

SPATIAL DISTRIBUTIONS OF *ANOPHELES FREEBORNI*, *GAMBUSIA AFFINIS* AND *LEPOMIS CYANELLUS* IN EXPERIMENTAL RICE PLOTS

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The mosquitofish, *Gambusia affinis* Baird and Girard, has been shown to be quite effective at reducing mosquito populations in rice fields in some experiments (Hoy and Reed 1971, Hoy et al. 1971) but quite ineffective in other experiments (Ahmed et al., 1970, Hoy et al. 1972, Cech and Linden 1987, Kramer et al. 1987, Blaustein 1989a). Assessments of green sunfish, *Lepomis cyanellus* Rafinesque, as regulators of mosquito populations in rice fields have also yielded inconsistent results (Davey et al. 1974, Davey and Meisch 1977, Blaustein 1988², 1989a). For a biological control agent to be effective at reducing a pest population, it must have high spatial overlap with the pest. Part of the inconsistency of larvivorous fishes to control mosquitoes in rice fields may be due to differences in overlap between predator and prey in different fields. Factors such as water depth and type and density of vegetation vary considerably among rice fields and could cause overlap between mosquitoes and fishes to vary.

In a field experiment, I found that mosquitofish, which were stocked at a high rate (2,318 adults [1:1 sex ratio] per ha) and reached high densities, failed to control mosquitoes in rice plots (Blaustein 1989a). In the same study, I was also unable to demonstrate control by green sunfish stocked at the same rate. I suggested that the inefficiency of these predators to control mosquitoes may have been due, in part, to low spatial overlap between predators and prey as a result of environmental heterogeneity and the dense submergent vegetation of these rice plots which may have provided refugia for immature mosquitoes.

I selected two of the experimental rice subplots from this study which contained high densities of fishes and *Anopheles freeborni* Aitken to determine if a low spatial overlap between the populations of the mosquito and the fishes may have contributed to the inability of these two fishes to reduce *An. freeborni* populations.

The description of the experimental rice plots used in this study are described in detail elsewhere (Blaustein 1989a). Briefly, six plots, each subdivided into four 83-m² subplots, were maintained during the 1982 rice season. Two subplots from one of these plots were chosen for the study. Both subplots contained high densities of *An. freeborni*. One subplot, hereafter referred to as subplot 1, contained high densities of green sunfish, while a second (subplot 2) contained high densities of mosquitofish and a lower density of green sunfish.

Subplot 1 contained a very shallow borrow pit on the south end, ranging in depth from 15 to 18 cm. It was devoid of emergent vegetation, but submergent vegetation (*Najas* sp. and *Chara* sp.) covered nearly 100% of the substrate and frequently reached the air-water interface. The southern half of the subplot contained a heavy stand of short-stem rice and ranged in depth from 13 to 16 cm. The northern half of the plot was deeper (15–25 cm) and was devoid of emergent vegetation but had submergent vegetation (mostly *Chara* sp.) covering ca. 75% of the substrate which did not reach the surface.

The north end of subplot 2 contained a borrow pit similar in structure to the borrow pit in subplot 1 (11–18 cm in depth). Short-stem rice, other emergent vegetation and submergent vegetation grew in the remainder of the subplot.

On September 1, 1982, 1000 h, a minnow trap (0.32 cm mesh) was placed in each of 13 stations in subplot 1 in four habitats: 1) an open water area devoid of emergent vegetation (4 traps), 2) the margin of the open area and a rice stand (3 traps), 3) a dense rice stand (3 traps) and, 4) a borrow pit (3 traps). At the same time, minnow traps were set up in subplot 2 at 25 stations in a grid of five rows and five columns in two habitats: a borrow pit contained 5 traps (one column), and the rice stand contained 20 traps (four columns). At each station, the depth was recorded, and populations of mosquitoes were determined by taking 10 dips with a standard 0.4 liter dipper. Approximately 4.5 hours later, the traps were lifted and the fish were counted. None of the traps contained any of the adult sunfish.

Previously, I found that trapping efficiencies of these fishes were not affected by vegetation but were affected by water depth (Blaustein

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² Blaustein, L. 1988. Biological interactions in rice fields: a community-ecology approach to mosquito control. Ph.D. Dissertation. Univ. of California, Davis.

1989b). However, the effects on trapping efficiency here do not change the qualitative results.

Because data from the replicated stations within a subplot were not independent, inferences drawn from results on spatial distributions apply only to these plots. In each subplot, water depth, numbers of *An. freeborni* (the sum of the 10 dips) and numbers of the fishes at each station were correlated (Spearman rank). Analyses of variance were used to determine the effect of the habitats on the natural log-transformed numbers ($\ln[x + 1]$) of *An. freeborni* and the fishes. Analyses of covariance, using the fishes as covariates, were conducted to determine if *An. freeborni* were more abundant in certain habitats in the absence of fish. Since water depth varied with the different habitats recognized, it was also entered into the model as a covariate.

Green sunfish and *An. freeborni* numbers were negatively correlated in the 13 stations of subplot 1 ($P < 0.10$, Table 1). Neither sunfish nor mosquito numbers were correlated to depth ($P > 0.10$). Significantly fewer sunfish were caught in the rice than the other three habitats ($F_{3,9} = 4.78$, $P < 0.05$; Fig. 1) even after controlling for depth ($F_{3,8} = 6.59$, $P < 0.05$). *Anopheles freeborni* densities were greatest in the rice and borrow pit habitats, significantly lower in the rice-open water margin and even significantly lower in open water ($F_{3,9} = 15.13$, $P < 0.001$; Fig. 1). This same relationship held after controlling for depth ($F_{3,8} = 16.89$, $P < 0.001$), sunfish ($F = 11.10$, $P < 0.01$), and both sunfish and depth ($F_{3,7} = 10.64$, $P < 0.01$).

Anopheles freeborni densities were negatively correlated with trap abundance of both fishes in subplot 1 ($P < 0.05$; Table 1). Depth and mosquitofish abundance showed a significant positive correlation ($P < 0.001$). More green sunfish were caught in the borrow pit than in the rice stand ($F_{1,23} = 9.66$, $P < 0.01$; Fig. 2) even after controlling for depth ($F_{1,22} = 6.33$, $P < 0.05$). Trap abundances of mosquitofish were greater in the deeper borrow pit than the shallower rice

stand ($F_{1,23} = 8.56$, $P < 0.01$), but after controlling for depth, habitat was no longer significant ($F_{1,22} = 0.79$, $P > 0.10$; covariate: $F = 2.89$, $P > 0.10$).

Anopheles freeborni were not distributed with respect to habitat ($F_{1,23} = 1.80$, $P > 0.10$; Fig. 2), even after controlling for depth, sunfish, mosquitofish, depth plus sunfish, depth plus mosquitofish and all three covariates (all ANCOVA models: $P > 0.10$).

Immature green sunfish prefer vegetation to open water in ponds (Werner and Hall 1976, Werner 1984). The results here do not contradict these pond studies because the other areas outside the rice zone (including the "open-water" habitat) had a fairly dense mat of submergent vegetation. However, I have also found green sunfish immatures to be more abundant in deeper borrow pits with little submergent vegetation than in rice stands (Blaustein 1988²).

It is not discernable whether the higher numbers of mosquitofish caught in the borrow pit than in the other habitats were due to deeper water or to some other characteristic (such as type of vegetation) which differed among habitats. Other studies have also shown mosquitofish to increase in numbers with increasing depth (Reed and Bryant 1972) and to have higher densities in the borrow pit (Norland and Bowman 1976, Coykendall 1981, but see Blaustein 1988²).

Few mosquito larvae would be expected in borrow pits of most rice fields since they rarely have vegetation that reaches the surface. However, the borrow pits of this study were shallow and contained a lot of submergent vegetation that reached the surface. The lower densities found at the rice-open water margin and the open water were expected since *An. freeborni* are generally associated with vegetation (Hess and Hall 1943).

The negative correlations and analyses of variance in both subplots show that *An. freeborni*, in general, are more abundant where fish are not abundant. This information alone cannot

Table 1. Spearman rank correlations for 13 stations in subplot 1 and 25 stations in subplot 2.

	<i>An. freeborni</i>	<i>L. cyanellus</i>	Depth
Subplot 1 (n = 13)			
<i>An. freeborni</i>			
<i>L. cyanellus</i>	-0.48*		-0.38
Depth			0.16
Subplot 2 (n = 25)			
<i>An. freeborni</i>			
<i>L. cyanellus</i>	-0.39**		-0.28
<i>G. affinis</i>		0.31	0.32
Depth			0.67***

* $P < 0.10$

** $P < 0.05$

*** $P < 0.01$

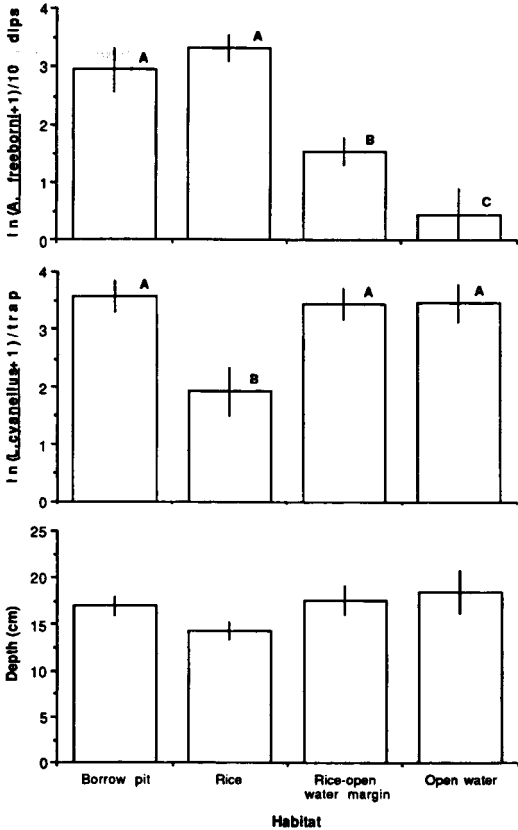


Fig. 1. Mean numbers (natural log-transformed) per station of *Anopheles freeborni* per 10 dipoles and *Lepomis cyanellus* per trap and mean depth of four habitat categories (see text for explanation of habitats) in subplot 1. Error bars are ± 1 SE. Different letters above histograms signify mean separation by Duncan's multiple range test. See text for effects of covariates.

differentiate whether mosquitoes have a different spatial distribution than the fishes in these rice fields or whether the negative associations may be due to predation by the fishes. Hence, the results from the correlations and analyses of variance give an estimation of a "realized" (*sensu* Hutchinson 1957) spatial distribution of the mosquito.

The analyses of covariance indicate that *An. freeborni* were more abundant in the borrow pit and rice stand habitats than in the open water in subplot 1 and were more abundant in the rice stand than in the borrow pit in subplot 2, even without the influence of fish—i.e., a closer estimation of a "potential" (*sensu* Hutchinson 1957) spatial distribution. These results support the hypothesis that poor spatial overlap between the fishes and *An. freeborni* contributed to the inability of the fishes to control this mosquito in the rice plot experiment of Blaustein (1989a).

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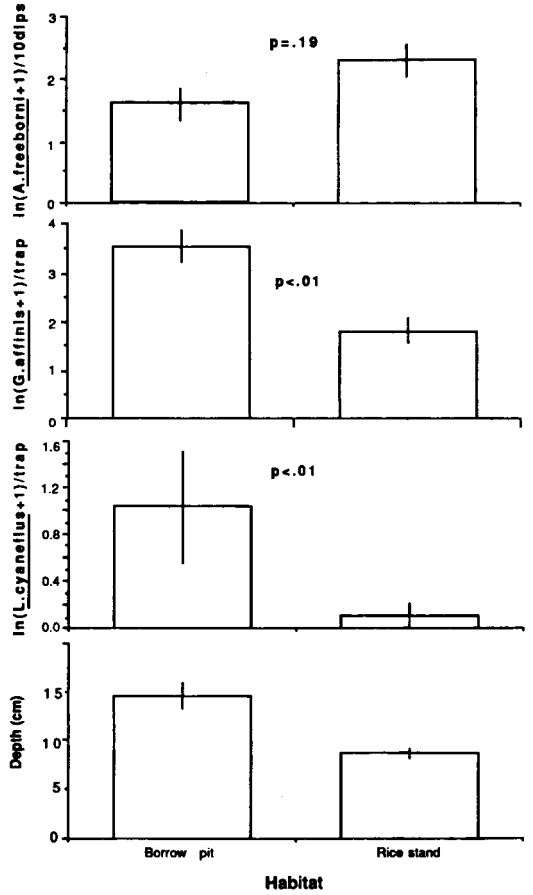


Fig. 2. Mean numbers (natural log-transformed) per station of *Anopheles* 10 dipoles, *Gambusia affinis* per trap and *Lepomis cyanellus* per trap and mean depth for two habitat categories in Subplot 2. Error bars are ± 1 SE. See text for description of habitats and effects of covariates.

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REFERENCES CITED

Ahmed, W., R. K. Washino and P. A. Gieke. 1970. Further biological and chemical studies on *Gambusia affinis* (Baird and Girard) in California. Proc. Calif. Mosq. Control Assoc. 38:95-97.
 Blaustein, L. 1989a. Larvorous fishes fail to control mosquitoes in experimental rice plots. Hydrobiologia (in press).
 Blaustein, L. 1989b. Effects of various factors on the efficiency of minnow traps to sample populations of

- mosquitofish (*Gambusia affinis*) and immature green sunfish (*Lepomis cyanellus*). *J. Am. Mosq. Control Assoc.* 5:29-35.
- Cech, J. J., Jr. and A. L. Linden. 1987. Comparative larvivoracious performances of mosquitofish, *Gambusia affinis*, and juvenile sacramento blackfish, *Orthodon microlepidotus*, in experimental paddies. *J. Am. Mosq. Control Assoc.* 3:35-41.
- Coykendall, R. L. 1981. Distribution and migratory movements of mosquitofish in a Sacramento Valley rice field. *Proc. Calif. Mosq. Vector Control Assoc.* 49:39-44.
- Davey, R. B., M. V. Meisch, D. L. Gray, J. M. Martin, K. E. Sneed and F. J. Williams. 1974. Various fish species as biological control agents for the dark rice field mosquito in Arkansas rice fields. *Environ. Entomol.* 3:823-826.
- Davey, R. B. and M. V. Meisch. 1977. Control of dark rice field mosquito larvae, *Psorophora columbiae* by mosquitofish, *Gambusia affinis* and green sunfish, *Lepomis cyanellus*, in Arkansas rice fields. *Mosq. News.* 37:258-262.
- Hess, A. D. and T. F. Hall. 1943. The intersection line as a factor in anopheline ecology. *J. Nat. Malaria Soc.* 2:93-98.
- Hoy, J. B., E. E. Kauffman and A. G. O'Berg. 1972. A large-scale field test of *Gambusia affinis* and chlorpyrifos for mosquito control. *Mosq. News* 32:161-171.
- Hoy, J. B., A. G. O'Berg and E. E. Kauffman. 1971. The mosquitofish as a biological control agent against *Culex tarsalis* and *Anopheles freeborni* in Sacramento Valley rice fields. *Mosq. News* 31:141-146.
- Hoy, J. B. and D. E. Reed. 1971. The efficacy of mosquitofish for control of *Culex tarsalis* in California rice fields. *Mosq. News* 31:567-572.
- Hutchinson, G. E. 1957. Concluding remarks. *Cold Spring Harbor Symp. Quant. Biol.* 22:415-427.
- Kramer, V. L., R. Garcia and A. E. Colwell. 1987. An evaluation of the mosquitofish, *Gambusia affinis*, and the inland silverside, *Menidia beryllina*, as mosquito control agents in California wild rice fields. *J. Am. Mosq. Control Assoc.* 3:626-632.
- Norland, R. L. and J. R. Bowman. 1976. Population studies of *Gambusia affinis* in rice fields: sampling design, fish movement and distribution. *Proc. Calif. Mosq. Vector Control Assoc.* 44:53-56.
- Reed, D. E. and T. J. Bryant. 1972. Interrelation between water depths and the distribution of *Gambusia affinis* and immature *Culex tarsalis* in Fresno County rice fields. *Proc. Calif. Mosq. Vector Control Assoc.* 40:122-123.
- Werner, E. E. 1984. The mechanisms of species interactions and community organization, pp. 360-382. *In:* D. R. Strong, D. Simberloff, L. G. Abele and A. B. Thistle (eds.), *Ecological communities: conceptual issues and the evidence*. Princeton Univ. Press, Princeton, NJ.
- Werner, E. E. and D. J. Hall. 1976. Niche shifts in sunfishes: experimental evidence and significance. *Science* 191:404-406.

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Blaustein, Leon. 1989. Spatial distribution of *Anopheles freeborni*, *Gambusia affinis* and *Leptomis cyanellus* in experimental rice plots. J. Am. Mosq. Control Assoc. 5:254-257.

Table 1 on page 255 should be replaced by the table below.

Table 1. Spearman rank correlations for 13 stations in subplot 1 and 25 stations in subplot 2.

	<i>G. affinis</i>	<i>L. cyanellus</i>	Depth
Subplot 1 (n = 13)			
<i>An. freeborni</i>	—	-0.48*	-0.38
<i>L. cyanellus</i>	—	—	0.16
Subplot 2 (n = 25)			
<i>An. freeborni</i>	-0.40**	-0.39**	-0.28
<i>L. cyanellus</i>	0.31	—	0.32
<i>G. affinis</i>	—	0.31	0.67***

* $P < 0.10$.

** $P < 0.05$

*** $P < 0.01$
