their harmful effects on biosphere, research studies are on the increase to find out the deleterious effects of pesticides on biological systems. Monocrotophos has withdrawn from use in developed countries but it is still used in many parts of the country. Present study is planned to understand the effect of monocrotophos on development of one breed of chick embryo namely, Gramapriya developed by the Project Directorate on Poultry (ICAR), Hyderabad by determining the heart beat rate, somite count and limb bud and tail bud formation in the developing embryo at 48 hours, 72 hours and 96 hours of incubation. The mortality rates of treated groups were predominantly higher than control group.

Keywords: Developmental Biology, Toxicology, Teratogeny, Monocrotophos, Gramapriya,

INTRODUCTION.

Monocrotophos is a red label pesticide in India which is highly hazardous, systemic and a less persistent organophosphate insecticide. It is an acaricide belonging to Vinyl Phosphate group. It is widely used to control pests of a variety of crops such as rice, cotton, castor, maize, peanuts, potatoes and onion. It is widely used to control a wide spectrum of chewing, sucking and boring insects. As it is sprayed on the crops, the birds tend to peck and feed on the infested grains. This will affect the bird as well as embryo. Prolonged exposure is known to cause chronic neurological syndrome, immune suppressive action, teratogenic effects in experimental animals (Meeker et al, 2006). Humans end up consuming these infested birds as well as eggs which can be harmful. Monocrotophos is one of the most toxic pesticides to birds, acute oral LD50 range from 0.9 -6.7 mg/kg body weight (Rajendran, 2009).

Monocrotophos has been withdrawn from use in developed countries due to its high toxicity against beneficial and non – target organisms. But its usage for the control of major insect pests

in agriculture is being continued in developing countries like India primarily due to lack of alternative replacement. Its low cost and many possible applications have kept up demand in India despite growing evidence of its negative impact on human health. Monocrotophos can be absorbed by ingestion, inhalation or skin contact.

Chick embryo has contributed enormously to experimental embryology and there is a vast amount of literature describing the development and their use as model system (Freeman and Vince, 1974). The chick embryo is commonly used in pharmacological and toxicological research owing to its ready availability, ease of handling and because many of its responses have predictive value for other species. (Rohlich, 1977).

Avian species largely lag behind in classical toxicological studies; however, in recent years some encouraging results have been obtained employing embryonic and neonatal chick. (Pallavi, 2015) Present study was conducted on eggs of Gramapriya Chicken, which were procured from I.C.A.R., Ela farm, Old Goa.

METHODOLOGY

The study was designed to investigate the teratogenic effects of Monocrotophos on chick embryo by determining the heart beat rate, somite count and limb bud and tail bud formation in the developing embryo at 48 hours, 72 hours and 96 hours of incubation to evaluate morphometric variations. Fertilized eggs from Gramapriya breed were divided into three groups depending upon the hours of incubation.i.e.48 hours,72 hours and 96 hours.Eggs from each group were marked as control, dose 1,dose 2 and dose 3 eggs and were injected with 0.2 ml chick ringer solution, 0.2, 0.4 and 0.6 ml of Monocrotophos chronic dose respectively using micro syringe. All the eggs were kept in the incubator at 37⁰ C. The eggs from each group were broken after 48 hours, 72 hours and 96 hours of incubation. The embryos were washed with chick ringer solution and were observed under microscope.

RESULTS

Chick embryos were examined for toxic and teratogenic effects of Monocrotophos. The malformations were observed sporadically in treated groups of chick embryos and no specific pattern of malformations was seen in normal group. The results show that experimental group had physiological and morphological changes, increase in heartbeat rate and decrease in somite count. Also mortality of chick embryos occurred at 0.4 ml and 0.6 ml dose of Monocrotophos treated groups. (Table.1)

The chick embryos treated with higher Monocrotophos were found to show the teratogenic effects. At 48 hours of incubation somite count decreased and heartbeat rate increased in chick embryos. Limb bud and tail bud formation was not seen in this breed and all the embryos with 0.40 ml dose were found dead and 0.60 ml dose were degenerated.

At 72 hours of incubation with increasing concentration of Monocrotophos gradual decrease in somite count and increase in heartbeat rate seen. At 96 hours of incubation a decrease in somite count and increase in heartbeat rate with increase in Monocrotophos dose was seen. Formation of limb buds and tail bud was seen more prominently in embryos of Gramapriya. All the embryos treated with 0.6 ml dose were degenerated in Gramapriya breeds and deformed tail bud and limb bud were seen in 0.4 ml dose. The mortality rates of treated groups were predominantly higher than control group.(Table.1)

DISCUSSION

The phase of embryogenesis and/or the beginning of organogenesis is very delicate period of prenatal development as rapidly differentiating and growing system of any developing embryo may be disturbed by introduction of toxic substances. During the development of chick embryo, the day of incubation is first critical period since the blastoderm in the incubated egg is on the threshold of gastrulation and may thus are more susceptible to any toxicant (Sahu and Ghatak,2002). The day 4 of incubation is the second critical period in which organogenesis and differentiation of various organ rudiments are established. (De Witt et al 2005). The present investigation shows that the growth and development of chick embryos were seriously affected at three sub lethal doses of Monocrotophos. Growth retardation is the manifestation of abnormal development. It is observed that this parameter is the most sensitive indicator of embryo toxicity (Maclaren and michie,1963; Kimbrough and Gains, 1968). Upshall et al (1968) suggested that in avian species, growth abnormalities may be due to nutritional deficiencies of the embryo to obtain ester hydrolysis products from the yolk. Budreau and Singh (1973) further contended that availability of nutrients to the foetus may be affected due to inhibition of esterase by the organophosphate compounds. Chemicals introduced into the eggs may cause a special type of toxicity because it destroys, alters or combines with essential nutrients such as vitamins, and minerals.(FDA Science Project series 1968).It is observed in the present investigation that exposure of fertilized eggs to three different sub lethal doses of Monocrotophos caused significant increase in heart beat and decrease in somite count in embryos of all three breeds of chicken in addition to this adversely affecting by the growth retardation and deformities in the embryos development. From this it is evident that with an increase in time of exposure the toxicity of Monocrotophos increased in chick embryo. The viability of the chick embryo was affected significantly at the high dose. Since embryos were affected by high doses of the Monocrotophosresulting the detectable histopathological and physiological aspects in chick embryos which pose a serious threat to profitable poultry production, use of this toxic product should be restricted or banned in the society with the help of enforcement authority.

CONCLUSION

From Our study it is concluded that “Terata” – formation at least in the chick embryos is definitely due to the toxic effect of Monocrotophos which can even cause abnormalities in morphological and histological aspects. Thus chicken consuming the pesticide treated grains

consume at least in lower doses, which in turn is consumed by the human beings through chick embryos or flesh causing human health hazards indicating its toxic potential.

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OBSERVATION TABLE

Table: 1 Effect of Monocrotophos on chick embryos of Gramapriya breed.

Categories		Somite count	Heartbeat	Tail bud Formation	Limb bud Formation
Hours	Doses				
48 hours	Control	26	105	Not seen	Not seen
	Dose 1 (0.2 ml)	25	109	Not seen	Not seen
	Dose 2 (0.4 ml)	24	-----	Not seen	Not seen
	Dose 3 (0.6 ml)	-----	Embryo degenerated	Embryo degenerated	Embryo degenerated
72 hours	Control	36	111	Seen	Seen
	Dose 1 (0.2 ml)	34	113	Seen	Seen
	Dose 2 (0.4 ml)	32	-----	Seen	Seen
	Dose 3 (0.6 ml)	-----	Embryo degenerated	Embryo degenerated	Embryo degenerated
96 hours	Control	41	116	Seen	Seen
	Dose 1 (0.2 ml)	34	118	Seen	Seen
	Dose 2 (0.4 ml)	32	-----	Seen	Posterior limb bud deformed
	Dose 3 (0.6 ml)	Embryo degenerated	Embryo degenerated	Embryo degenerated	Embryo degenerated

Figure.1. Effect of Monocrotophos on somite count of chick embryo of Grampriya breed.

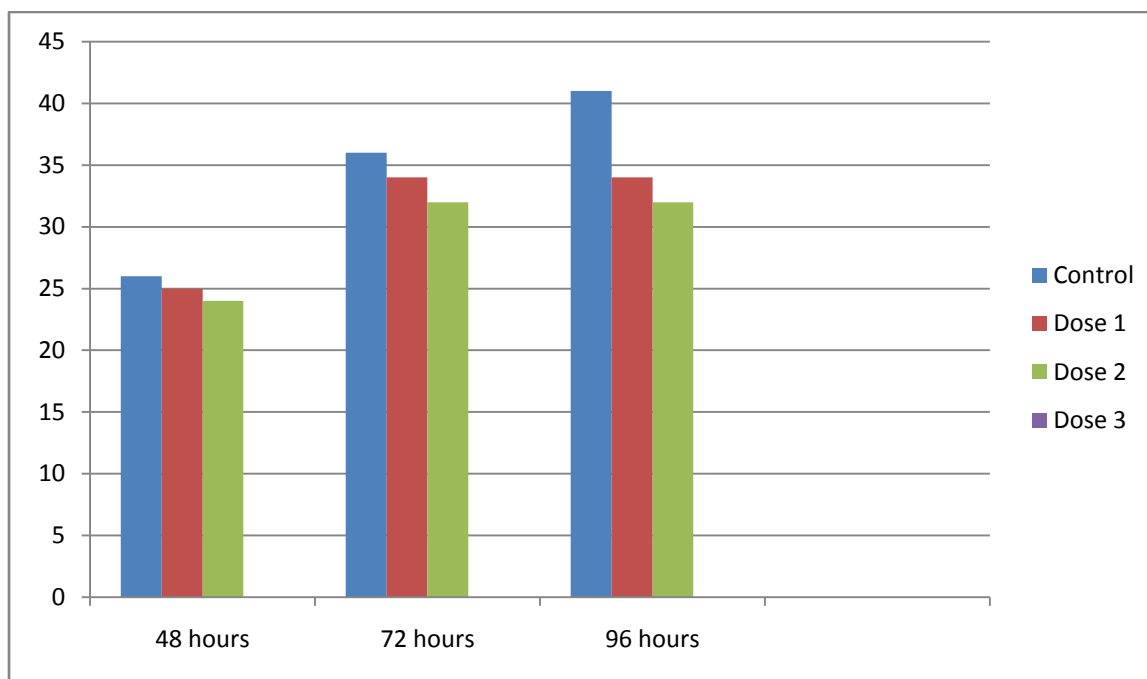
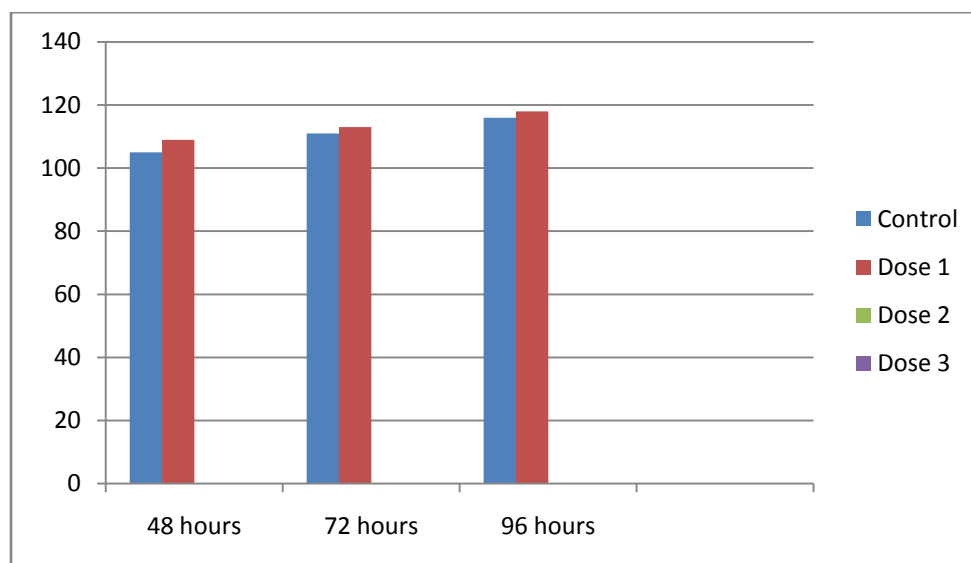
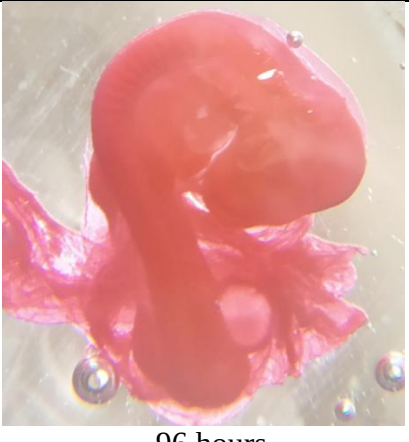
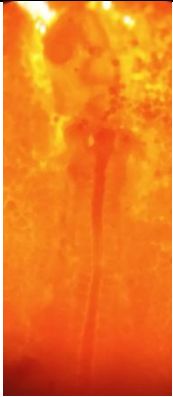
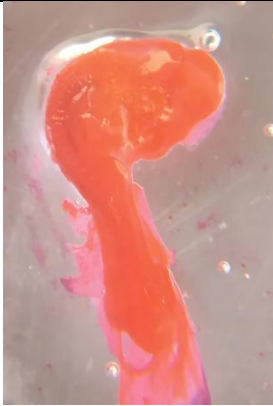
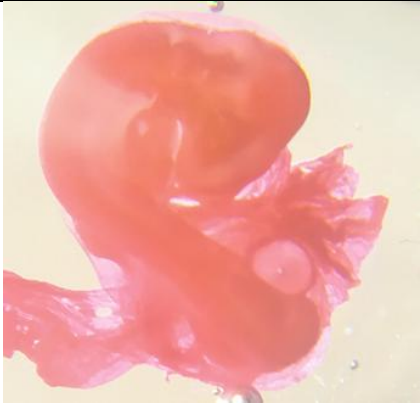
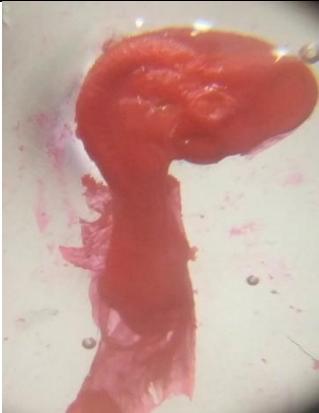


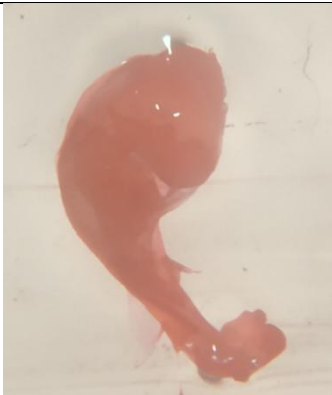
Figure.2. Effect of Monocrotophos on heartbeat of chick embryo of Grampriyabreed.



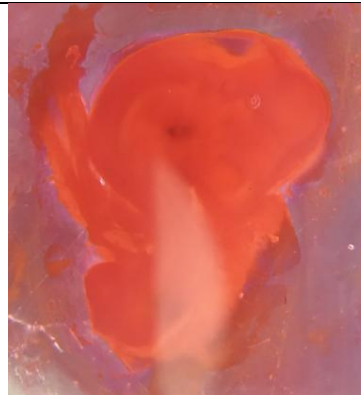
 48 hours Control Normal growth	 72 hours Control Normal growth	 96 hours Control Normal growth
 48 hours	 72 hours	 96 hours
 48 hours	 72 hours	 96 hours



48 hours
Embryo degenerated



72 hours
Embryo degenerated



96 hours
Embryo degenerated