EFFECTS OF MASOTEN (DYLOX) ON PLANKTON IN EARTHEN PONDS

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Abstract: The effects of Masoten, an organophosphate parasiticide, on phytoplankton and zooplankton in earthen ponds were studied. In 2 separate trials, 3 ponds (0.04 ha) received a single application of Masoten at a rate of 0.25 mg/l (active ingredient) and 3 ponds served as untreated controls. Net plankton samples were collected at pre-treatment, and 5, 24 and 48 hrs following treatment. Toxic effects were based on quantitative-qualitative plankton analyses. Phytoplankton and rotifers were unaffected by treatment. The copepod *Diaptomus* sp. and nauplii were also unaffected. However, variability in response of copepods and nauplii to Masoten can be anticipated. Cladocerans were the most sensitive to Masoten; losses are to be expected when the compound is employed for control purposes. Information regarding residues and decomposition of Masoten is presented.

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Pennak (1953) describes the Eubranchiopoda (tadpole, fairy, and clam shrimps) as "characteristic inhabitants of temporary ponds and pools, especially during spring and early summer". Hatchery personnel who annually contend with eubranchiopod depredation might expand Pennak's description by saying that these organisms are also characteristic of the typical pondfish production season. Where shrimp populations reach nuisance proportions, they compete and interfere with fish either directly or indirectly thus complicating production pond management.

For example, all shrimp compete to some degree with fry for desirable fish-food organisms. Further, tadpole shrimp (*Apus* sp.) and clam shrimp (*Cyzicus* sp.) can induce excessive turbidities that interfere with spawning, fry collection, and photosynthesis. Newly hatched fairy shrimp (*Streptocephalus* sp.) complicate collection of fry by their similar appearance in water to fry. When shrimp are intermixed with fingerlings following harvest, valuable time is often lost during the "cleaning" process prior to shipment (Hornbeck et al. 1965, Dexter and McCarraher 1967).

Hornbeck et al. (1965) reported that fairy and tadpole shrimp could be effectively controlled in hatchery ponds with single applications of Dylox, an organophosphate insecticide, at a rate of 0.25 mg/l (active). At lower levels, Dylox is also known to be an effective control for clam shrimp (McCraren et al. unpublished data). Effects upon desirable fish-food organisms are not as well defined in the literature since most workers have been principally interested in the compound's attributes as a control agent. The impact upon plankton and benthos has been of secondary importance.

In a study designed to evaluate Dylox as a control for the anchor parasite (Lernaea sp.), Meyer (1966) reported that zooplankton was reduced in a pond repeatedly treated with Dylox at 0.25 mg/l. Two weeks following the final treatment, zooplankton were again evident in great abundance. Chironomid larvae were reported to be abundant throughout the study period. Hornbeck et al. (1965) stated that plankton populations appeared to be unaffected in largemouth bass (Micropterus salmoides) rearing ponds treated with single applications of Dylox.

This paper reports on a study of the effects of the standard application rate of 0.25 mg/l on the plankton community. It was thought that quantitative-qualitative plankton data would be of particular value in those instances where the chemical is employed for shrimp control either prior to or following the hatching of fry in spawning ponds, or in newly stocked rearing ponds where an abundance of desirable plankton must be present if optimum fry survival and growth are to be achieved. Masoten was used in this study rather than Dylox, as it is the current alternative to the latter and the active ingredient is the same.

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METHODS AND MATERIALS

Masoten (dimethyl [2,2,2-trichloro-1-hydroxyethyl] phosphonate) is an organic phosphate parasiticide developed and produced commercially by Chemagro Corporation of Kansas City, Mo. Trichlorfon is the active ingredient in Masoten (Dylox, Dipterex, Neguvon, etc.). The compound is available as an 80 percent soluble powder and is registered by EPA for use as a parasiticide on non-food fish in impounded waters. It is incompatible with alkaline materials such as lime and lime sulfur. It is subject to hydrolysis; decomposition is increased at high temperatures (25-30 C) and at a pH greater than 7.4 (Ellis 1974).

Studies conducted in 1967 with Dylox suggest that residues are apparently noncumulative. Ponds were treated once or 4 times at 0.25 and 1.0 mg/l. Samples of water, mud, and fish tissues were collected and analyzed. No residues (as DDVP) were found in samples of pondwater or bottom mud. Bluegill (*Lepomis macrochirus*) offal contained 0.02 mg/l DDVP when collected 4 hrs after a single application of 1.0 mg/l. In another study, only 0.06 mg/l DDVP was detected after 4 hrs in pondwater at pH 8.5 (room temperature) following treatment with 1.0 mg/l Dylox (Meyer 1968).

Masoten is best known as an effective control for a number of parasitic copepods and monogenetic trematodes of fishes. It has an apparent low toxicity to fish. Treatment is generally based on 0.25 mg/l applied at weekly intervals for 4 weeks (Ellis 1974).

This study was conducted in 6 0.04 ha earthen ponds located on the grounds of the San Marcos, TX state fish hatchery in April-May 1975. Each pond has an approximate volume of 12.4 kl. Well water from a single source was used to fill the ponds. Prior to treatment, water temperature was determined to be 18 C, pH was 8.7, and total hardness (expressed as CaCO₃) was 240 mg/l. The ponds were fully watered approximately 2 weeks prior to treatment. Each pond was fertilized shortly thereafter with 3.6 kg of a 16-20-0 inorganic source and .8 kg of triple superphosphate.

The study was conducted twice at San Marcos using the aforementioned single application of Masoten at 0.25 mg/l (active ingredient). On each occasion, 3 ponds served as untreated controls and 3 were treated with the 80 percent wettable powder formulation. The chemical was applied with a sprayer to ensure uniform coverage. The first pre-treatment plankton sample was collected on 3 April, 24 hr prior to treatment. Post-treatment samples were collected at 5, 24, and 48 hr. A pre-treatment sample for the second phase of study was collected on 9 April, 1 hr prior to treatment. Posttreatment samples were again collected at 5, 24, and 48 hr. Sampling frequency and duration were purposely abbreviated due to the compound's rapid degradation rate.

All samples were collected at a depth of approximately 15 cm by towing a No. 20 mesh Wisconsin type plankton net around each pond's perimeter. Length of each tow was approximately 40 m; volume of water sampled was determined to be approximately 1,600 l. To avoid the effects of diel periodicity, all samples were consistently taken within the same period. Organisms trapped in the net were washed with deionized water into 30 ml vials and immediately preserved with Lugol's solution. Each 30 ml concentrate was diluted to 200 ml prior to subsampling.

For each sample, two, 1 ml subsamples were examined in a Sedgwick-Rafter counting chamber. Each sample vial was swirled and inverted several times immediately prior to subsampling. The entire counting chamber was examined for zooplankton and phytoplankton at 100X using a stereoscopic Bausch and Lomb microscope. Filamentous forms of phytoplankton equal to or greater than 250 μ in length were counted as 1 unit. The average count was determined from the dual subsamples and organisms/l were calculated from that value.

A concentration factor (CF) was determined as follows. The original sample (i.e 1,600 l) was concentrated to 200 ml. The 1 ml subsample represented 1/200 of the original sample or 8 l. Thus, a factor of 0.12 (CF = 0.12) was required to convert the number of organisms counted in 1 ml of concentrate to number of organisms/1 (Colbert 1973).

It is assumed that some water was pushed by the net and not filtered through it. This would consistently lower the calculated value of organisms/l. Zooplankton were identified by referring to Edmondson (1959) and Pennak (1953). Phytoplankton were identified by referring to Prescott (1962).

To obtain the data points in Figs. 1-6, an average plankton density was calculated using 3 control ponds and 3 sample ponds for each sample period. Water temperature, pH and total hardness were monitored periodically during the study.

RESULTS AND DISCUSSION

While conducting a study concerning control of *Lernaea* sp. with Dylox. Rogers (1968) concluded that phytoplankton was apparently unaffected in ponds after treatment at 0.25 mg/. Based on the results of our study, we agree with his conclusion. Trends in phytoplankton population densities correlated well between treated and control ponds. Population densities at 48 hr were similar to or exceeded those at pre-treatment (Figs. 1 and 2). A variety of phytoplankton was found in all ponds during the course of study (Table 1). These results are important from the standpoint of phytoplankton's basic relationship to overall pond productivity.

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Division Chlorophyta	Order Oedogoniales
Order Volvocales	Family Oedogoniaceae
Family Volvocaceae	Oedogonium sp.
Volvox sp.	Division Pyrrhophyta
Order Ulotrichales	Order Peridiniales
Family Ulotrichales	Family Ceratiaceae
Ulothrix sp.	Ceratium hirundinella
Geminella sp.	Ceratium sp.
Order Chlorococcales	Division Cyanophyta
Family Hydrodictyaceae	Order Hormogonales
Pediastrum sp.	Family Nostocaceae
Family Scenedesmaceae	Anabaena sp.
Scenedesmus sp.	Nodularia sp.
Order Zygnematales	Family Oscillatoriaceae
Family Desmidiaceae	Oscillatoria sp.
Staurastrum sp.	Order Chroococcales
Cosmarium sp.	Family Chroococcaceae
Arthodesmus sp.	Merismopedia sp.
Family Mesotaeniaceae	Division Chrysophyta
Closterium sp.	Order Pennales
Family Zygnemataceae	FamilyTabellariaceae
Zygnema sp.	Tabellaria sp.
Spirogyra sp.	-

Although they were not specifically studying the effects of Dipterex on plankton, Sarig et al. (1965) observed that rotifers persisted in ponds after several treatments at 0.8 mg/l. Our results substantiate their observation. Rotifer numbers were essentially the same in treated ponds at 48 hr as they were at pre-treatment (Figs. 3 and 4). Rogers (1968) also reported no apparent effect on rotifers in ponds treated with Dylox at 0.25 mg/l. A wide variety of rotifers were preesnt in the ponds during the study period (Table 2).

Our results with copepods are based entirely on *Diaptomus* sp. and nauplii. Too few *Cyclops* sp. were present in the samples to allow us to make any judgments regarding their response to treatment. Overall, the *Diaptomus* sp. adults appeared to be unaffected by treatment.

Numbers of adults at termination were similar to or exceeded those present at pretreatment in treated ponds. More adult Diaptomus sp. were present in treated pond



Fig. 1. Effects of a single application of 0.25 mg/l Masoten upon phytoplankton in treated ponds (first trial)



Fig. 2. Effects of a single application of 0.25 mg/l Masoten upon phytoplankton in treated ponds (second trial)



Fig. 3. Effects of a single application of 0.25 mg/l Masoten upon rotifers in treated ponds (first trial)



Fig. 4. Effects of a single application of 0.25 mg/l Masoten upon rotifers in treated ponds (second trial)

Table 2. Taxonomic checklist of zooplankton present in ponds during course of study.

Rotifers Order Ploima Family Synchaetidae Polyarthra sp. Family Trichocercidae Trichocerca sp. Family Brachionidae Brachionus caudatus B. auadridentata Euchlanis sp. Keratella cochlearis K. hiemalis Platyias quadricornis Trichotria sp. Lepadella sp. Monostyla sp.

Copepods

Order Copepoda Family Diaptomus Diaptomus sp. Family Cyclopidae Cyclops sp.

Cladocerans Order Cladocera Family Daphnidae Daphnia sp.

samples at 48 hr in both trials than in those collected from control ponds. The abrupt decline in treated adults at 5 hr post-treatment in the first trial is not believed to be due to treatment. Unexplained oscillations are not uncommon in studies of this type and may be due in part to sampling technique.

Copepod nauplii were also apparently unaffected to any degree by treatment. In the first trial, nauplii numbers decreased in treated and control ponds by 48 hr. Fewer nauplii were present in control samples at 5 hr in the second trial than in treated samples. However, by termination, greater numbers of nauplii were tallied in control pond samples than in those from treated ponds (Figs. 5 and 6).

Published results regarding the effects of this compound on copepods are also limited. Sarig et al. (1965) reported that large numbers of Cyclops sp. persisted in ponds after several treatments with Dipterex at 0.8 mg/l. Working with *Lernaea* sp., Lahav et al. (1964) found that Dipterex at 0.5 mg/l would destroy the copepodid larvae of the parasite but not the nauplii. McCraren (unpublished data) conducted a series of static bioassays in 1972 to ascertain the effects of Dylox upon zooplankton at levels ranging from 0.0125 to 0.25 mg/l (active ingreident). Appropriate controls were used; all assays were run in duplicate. Two common copepod genera present throughout the study were *Diaptomus* sp. and *Cyclops* sp. Representatives of *Cyclops* were immobilized or dead after 48 hr at levels greater than 0.025 mg/l, whereas *Diaptomus* sp. was unaffected at all levels. Unidentified copepod nauplii responded similarly, some were obviously affected and some were not. These observations substantiate our results but perhaps more importantly, suggest that some copepods, at least at the specific level, are much more sensitive to Masoten than others.

Unfortunately, too few cladocerans were present in the study ponds to provide us with any information. However, results of a number of bioassays are available which suggest that cladocerans are the group most sensitive to Masoten. For instance, Rogers (1968) reported that concentrations of Dylox in excess of 0.1 mg/l immobilized Daphnia



Fig. 5. Effects of a single application of 0.25 mg/l Masoten upon Diaptomus sp. and nauplii in treated ponds (first trial)



Fig. 6. Effects of a single application of 0.25 mg/l Masoten upon *Diaptomus* sp. and nauplii in treated ponds (second trial)

sp. within 1 hr with death occurring after the second day. Ellis (1974) reported that levels of Chlorophos as low as 0.05 mg/l caused paralysis and death of 3 species of cladocerans. Working with concentrations of Dylox as low as 0.0125 mg/l, McCraren (unpublished data) found individuals from 7 genera of cladocerans to be either immobilized or dead after 48 hr. Results of a study by Sanders and Cope (1966) lends additional credence to the suggested overt sensitivity of cladocerans to Masoten. In their study, 2 species of daphnids were challenged with organophosphate and chlorinated hydrocarbon insecticides, acaricides, herbicides, and several other insecticides. The organophosphates were found to be generally more toxic than the chlorinated hydrocarbons to both species. Estimated 48 hr EC₅₀ immobilization values for Dipterex ranged as low as 0.0007 and 0.00032 mg/l for one species and 0.00018 mg/l for the other.

To summarize briefly, our data coupled with that of other investigators, indicates that phytoplankton is unaffected by levels of Masoten up to and including 0.25 mg/l. Rotifers appear to respond to Masoten in a similar fashion, but investigators should be aware of the possibility of variability in sensitivity between species. Copepods and nauplii are affected differently by Masoten at least at the specific level. The copepod *Diaptomus* sp. was found to tolerate Masoten at 0.25 mg/l, whereas a species of *Cyclops* sp. succumbed to levels as low as 0.025 mg/l. Of the numerous genera of cladocerans encountered in our work and that of others, all were found to be much more sensitive to Masoten than any of the other major taxonomic groups studied. Losses of cladocerans can be anticipated when Masoten is employed for control purposes of the type discussed in this paper.

Lastly, if a fish culturist is to employ Masoten knowledgeably as a control in earthen ponds, he should first attempt to recognize the problem at an early date. He should know the temperature, pH, and hardness of the pondwater in order to anticipate its effects upon degradation of the compound. Using the information contained in this paper, coupled with an understanding of the treatment rates required to control various problematical organisms, he should be in a position to anticipate the effects of Masoten on plankton and how they will relate to the requirements of the fish he is attempting to culture. If time is available, much could be learned by conducting a series of onsite bioassays.

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