quitoes: Isolation and identification of oviposition repellents for *Culex* mosquitoes. J. Chem. Ecol. 6:71–80.

Hwang, Y.-S., G. W. Schultz, H. Axelrod, W. L. Kramer and M. S. Mulla. 1982. Ovipositional repellency of fatty acids and their derivatives against *Culex* and *Aedes* mosquitoes. Environ. Entomol. 11:223–226.

Kramer, W. L. and M. S. Mulla. 1979. Oviposition attractants and repellents of mosquitoes: Oviposition responses of *Culex* mosquitoes to organic infusions. Environ. Entomol. 8:1111–1117.

Kramer, W. L., Y.-S. Hwang and M. S. Mulla. 1980. Ovipositional repellents of mosquitoes: Negative responses elicited by lower aliphatic carboxylic acids. J. Chem. Ecol. 6:415–424.

Mulla, M. S. and H. A. Darwazeh. 1981. Efficacy of petroleum larvicidal oils and their impact on some aquatic nontarget organisms. Proc. Calif. Mosq. Vector Control Assoc. 49:84–87.

Schultz, G. W., Y.-S. Hwang, W. L. Kramer, H. Axelrod and M. S. Mulla. 1982. Field evaluation of ovipositional repellents against *Culex* mosquitoes. Environ. Entomol. 11:968–971.

SURVIVAL AND PREDATORY EFFICIENCY OF *GAMBUSIA AFFINIS* FOR CONTROL OF MOSQUITOES IN UNDERGROUND DRAINS

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ABSTRACT. Mosquitofish, *Gambusia affinis,* survived in an impounded water area within an underground storm drain system in Fresno, CA through a 14-wk period during the summer of 1982. The number and relative condition (K-factor) of the stocked fish slowly declined with the depletion of the food supply, as determined by fish gut content analysis, at the tenth week after introduction. However, the mosquitofish were effective in reducing the number of adult *Culex quinquefasciatus* produced in the drain system. Reductions of 75, 89 and 94% below those of an untreated control area were obtained after the first, second and third month, respectively. While female mosquitofish (gravid before introduction) produced offspring, no mating of fish within the drain was found.

INTRODUCTION

As urban sprawl displaces agriculture and destroys natural habitats, the production of some mosquito species is diminished. With a reduction in breeding habitat, such species as *Culex tarsalis* Coquillett and the floodwater *Aedes* spp. decrease in numbers. This is not the case for *Cx. quinquefasciatus* Say, which is proliferating in the breeding habitats provided by urbanization. A major breeding source in the urban environment is the underground storm drain system.

Chemical applications in storm drains cannot be solely relied upon because of the potential for insecticide resistance (Mulligan and Schaefer 1981). Physical and biological methods which show potential should be incorporated into an integrated control program. One example is a physical barrier system which prevents access of mosquitoes to potential breeding areas within the drain line (Mulligan and Schaefer 1982).

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Complementing this integrated approach is the preliminary report by Farley and Caton (1982) on the success of the mosquitofish, *Gambusia affinis* (Baird and Girard) in underground drains. The following report is a further investigation into the predatory efficiency and survival potential of *G. affinis* in underground storm drain lines in Fresno, CA.

MATERIALS AND METHODS

The study areas were sections of a single, underground storm drain line, totaling ca. 3.5 km in length (Fig. 1). Two separate water impoundments were formed by flow barriers in the drain line. One impoundment included the entire upper 1.5 km portion of the line. A second, 0.7 km long impoundment was formed above a barrier located 1.3 km downstream from the first.

Adult mosquito populations were monitored by collections with miniature, CDC-type traps (Bio-Quip #2802 EVS) without CO_2 bait. Traps were operated overnight (ca. 1600 hr–0900 hr) with new batteries at each use. A single trap was placed in three manhole chambers in the two water impoundment areas. Species and num-

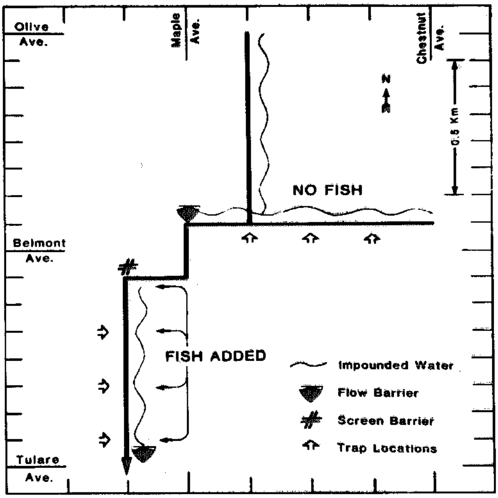

Fig. 1. Location of water impoundments and sampling sites within an underground storm drain line in Fresno, CA. *Gambusia affinis* were released at a rate of 2.82 kg fish per km drain line.

bers of each sex were determined in the laboratory. When a collection was large (<1000), the number was estimated by counting and weighing a subsample. Collections were made on a weekly basis from June through October 1982.

Unsorted, adult mosquitofish, *G. affinis,* were evenly distributed at four sites (manhole chambers) in the downstream impoundment on July 13, 1982. The release rate was 2.82 kg of fish per km of drain line (10 lb/mile). The upstream impoundment served as an untreated control area. Discontinuity of water between the two impoundments prevented movement of fish from the downstream to the upstream area. As a precaution, screening was placed across the drain line at the upper end of the downstream impoundment.

Every other week, modified Gee® minnow traps were placed at three locations in each study area to assess the fish population. The interiors of the 40 × 22 cm diam, barrel-shaped traps were lined with 1.5 mm window screen. A 3 volt light bulb was suspended 0.75 m above the trap. Traps were left in the line for 24 hr.

Once a month a fish sample was netted from the drain line for gut content analysis and determination of relative condition of the fish population. Relative condition (K-factor) was determined by the formula:

$$K = W \times 100/L^3,$$

where W = weight in grams and L = length in centimeters. The K factor can be used to determine changes in the physical condition of a fish population over time (Lagler 1956). A decreasing K-factor indicates a deterioration in the condition of the fish. K-factors for post-stocking collections were compared with pre-stocked fish at similar lengths and reproductive status to minimize variation caused by these factors.

A quality analysis of the impounded water in the fish study area was made. Dissolved oxygen was recorded with a YSI Model 51 oxygen meter. Total ammonia was determined colorimetrically with a chemical test kit (Hill's Kordon, Aquatru Test, Cat. No. 35510).

RESULTS

Monthly means, ± SE, were derived from weekly, triplicate captures of adult mosquitoes for each of the two study areas (Fig. 2). The only mosquito species collected within the drain line was *Cx. quinquefasciatus.* Mosquito production developed slower in the area lacking fish, however, by July there was no significant difference ($P>0.05$) between the means of the two areas by analysis of variance (ANOVA). After the addition of fish in mid-July, the numbers of adult mosquitoes captured were reduced from August through October. While mosquito numbers also declined through September in the area without fish the numbers in the area with fish were significantly lower (ANOVA, $P<0.05$). Capture means in the area with fish were reduced 75, 89 and 94% below those of the area without fish for August, September and October, respectively.

During the course of the study, no fish were captured from the unstocked, upstream area. Results of minnow trap collections from the

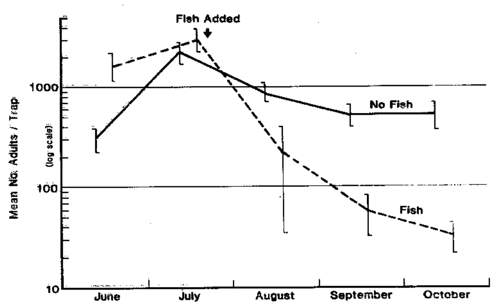

Fig. 2. Monthly mean numbers ± SE of *Culex quinquefasciatus* adults captured from two areas of impounded water within a Fresno, CA underground storm drain line. *Gambusia affinis* were released at a rate of 2.82 kg fish per km drain line.

Table 1. Number, by age group, of stocked *Gambusia affinis* captured with minnow traps from three locations in a drain line.

	Number of weeks after release of fish							
	0	2	4	6	8	10	12	14
	Mean no. fish ± SE/trap							
Adults	32 ± 16	3 ± 2	8 ± 6	7 ± 4	10 ± 8	4 ± 2	3 ± 2	1 ± 1
Juveniles	0	0	2 ± 2	3 ± 2	2 ± 2	3 ± 2	0	0
Fry	1 ± 1	6 ± 3	1 ± 1	4 ± 2	0	0	0	0

downstream area are shown in Table 1. At one day after release, before dispersal of the fish, a high mean of 32 adult fish were captured. Subsequent numbers of adult fish captured were lower, but remained fairly constant through 8 weeks. A drop in numbers occurred at 10 weeks and numbers gradually declined thereafter. Fewest fish were taken at 14 weeks. Fry were collected at each trapping date through 6 weeks after stocking; however, no fry were collected subsequently. Consistent numbers of juveniles were captured from 4 to 10 weeks after release of fish.

Results of the relative condition determinations and gut content analysis of netted fish are shown in Table 2. Prior to introduction of the *G. affinis* into the study area, the mean K-factor for females was 1.15. Thereafter the K-factor gradually decreased to 0.97 and 0.98 for 10 to 14 weeks respectively. The numbers of males netted in the samples were low after the second week.

Fish netted from the drain fed mainly on mosquitoes and ostracods at 2 weeks after release. By 6 weeks mosquito consumption had diminished and the fish were feeding heavily on copepods, ostracods and psychodid immatures. Gut content was minimal by the tenth week and was chiefly mosquito in origin.

A mean ammonia (NH_3) concentration of 0.04 mg/liter (range 0.03–0.06 ppm) was calculated from total ammonia readings in the impounded water areas. Dissolved oxygen ranged from 0.5–4.0 ppm, with an average of 2.5 ppm.

DISCUSSION

Gambusia affinis were effective in reducing adult *Cx. quinquefasciatus* numbers in a water impoundment area of an underground storm drain line. Gut content analysis of collected specimens confirmed fish predation upon mosquito immatures. At the onset of the study, mosquitoes, and to a lesser degree ostracods, formed the major part of the diet. Mosquitoes continued to be found in the fish guts through the last sampling date.

With an abundance of prey organisms through the 6-week period after stocking, the condition of the fish was excellent with K-factors well above 1.00. By the tenth week, gut content analysis showed the food supply to be depleted and, as a result, the condition of the fish had significantly decreased. Corresponding to the reduced food supply, the numbers of fish trapped began to decline. Depletion of food and resultant emigration of fish from the study area were probably the causes for this decline. A contributory factor, possibly resulting in loss of fish to downstream areas, was the occurrence of 5.6 mm of rainfall immediately prior to collection of fish trap samples at the tenth week.

Table 2. Relative condition (K-factor) and gut content analysis of adult *Gambusia affinis* netted from a storm drain line after initial stocking.

	Number of weeks after release of fish				
	Prerelease	2	6	10	14
	Mean K-factor (sample size)				
Males	0.89 (20)	1.03 (8)	1.09 (2)	0.74 (1)	0.68 (1)
Females	1.15 (35)	1.04 (7)	1.08 (10)	0.97 (7)	0.98 (3)
	Mean no. organisms/fish gut				
Culicidae		6.1	0.5	1.1	
Psychodidae		0.7	3.8	0	
Ostracoda		4.7	6.8	0	
Copepoda		0	20.8	0	
Mollusca		0.1	0.2	0.1	
Unidentified[1]		0.2	0.7	0.3	

[1] Category includes terrestrial organisms and extraneous matter.

Exhaustion of the food supply was probably inevitable in such a confined and simplified habitat, regardless of the initial stocking rate.

Production of fry ceased after the sixth week and fertile females were not found after that time. This confirmed the report by Farley and Caton (1982) that mating does not occur in the drain lines. The cause of cessation of mating in the drain, whether an influence of the environment on behavior or the effect of a dwindling food supply, was not determined.

While neither the ammonia level nor the dissolved oxygen concentration was within a known toxic range for *G. affinis,* both were outside their respective criteria (0.02 ppm NH_3 and 5.0 ppm dissolved oxygen) set by the U.S. Environmental Protection Agency for optimum fresh-water aquatic life (U.S. Environmental Protection Agency 1976). However, morphological adaptation in *G. affinis,* enabling survival in oxygen deficient waters has been shown by Lewis (1970). Moreover *G. affinis* survived in the drain environment and effectively suppressed mosquito production. If used where impoundment areas produce contiguous water, *G. affinis* can be an important component in an IPM program of mosquitoes which breed in underground storm drain lines.

ACKNOWLEDGMENT

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References Cited

Farley, D. G. and J. R. Caton. 1982. A preliminary report on the use of mosquitofish to control mosquitoes in an urban storm drain system. Proc. Calif. Mosq. Vector Control Assoc. 50:57–8.

Lagler, K. F. 1956. Freshwater fishery biology. Wm. C. Brown Co., Dubuque, IA. 421 p.

Lewis, W. M., Jr. 1970. Morphological adaptations of Cyprinodontoids for inhabiting oxygen deficient waters. Copeia 2:319–26.

Mulligan, F. S., III and C. H. Schaefer. 1981. The breeding of *Culex quinquefasciatus* within the Fresno urban storm drain system. Proc. Calif. Mosq. Vector Control Assoc. 49:101–3.

Mulligan, F. S., III and C. H. Schaefer. 1982. A physical barrier for controlling mosquitoes which breed in urban storm drains. Mosq. News 42:360–5.

U.S. Environmental Protection Agency. 1976. Quality criteria for water. U.S. Gov. Printing Office, Washington, DC. 256 p.