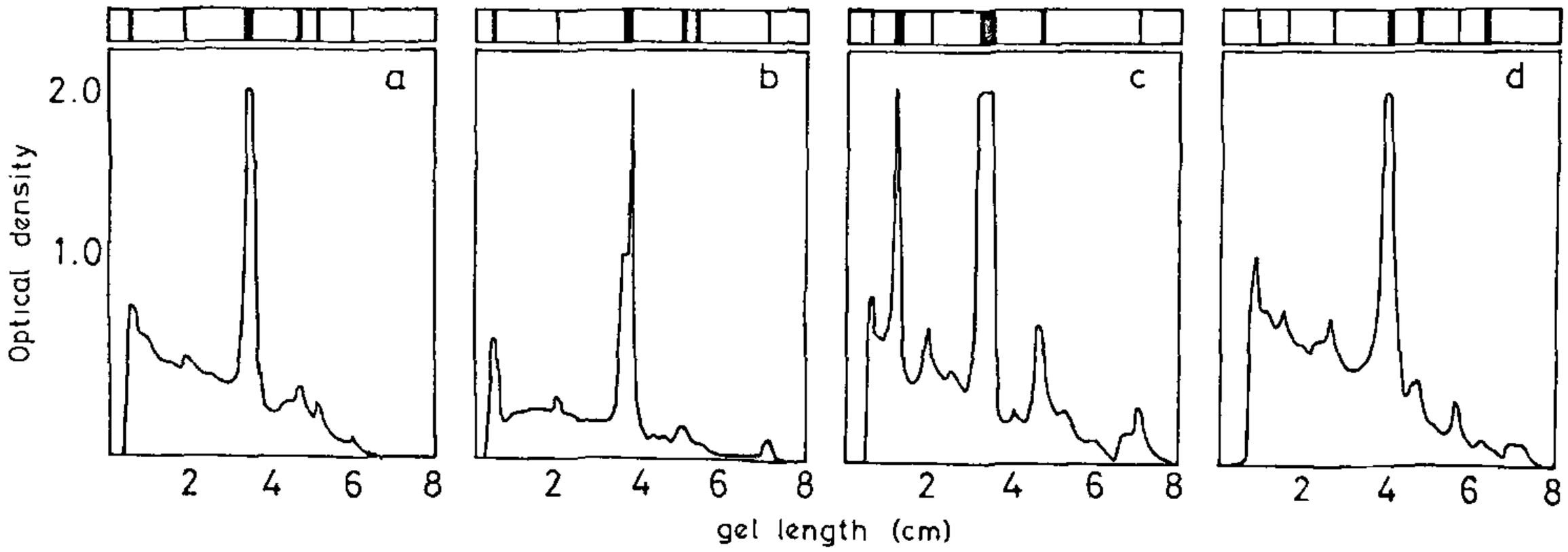

Figure 1. Zymograms showing the myogen patterns of **a.** *Ichthyophis glutinosus*, **b.** *I. beddomi*, **c.** *I. bombayensis* and **d.** *Uraeotyphlus narayani.*

cated against each of the four species (*Ichthyophis glutinosus, I. beddomi, I. bombayensis* and *Uraeotyphlus narayani*) conform to the key given by Taylor[3].

Figure 1 shows the zymograms of the four species. Their soluble muscle proteins showed considerable variations in their mobility patterns, indicating the distinctness of each species. While the myogen patterns reported here for *I. glutinosus* are not comparable with the serum protein patterns reported for a Ceylonese population of the same species by Case and Wake[2], it is interesting that the two banded species, *I. glutinosus* and *I. beddomi*, have comparable myogen paterns. *I. bombayensis* showed distinct differences in its myogen patterns.

The available data on the myogen patterns in caecilians are limited and it is difficult to establish conclusively the taxonomic relationships of individuals belonging to the group. Clearly, such studies on biochemical taxonomy of caecilians are warranted.

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EFFECTS OF TECHNICAL GRADE MALATHION AND DDT ON THE ACTION OF CHLORPROMAZINE, DIAZEPAM AND PENTOBARBITONE WITH REFERENCE TO FIGHTING BEHAVIOUR IN MICE

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INSECTICIDES are increasingly being used to control vectors and parasites affecting man and animals. Measurable quantities of insecticides have been demonstrated in mammalian tissues all over the world[1]. In view of this it is important to know how the living organisms react to these chemicals. The exposed individuals may also consume drugs which may be resulting in an interaction of drugs with insecticide residues already present[2].

The manifestation of fighting behaviour in humans is as simple as in animals. Behaviour characteristic of anger can be induced in the laboratory animals by a variety of experimental methods[3]. The present study was undertaken to determine the influence of DDT and malathion, on shock-induced fighting behaviour, and the interaction of these chemicals with some centrally acting drugs namely chlorpromazine, diazepam and pentobarbitone.

Male mice (750 in number) of Swiss strain were used. Animals were maintained on standard feed (Hind Levers animal feed, India) and water *ad libitum*. They were acclimatized in the laboratory prior to experimentation. To induce fighting in mice, foot shock method[3] using a square wave stimulator (pulse duration 1 msec, frequency 10/sec, 30 V) was employed. The frequency of fighting episodes of each

pair of mice was noted and the pairs showing at least 3 episodes per minute were used in this study. The mice were then tested at an interval of 15 days to study the effect of insecticides and/or drugs.

Technical grade malathion (97.72%) and DDT were used. Three dose levels of malathion and DDT, equivalent to 20, 10 and 4% of their LD_{50} in mice determined earlier[4] were tested. Malathion and DDT were suspended in arachis oil and injected i.p. 2 and 3 hr, respectively before mice were subjected to foot-shock. Animals in the control group received arachis oil only. In another set of experiments, chlorpromazine (CPZ), diazepam (DZP) or pentobarbitone (PB) was given to different groups of mice 30 min before the animal were subjected to foot-shock. A drug was considered to have a blocking effect when the pairs of mice exhibited less than 3 episodes during one minute of foot shock. Each drug was tested at three or more dose levels using six pairs of mice per dose. Using quantal response assay, ED_{50} of each drug for suppressing the shock-induced fighting behaviour was calculated in control and malathion/DDT-treated groups.

Paired male mice when subjected to foot shock exhibited fighting episodes lasting 1–3 sec. Under the influence of DDT or malathion, the number of fighting episodes increased significantly (table 1). The number of episodes increased by 119 and 102% in mice treated with malathion at a dose rate of 200 and 100 mg/kg, respectively. The corresponding 20 and 10% LD_{50} of DDT showed a lower though significant effect. Further, the increase in episodes was greater in animals receiving malathion (40 mg/kg) or DDT (13 mg/kg) daily for 15 days. Also, these mice bit severely each other, resulting invariably in bleeding at mouth.

TABLE 1

Effect of malathion (MAL) and DDT on electroshock fighting behaviour in mice

Treatment mg/kg i.p.	Average number of episodes	Increase in episodes (%)
Control	7.0	—
MAL 200	15.3*	119
MAL 100	14.1*	102
DDT 65	13.0*	85
DDT 32.5	11.7*	67
Control	6.7	—
MAL† 40	17.2*	157
DDT† 13	14.2*	112

† MAL/DDT was administered daily for 15 days.

* Statistically significant when compared with corresponding control value ($P < 0.01$).

The effect of malathion and DDT pretreatment on the blockade of shock-induced aggressive behaviour by CPZ, DZP and PB in mice is presented table 2. It is seen that pretreatment with malathion decreased the effectiveness of CPZ, DZP and PB in suppressing fighting episodes in mice though only the ED_{50} of CPZ was increased significantly. The effect of DDT (32.5 mg/kg) pretreatment was, however, negligible. As is evident from table 3,15-day pretreatment with malathion and DDT could modify the effect of CNS active drugs. Even in this case, pretreatment with malathion produced more effect than DDT since it raised the ED_{50} of CPZ and DZP to a greater extent. The effect of daily administration of DDT (13 mg/kg) for 15 days in modifying the effect of drugs was, however, greater than the effect of its single dose (32.5 mg/kg).

TABLE 2

Effect of malathion (MAL) and DDT pretreatment on the blockade of electroshock fighting behaviour by CPZ, DZP and PB in mice

	ED_{50} mg/kg		
Treatment mg/kg, i.p.	CPZ	DZP	PB
Control	4.0	6.2	19.0
MAL 100	8.5*	7.8	22.3
DDT 32.5	4.7	6.2	20.8

* Statistically significant when compared with corresponding control value ($P < 0.05$).

TABLE 3

Effect of malathion (MAL) and DDT pretreatment on the blockade of eletroshock fighting behaviour by CPZ, DZP and PB in mice

	ED_{50} mg/kg		
Treatment mg/kg, i.p.	CPZ	DZP	PB
Control	4.0	5.8	19.9
MAL† 40	7.3	9.4*	22.5
DDT† 13	5.9	7.2	22.0

† MAL/DDT was administered daily for 15 days.

* Statistically significant when compared with corresponding control value ($P < 0.05$).

The suppressant effect exerted by CPZ, DZP and PB on shock-induced fighting aggression in mice agrees with earlier reports[3,5,6] but there is no report to substantiate that DDT or malathion augmented fighting behaviour. The mechanism involved, therefore, can only be speculated. Irritation aggression which resembles rage reaction is elicited by a wide range of external stimuli which in the present case is pain caused by electric shock. Hypothalamus and limbic forebrain structures such as amygdala through certain neurochemical changes have been implicated in the aggressive behaviour. Also, there is a greater activation or arousal of the autonomic and endocrine components[7].

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INSULIN TOLERANCE TEST IN THE NORMAL AND HYPOPHYSECTOMIZED TOAD, *BUFO MELANOSTICTUS* (SCHNEIDER)

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THE hypoglycemic effect of insulin is very slow in poikilotherms as was reported in amphibians, *Bufo arenarum*[1], *Leptodactylus ocellatus*[2] and *Rana tigrina*[3] and in snakes[4,5] and crocodiles[6]. Hypophysectomized toads had a greater hypoglycemic action to insulin than normals[7]. This reaction could be reduced by the injection of extracts of the hypophysis, particularly the pars distalis[8]. The following experiment was performed to study the comparative tolerance to insulin of normal and hypophysectomized toad, *Bufo melanostictus*.

The animals used for this study were kept in cages containing water for two days and during this period they were not fed. Hypophysectomy was performed under ether anaesthesia. The mouth of the toad was kept open as wide as possible with the help of rubber bands. A medial cut (2.5 cm) was then made in the mucous membrance of the palate. The margins of the cut were pulled apart to expose the parasphenoid bone. A small window was made in the parasphenoid bone just above the position of the pituitary with a sharp scalpel. The cartilage which thus became visible was removed using a pointed needle. The pituitary which now lay exposed was removed with pointed curved forceps. The square piece of the parasphenoid bone was then put back in place and the cut ends of the mucous membrane were sewn together with surgical thread. Neosporine antiseptic was applied over the wound to prevent infection.

Insulin tolerance test was performed on normal and hypophysectomized animals by injecting a dose of 10 IU/kg of insulin (insulin lente, insulin zinc suspension, Lot No. 618, The Boots Company (India) Ltd., Bombay). For these tests 27 normal and 27 hypophysectomized animals were used. The normal animals were sacrificed after 0, 0.5, 1, 2, 3, 4, 24, 48 and 96 hr. In the hypophysectomized animals insulin was injected 24 hr after the operation and the animals were sacrificed after 0, 0.5, 1, 2, 3, 4, 24, 48 and 96 hr. All injections were given in the dorsal lymph sac. The animals were sacrificed by pithing. Blood samples were withdrawn from the conus arteriosus and were received in oxalated tubes. Blood sugar was estimated by Folin-Wu method[9], and the values obtained were plotted (figure 1).

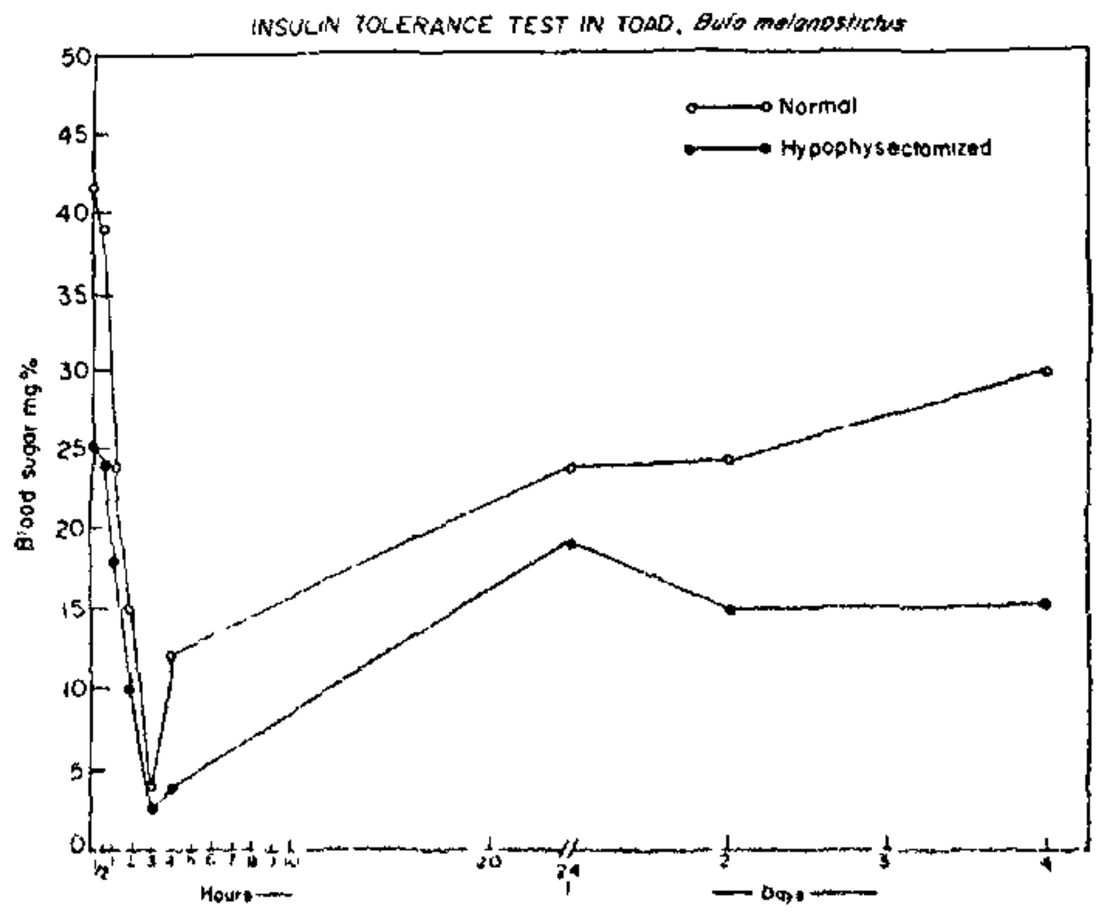

Figure 1. Hypoglycemic action of insulin (10 IU/kg) in normal and hypophysectomized toads (each point on the graph represents the average of three blood sugar values).