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Use of TFM (3-Trifluoromethyl-4-Nitrophenol) to Selectively Control Frog Larvae in Fish Production Ponds

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Abstract.—The efficacy of TFM (3-trifluoromethyl-4-nitrophenol) for the selective control of frog larvae in fish culture ponds was examined. Mortalities of frog larvae and fathead minnows (*Pimephales promelas*) in exposure cages as well as end-of-season standing crops were used to quantify the selective effects of TFM in three treatment ponds, A, B, and C. The chemical was completely effective in controlling frog larvae in treated ponds A and B; no tadpoles were recovered after treatment. These ponds were filled just before the study. The standing crop of tadpoles in the untreated control pond, which was also filled just before the study, was 219.7 kg/hectare. Treatment of pond C, which was filled throughout the year and had an established population of older, larger tadpoles before TFM application, was not effective, leaving 243.3 kg frog larvae per hectare). Exposure-cage mortalities of frog larvae ranged from 22.0 to 95.0%. Failure to kill all frog larvae was most likely due to insufficient TFM concentration for the life stage of the tadpoles treated. The half-life of TFM was 10.3 d in ponds A and B, and 20.1 d in the more sediment-laden pond C. No stratification of the toxicant was observed in any of the ponds. Results indicate that TFM effectively controls infestations of frog larvae if applied to ponds when the tadpoles are relatively young or newly hatched. Application of TFM will probably not selectively control tadpoles at older life stages.

Populations of frog larvae in warmwater fish culture ponds have long caused a variety of problems. Sorting and removal of tadpoles in harvest nets are time-consuming and subject fish to severe mechanical injury and stress. In addition, frog larvae effectively compete for food resources (Dickman 1978; Seale 1980) and make production quantification difficult. Tadpole biomass in production ponds has been reported to be as high as 2,242 kg/hectare (Prather et al. 1953). In the bait-fish production ponds in our study, tadpoles often constituted 50–75% of the harvest biomass (Figure 1).

Current methods used to control tadpoles include hand removal or poisoning of egg masses, mowing pond banks to reduce frog habitat, using ducks to eat the eggs and larvae, and killing the adult frogs. These methods are costly, often haphazard, and not highly effective. Helms (1967) reported limited success with formalin to control tadpoles in fish production ponds. Formalin treatments, however, caused oxygen depletion as well as incomplete tadpole mortality. Formalin has been used to separate tadpoles and fingerling largemouth bass (*Micropterus salmoides*) in raceways (Carmichael 1983). This technique, although economical and coincidental to ectoparasite treatment, does not alleviate harvest and preharvest difficulties. At present, there is no safe and effective method for controlling tadpoles in fish culture ponds (Piper et al. 1982; Kane et al. 1985).

The lampricide TFM (3-trifluoromethyl-4-nitrophenol) has been used to control sea lampreys (*Petromyzon marinus*) in the Great Lakes of North America since 1958 (Applegate et al. 1961). Howell (1966) reported that TFM may be selectively toxic to amphibians. In his study, mud puppies (*Necturus maculosus*) appeared to be as susceptible to TFM as lampreys. Gilderhus and Johnson (1980) noted mortality of mud puppies as well as frog larvae during stream treatments with TFM. Results of static bioassays conducted with larvae of both grey tree frogs (*Hyla versicolor*) and leopard frogs (*Rana pipiens*) and flow-through tests with larval bullfrogs (*R. catesbeiana*) were reported by Chandler and Marking (1975). In these tests, the acute toxicity of TFM to tadpoles was 1.2–8.2 times greater than the toxicity to selected fish species reported by Marking and Olsen (1975). These conclusions were confirmed by further studies in which TFM was four times more toxic to bullfrog larvae than to fathead minnows (*Pimephales promelas*; Kane et al. 1985). The chemical is photodegradable (Carey and Fox 1981) and, under controlled usage, is not toxic to mammals or waterfowl (NRCC 1985).

In this study, we examined the efficacy of TFM as a selective amphibicide in fish culture ponds. End-of-season standing crops as well as mortali-

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FIGURE 1.—Net haul from a tadpole-infested baitfish production pond. Dark-colored organisms are tadpoles.

ties of frog larvae and fathead minnows held in exposure cages were used to quantify the selective effect of TFM. Persistence of TFM in shallow, static impoundments was evaluated by determining its postapplication distribution and half-life in the different ponds.

Methods

Four central Ohio fish ponds known to have tadpole infestations were used. Ponds A and B and the control pond were adjacent, drainable hatchery ponds measuring 0.41 hectares. Pond C was privately owned, filled year-round, and had an established tadpole population. It measured 0.03 hectares and was somewhat turbid due to suspended sediment.

Initial TFM application rates for ponds A and B (4.0 mg/L) and pond C (7.5 mg/L) were based on prior tests with fathead minnows, bluegills (*Lepomis macrochirus*), bullfrog larvae, and larval green frogs (*Rana clamitans*) in on-site bioassays. These tests were conducted in 250-L polyethylene bags suspended in the ponds (Burress 1975). The 4.0-mg/L pond treatments, however, killed only a few test animals held in exposure cages suspended in the ponds, so the TFM concentrations in ponds A and B were raised to 7.5 mg/L 3 d after the first toxicant application.

Field-grade TFM (35.7% active ingredient) diluted 1:4 with pond water was used. A uniform application was made subsurface with a pressure sprayer from a boat zigzagging across the ponds. The TFM was applied in early summer to ponds A and B. Exposure cages in ponds A and B and the control pond consisted of 18.9-L covered buckets drilled with enough 9.5-mm holes to yield approximately 20% open surface. Thirty-six exposure cages were suspended throughout each pond. Each cage held 20 young bullfrog larvae and 20 fathead minnows, which were collected by seining from a nearby private farm pond. Standing crops in these ponds were determined by hand-counting the remaining fish and frogs after ponds were drained in midsummer.

In pond C, TFM was applied in late summer. A single floating mesh-net box (1.0 × 1.3 × 1.0 m) anchored at the center of the pond served as an exposure cage. The container held 100 green frog larvae, 100 second-year bullfrog larvae, and 65 adult fathead minnows, all of which were resident in the pond and collected by seining. The

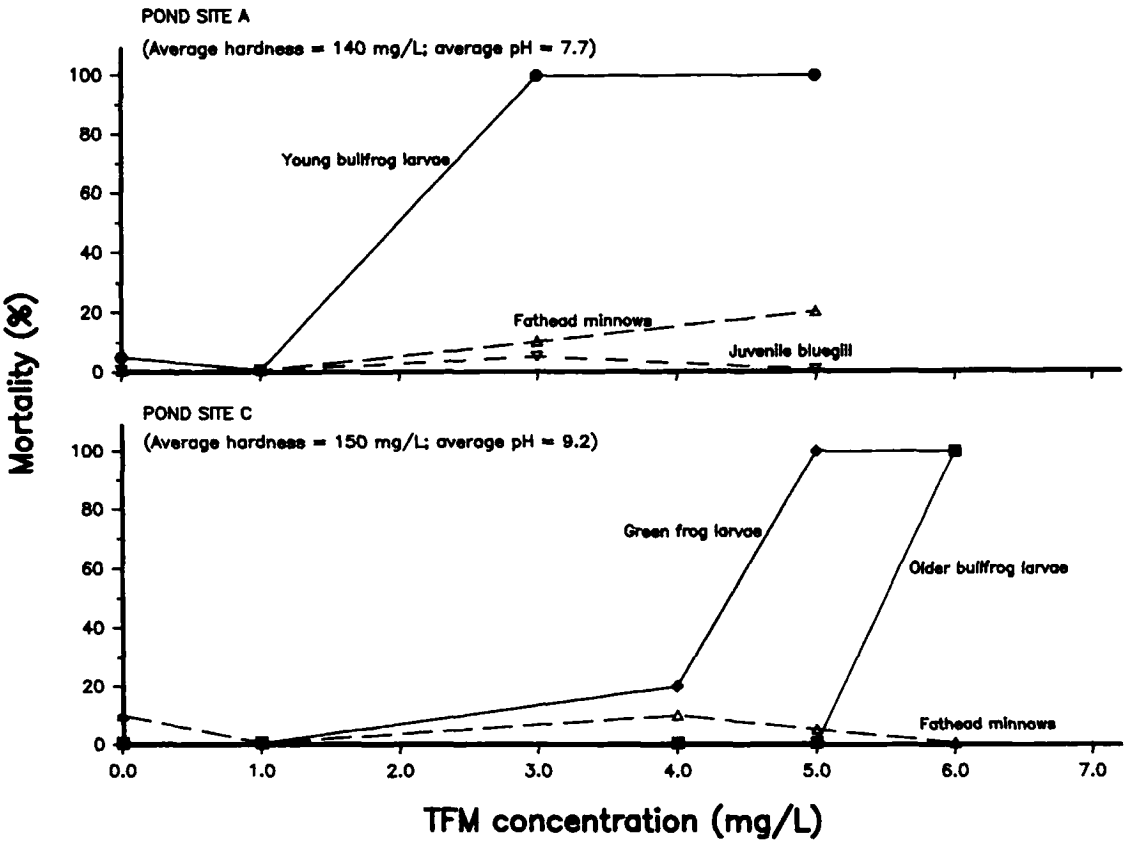

FIGURE 2.—Mortalities of fathead minnows, juvenile bluegills, and frog larvae exposed to different concentrations of TFM. Bioassays were conducted with ambient pond water in 250-L polyethylene bags suspended in different ponds (A and C).

removal method of population estimation (Zippen 1958) was used to estimate standing crop in the whole pond in early fall.

The TFM concentrations in the ponds were determined spectrophotometrically (Olsen and Marking 1973). Dissolved oxygen and pH were measured with a membrane electrode, and total hardness (as $CaCO_3$ equivalents) was measured with Bausch and Lomb Spectrokits® and EDTA titration (APHA et al. 1980).

Results

On-Site Bioassays

Bag tests conducted in pond A, which had water quality similar to that in pond B and the control pond, showed complete mortality of young (age-0) bullfrog larvae at 3.0 mg TFM/L, whereas no more than 10% mortality of fathead minnows resulted. The bag test done in pond C yielded complete mortality of green frog larvae and older (age-1) bullfrog larvae at 5.0 and 6.0 mg TFM/L, respectively, whereas mortality of fathead minnows again did not exceed 10% (Figure 2).

Mortalities in Exposure Cages

Exposure cage mortalities were cumulative over 30 d in ponds A and B and the control pond, and over 14 d in pond C. Mortality was 45–75% for young bullfrog larvae in pond A and B exposure cages. Mortality of older bullfrog larvae and green frog larvae in the pond C exposure cage was 22 and 95%, respectively. Mortality of fathead minnows in the exposure cages in ponds A, B, and C was 16.9, 22.4, and 28.0%, respectively. Mortality of fathead minnows and young bullfrog larvae in the untreated control pond exposure cages was 2.5 and 7.9%, respectively (Figure 3).

End-of-Season Standing Crops

The TFM significantly suppressed the seasonal production of frog larvae in drainable ponds A

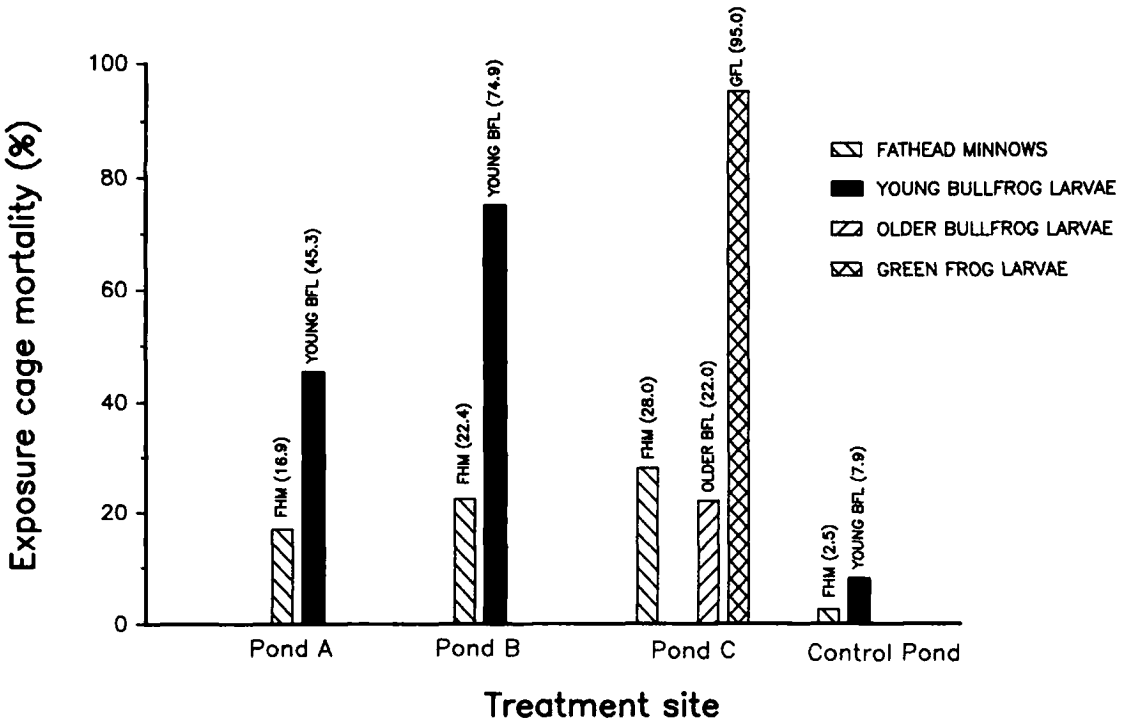

FIGURE 3.—Exposure-cage mortalities of frog larvae and fathead minnows in three ponds treated with 7.5 mg TFM/L and in an untreated control pond.

and B; when ponds were drained after treatment, standing tadpole crops were 0.0 kg/hectare in ponds A and B, but 219.7 kg/hectare in the untreated control pond ($P < 0.01$). Treated pond C, which had a previously established population of frogs, had a crop of 243.3 kg tadpoles/hectare when it was drained (Figure 4).

Toxicant Distribution and Water Quality

Distribution of TFM in treated ponds A, B, and C 12 h after application was relatively even vertically and horizontally (±0.5 mg/L). Within 24 h, concentrations were homogeneous (±0.1 mg/L) throughout the ponds. The degradation of TFM in the ponds followed first-order kinetics (Figure 5). The half-life of the toxicant was 10.3 d in ponds A and B and 20.1 d in the moderately turbid pond C.

Diurnal measurements of dissolved oxygen (taken at middepth from all ponds) ranged from 8.0 to 13.0 mg/L, and temperatures ranged from 16 to 25°C. Predawn levels of dissolved oxygen in ponds A and B and the control pond ranged from 4.2 to 7.3 mg/L at the shallow end and from 7.6 to 11.0 mg/L at the deeper end. Dissolved oxygen in on-site bioassay bags and in exposure cage buckets in ponds A and B was at times not more than 20% of pond levels. Dissolved oxygen levels in the ponds were unaffected by TFM treatment.

Discussion

Treatment of ponds A and B with TFM was highly effective for controlling tadpoles. This was not the case for pond C or the exposure cages in all of the treatment ponds. The resident tadpoles in pond C and those collected for exposure cage testing all had a developed operculum, but their limb buds were not yet present or just barely appearing (stage 25-26; Gosner 1960). Frog eggs or newly hatched and very young tadpoles (stages 1–17) appear to be more susceptible to the toxicant than older tadpoles. This is supported by comparison of pond production data with exposure-cage mortality in this study and previous studies (Kane et al. 1985) in which younger bullfrog larvae succumbed at lower TFM concentrations than older ones.

Although ponds A and B and the control pond were not stocked with finfish after the ponds were filled, 25–35-mm gizzard shad (*Dorosoma cepedianum*) were present in the end-of-season harvests. These fish were introduced through the fil-

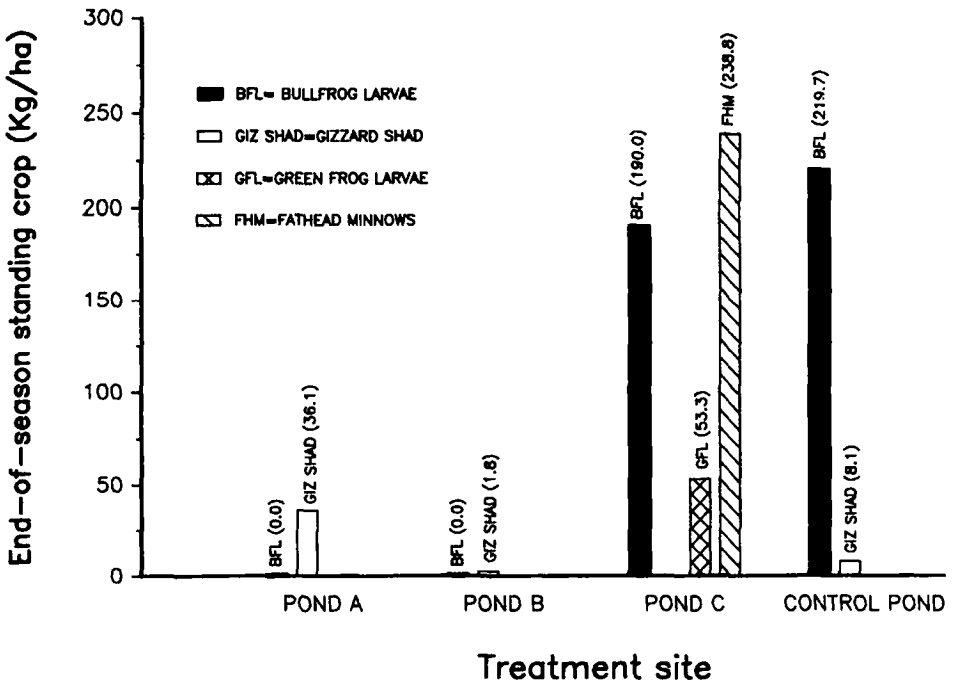

FIGURE 4.—End-of-season standing crops (kg/hectare) of fish and frogs in ponds treated with 7.5 mg TFM/L and an untreated control pond.

tration socks during pond filling. Gizzard shad production, although low, appeared to be unaffected by the TFM treatment.

The mortality of fathead minnows in all pond exposure cages was unacceptably high (16.9–28.0%). Depressed dissolved oxygen levels (≤20% of pond levels) in exposure cages (buckets) that had high tadpole mortality indicated retarded water circulation within the cages. This, in part, may have accounted for the elevated fathead min-

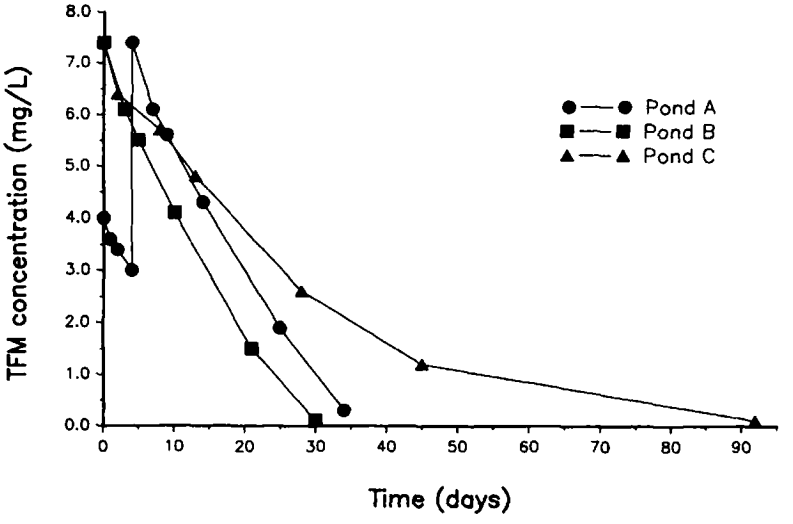

FIGURE 5.—Degradation of TFM in three fish culture ponds. Ponds A and B were initially treated with 4.0 mg TFM/L then treated again to increase the TFM concentration to 7.5 mg/L. (Pond B data are displayed, for clarity, 3 d before measurements were actually made.) Pond C was treated once with 7.5 mg TFM/L.

now mortality in cages in ponds A and B. Capture, transport, handling, and confinement stress may have also contributed to increased susceptibility of the fish to the toxicant. In pond C, a columnaris outbreak occurred just before TFM appliction and may have made the fish more sensitive to the toxicant.

The on-site bioassays did not accurately predict mortalities in the pond and exposure cages, although they did indicate the relative toxicities of TFM to the organisms tested. Low dissolved oxygen levels in the bags (<20% of pond levels) as well as confinement stress may have enhanced tadpole mortalities in the bag tests.

Pond distributions of TFM were relatively even within 12 h and homogeneous within 24 h. In contrast, Scott et al. (1984) found that TFM approached homogeneity in small, inverse-pyramidal, plastic-lined ponds after 72 h. The more gradual mixing in their ponds was most likely due to a relatively low surface-to-volume ratio. In our ponds, which are more representative of hatchery ponds, gentle wind action was sufficient to evenly distribute the toxicant.

The half-life observed in ponds A and B (10.3 d) agrees with the observed 11-d half-life in ponds reported by Scott et al. (1984). Our data also closely agree with the predictive values (Zepp and Cline 1977) used by Carey and Fox (1981): for a mean pond depth of 1 m with autumn solar incidence, half-life is about 10 d. The extended half-life of TFM observed in pond C was most likely due to less available light for photodegradation due to the suspended sediment (Carey and Fox 1981) and to the gradual desorption of the toxicant over time as it was released from the fine sediment, which was relatively high in organic matter (Dawson et al. 1986).

In conclusion, TFM can be used to control young tadpoles in fish ponds, but the larvicide probably will not selectively control older tadpoles. Previous work (Kane et al. 1985) indicated that earlier tadpole stages may be controlled with a much lower dose of the toxicant than we used. Therefore, it is plausible that the application concentration used in this study (7.5 mg/L) exceeded the necessary concentration for the effective control of newly hatched frog larvae. The degree of selectivity of the toxicant will depend on several variables, including the amount of toxicant used, the fish being cultured (Marking and Olsen 1975), physical characteristics of the pond (i.e., sediment type and organic content; Dawson et al. 1986), and water quality (Dawson et al. 1975).

Use of TFM has been registered solely for lamprey control in the tributaries of the Great Lakes by authorized Great Lakes Fishery Commission personnel. The toxicant would need to be relabeled by the U.S. Environmental Protection Agency for use as a *selective amphibicide* in static culture ponds before it could become available for use as described here. Additional considerations regarding the future of TFM as an amphibicide include economic feasibility.

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