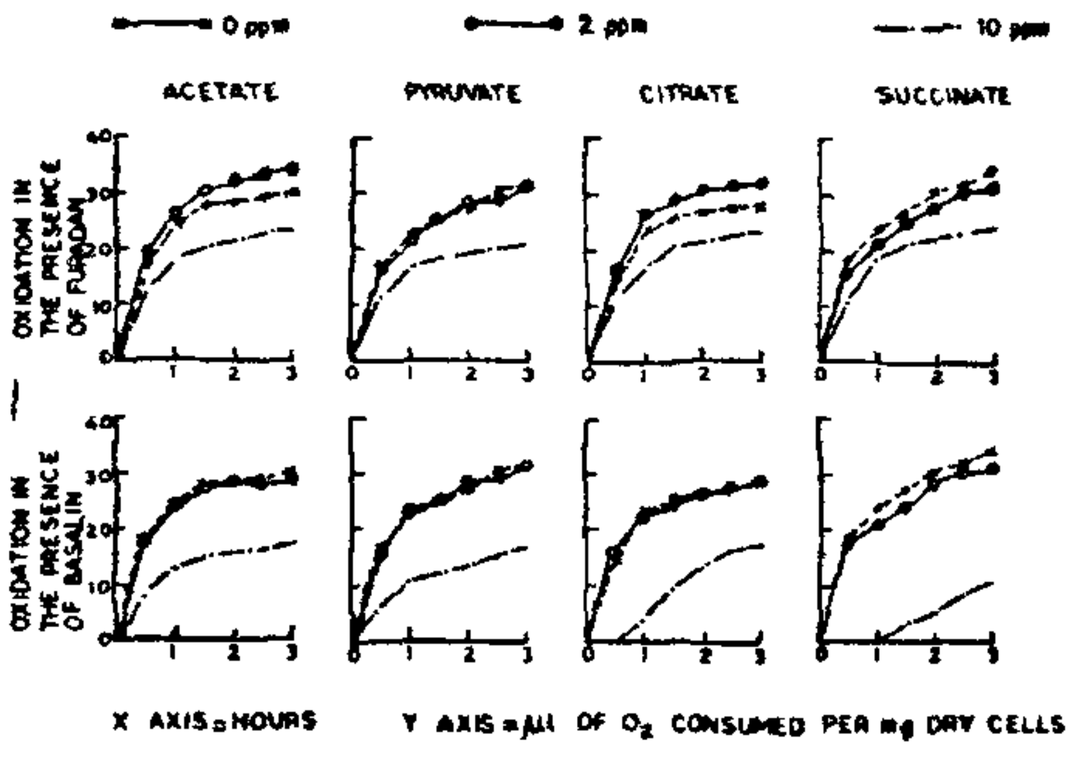

Figure 1. Effect of Furadan and Basalin on oxidative metabolism of cowpea bacteroids.

Soil application of furadan, even upto a level of 10 ppm had no significant effect on the oxidation of the substrates tried, except in the case of acetate where significant stimulation was observed (figure 1). The oxidation of all substrates except that of fumarate was significantly reduced in the presence of furadan *in vitro* at 10 ppm only, lower levels not exhibiting any inhibitory effect. Stimulation was noticed at 2 ppm level in acetate and citrate alone. Since Furadan persist less in soil[6], the chemical level at flowering stage might be too low to influence the bacteroids.

In Basalin, oxidation of all the five substrates by the bacteroids of 10 ppm soil treatment was significantly low, while that of 2 ppm showed no effect. Very similar results were also obtained *in vitro* studies. It is obvious that the nodule development and bacteroid metabolism could have been affected since the herbicide was applied right at the time of sowing and has longer persistence (more than two months as per the manufacturers report). It was observed that among the substrates tried the succinate oxidation at 0 ppm was maximum (more than 32 μ l of O_2/ mg dry cells) and it was more sensitive to Basalin than other substrates. This might be due to the succinate-stimulated acetylene reduction, by isolated bacteroids[7], indicating the key role played by succinate dehydrogenase in bacteroid metabolism. However, extensive research in this field is needed to explore these details.

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EFFECT OF SUBLETHAL CONCENTRATION OF METASYSTOX ON THE CIRCADIAN RHYTHM OF BIMODAL OXYGEN UPTAKE IN *CHANNA STRIATUS*

G. M. NATARAJAN. G. SUNDARA RAJULU, S. SIVAGAMA SUNDARI AND S. SUBRAMANIAN.
Department of Zoology, Bharathiar University, Comibatore 641 041, India.

ORGANOPHOSPHORUS insecticides are widely used in crop protection. Indiscriminate application of these insecticides may affect non-target organisms including economically important freshwater fish. Recent studies have shown that metasystox, the most widely used insecticide against cotton pests inhibited various physiological activities in fresh water fish[1]. However, no information is available on the sublethal effect of metasystox on the respiratory rhythms of fish. Since fish exhibit clear-cut respiratory rhythms[2], an attempt has been made in the present work to study the sublethal concentration of metasystox on the circadian rhythm of bimodal oxygen uptake in an economically important air breathing fish, *Channa striatus*.

One hundred snakehead of both sexes weighing 10–15 g were collected from local freshwater sources and maintained in the laboratory in constantly running aerated tap water under a 12 hr light: 12 hr dark cycle (LD 12:12) at 29 ± 1°C for 15 days before starting the experiments. The fish were fed on chopped goat liver on alternate days. Feeding was discontinued a day before the fish were used in the experiments. The circadian rhythm of bimodal oxygen uptake was studied at 29 ± 1°C using respiratory chambers designed by Reddy and Natarajan[3]. The amounts of oxygen consumed under water and in air were separately determined for a day at regular intervals of 3 hr each. The oxygen content of the water was kept constant throughout the study (6.5 ± 0.2 mg/l). Oxygen obtained by the fish from water was measured using Winkler's method. Oxygen consumed from the gas

TABLE 1

Effect of sublethal concentration of metasystox on the circadian rhythm of bimodal oxygen uptake of C. striatus (cc/hr) N = 12..

Time of day (Hrs)	Oxygen uptake								
	Aquatic			Aerial			Total		
	Control	Metasystox	% Change	Control	Metasystox	% Change	Control	Metasystox	% Change
6	0.063	0.052	−17.5	0.212	0.330	+55.7	0.275	0.382	+35.3
9	0.071	0.048	−32.4	0.208	0.342	+64.4	0.279	0.390	+39.8
12	0.074	0.031	−58.1	0.192	0.416	+111.4	0.266	0.447	+68.0
15	0.080	0.033	−50.9	0.220	0.421	+91.4	0.300	0.454	+51.4
18	0.083	0.027	−67.5	0.268	0.440	+64.2	0.351	0.467	+33.0
21	0.080	0.049	−38.9	0.246	0.443	+80.0	0.326	0.492	+50.9
24	0.076	0.048	−36.8	0.250	0.476	+90.0	0.326	0.524	+60.7
3	0.069	0.036	−47.8	0.236	0.408	+72.9	0.305	0.444	+45.6

Values expressed are mean for 6 individual observations significant at $P < 0.01$. The signs + or — indicate percent increase or decrease over control.

phase of the respiratory chamber was calculated as described by Umbreit *et al*[4]. In all these measurements, experiments were started only after the fish had adjusted themselves to the respiratory chamber for atleast 12 hr.

The technical grade of metasystox (Demeton: O, O-dimethyl s-2 (Sulphinyl) ethyl phosphorothioate) of 95% purity was obtained from Bayer Ltd, Bombay. Lethal (LC 50/48 hr) concentration was calculated by the probit method[5] and approximately 1/3 of the LC 50/48 hr concentration (1.7 mg/l) was selected for sublethal treatment. No mortality was recorded during the experiments and thus the results obtained describe sublethal reaction.

The data on circadian rhythm of bimodal oxygen uptake of normal and metasystox exposed (ME) fish have been given in table 1. A definite circadian rhythm is noticed in the oxygen uptake of control fish. Maximum oxygen uptake (0.268 cc/g/hr) through the air-breathing organs is recorded during 1800 hr and the minium (0.192 cc/g/hr) during noon (1200 hr). There is little variation in the aquatic oxygen uptake in different hours of the day. Maximum oxygen uptake through water is recorded during dusk (1800 hr) when aerial respiration is also at its peak. Accordingly, the oxygen uptake through the bimodal gas exchange machinery is maximum (0.351 cc/g/hr) during dusk and a minimum is recorded at noon (0.266 cc/g/hr). Morning and night hours of the day show no significant variation. This unimodel rhythm of bimodal oxygen uptake corroborates the findings of earlier authors[2] who have reported a maximum oxygen uptake during early parts of night in *C. striatus*.

Following ME the respiratory patterns were significantly reversed. When compared to the control, the aquatic oxygen uptake decreased at almost all the hours of the day. The decrease does not indicate any specific trend. However, from morning (0600 hr), there appeared a continuous rising trend in the percent decrease, but, at 1500 hr the trend reversed. This indicates that pesticide-treated fish, even at sublethal concentration are not able to maintain a normal physiological balance. It is quite interesting to note that minimum aquatic oxygen uptake is recorded at 18 hr where the control fish exhibited maximum oxygen uptake. The pesticide-treated fish shifted its peak to morning hr. Similar disruption of circadian rhythms were reported for heavy metals in fish[6].

Dramatic changes were also noticed in the aerial respiration. As in aquatic respiration, airbreathing advanced its peak to midnight hour (2400 hr). Further, ME increased the aerial respiration at all times of the day. This might be due to dependence on air-breathing organs for respiration during severe pollution stress in the aquatic environment. The overall increase of total oxygen uptake during bimodal conditions appears to be due to behavioural manifestations like hyperactivity, darting movements etc on exposure to pesticide. This may also be due to the escape behaviour of the fish from the pesticide toxicity as reflected in its aerial respiration. Obviously, this has got great ecological significance. Normally, *C. striatus* is an inhabitant of CO_2 rich waters[2]. Under these ecological conditions, greater participation of airbreathing organs may be advantageous for survival. A similar conclusion has also been arrived by Natarajan[7] in the air breathing climbing perch, *Anabas scandens* for lethal exposure to sumithion.

With regard to the decrease in oxygen uptake from water, it appears mainly due to shrinkage of the respiratory epithelium since there is no apparent cellular damage of the gill tissue except occasional swelling of the secondary lamellar tips. Recently, shrinkage of the respiratory surface area and enlargement of the water/blood barrier of the gill in the sublethal concentrations of heavy metal salts has been reported[10]. Singh and Singh[8] correlated increased opercular movement with decrease in oxygen consumption in case of *Mystus vittatus* exposed to sublethal concentrations of Zinc and Copper. They assumed that decrease in aquatic oxygen uptake was mainly due to the thickening of secondary lamellae leading to the enlargement of the water/blood pathway. Crandall and Goodnight[9] suggested that prolonged exposure of fish to low levels of pollutants subjects them to stress which causes a hormonal imbalance ultimately leading to a variety of internal pathological changes. The present disruption of circadian rhythm of bimodal oxygen uptake on exposure to metasystox may also be due to a similar mechanism which warrants further studies.

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TEMPORAL EFFECTS OF BARBITUARATES ON PREGNANCY IN ALBINO RATS

SARASWATI B. PATIL AND M. APPASWAMY RAO*
Department of Zoology, Gulbarga University, Gulbarga 585 106, India.

* Present Address: 5th Main, Yadavgiri, Mysore 570 020, India.

EXTENSIVE investigation on the effects of barbiturates in the field of female reproduction has been carried out. Barbiturates are known to block ovulation by inhibiting the pituitary gonadotrophins and prolactin release in rats[1-2]. Administration of phenobarbital or barbital sodium inhibits the ovarian compensatory hypertrophy and also interrupts pregnancy[3-4]. As pregnancy is maintained by pituitary gonadotrophins during early part and by placental luteotrophins in the later part[5,6], it is of interest to study the temporal effects of these drugs on pregnancy in albino rats.

Nulliparous, female albino rats of Hotzman strain, with regular established estrous cycle, weighing 140–150 g, 80–90 days old, were mated with fertile males at proestrus. The rats showing spermatozoa in the vaginal smears on the subsequent day were selected for experimentation and that day was designated as day 1 of pregnancy. The selected rats were laparotomized under ether anaesthesia on day 8 of pregnancy to note the number of implantations and those having normal number of implantations were selected for further treatment. Earlier tested doses of phenobarbital (7.5 mg/100 g body weight, twice a day) or barbital sodium (20 mg/100 g body weight, twice a day)[4] was injected subcutaneously in 0.5 ml saline from day 8, 10, 13 or 15 through 19 of pregnancy. Saline-treated controls were maintained for these groups. Barbiturate treatment was stopped if the profuse vaginal bleeding was observed. All rats were autopsied on day 20, the number of live foetuses was recorded and the percent foetal survival was calculated. The ovaries were weighed to the nearest mg.

The results suggest that (table 1) the effective period for the action of barbiturates ranges from day 8 to 12 of pregnancy as there is either nil or 11.1% foetal survival with respective phenobarbital or barbital sodium treatment. Administration of phenobarbital from day 10 causes complete foetal resorption, but barbital sodium treatment from day 10 is less effective as 34.5% foetal survival is seen. The drug treatment from day 13 through 19 seems to be less toxic, as the gestation is maintained with 73.8 or 92.7% foetal survival by the administration of phenobarbital or barbital sodium respectively. It is more ineffective when the barbituarates are administered from day 15 onwards, as more than 90.5% foetal survival is seen in both cases. The ovaries of the rats wherein the pregnancy is interrupted are small with many moderate-sized corpora lutea and developing follicles resembling those of nonpregnant rats, as these rats returned to estrus soon after the cessation of vaginal bleeding. But the ovaries of rats where the pregnancy is not interrupted are large, and resemble those of pregnant rats, probably due to the maintenance of large corpora lutea.

It has been demonstrated that hypophysectomy performed prior to day 11 of pregnancy promptly terminates gestation, but pituitary ablation during later half does not disturb the pregnancy in rats[5-6]. Moudgal and co-workers showed the importance of LH in maintaining the pregnancy in hypophysectomized or LH-antiserum treated rats by the administration of proper doses of LH. Studies on pheno- and pentobarbital indicate that these drugs inhibit pituitary surge of LH and also tonic release of LH, FSH and prolactin[1-2]. Therefore, in the present investigation the Interruption of pregnancy by barbiturate administration from day 8 to 10 of pregnancy is probably due to the continued inhibition of pituitary gonadotrophins and prolactin release. As these luteotrophins are essential during early part of pregnancy in rats[7-8] and subnormal production of these hormones after barbiturate treatment may cause low progesterone production resulting in the interruption of pregnancy[9]. These drugs are less effective after day 12, as the placental gonadotrophins take over the function of pituitary gonadotrophins during later half of gestation.

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