

OVIPOSITION RESPONSES OF GRAVID *CULEX QUINQUEFASCIATUS* AND *CULEX TARSALIS* TO BULRUSH (*SCHOENOPLECTUS ACUTUS*) INFUSIONS

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ABSTRACT. Laboratory bioassays demonstrated that fermented infusions of dried bulrushes (*Schoenoplectus acutus*) strongly attracted and stimulated oviposition by gravid female *Culex quinquefasciatus* and *Culex tarsalis*. The responses of the 2 species varied with the concentration and method of preparation of the infusions, with responses generally increasing with increasing concentration. No major differences were found in the responses of either species to infusions prepared with bulrushes alone, or with bulrushes plus lactalbumin hydrolysate and brewer's yeast. Infusions remained more attractive than distilled water controls to both species for up to 8 wk. Field tests corroborated the results from the laboratory bioassays. *Culex quinquefasciatus*, *Cx. tarsalis*, and *Cx. stigmatosoma* egg rafts were collected from water pan traps baited with bulrush infusions. A few *Culiseta incidens* eggs also were collected. In multiple-choice tests using gravid female or egg traps, *Cx. quinquefasciatus* preferred the most concentrated bulrush infusions, whereas *Cx. tarsalis* preferred intermediate concentrations. Female *Cx. stigmatosoma* and *Culiseta incidens* also were attracted. Overall, these results may provide new leads towards developing synthetic chemical baits to attract bloodfed mosquitoes.

KEY WORDS Oviposition attractant, oviposition stimulant, mosquito, bulrush, *Schoenoplectus acutus*, *Culex quinquefasciatus*, *Culex tarsalis*

INTRODUCTION

Several *Culex* mosquito species are known to transmit a number of disease agents to humans and livestock. For example, *Culex quinquefasciatus* Say is the principal urban vector of Bancroftian filariasis in the tropics (Carpenter et al. 1946), and may be of minor concern as a secondary vector of St. Louis encephalitis virus (SLE) in the USA (Reiter 1986). In contrast, *Culex tarsalis* Coquillett is the primary vector of SLE and western equine encephalomyelitis virus in California and other parts of the USA (Reisen and Reeves 1990, Reisen et al. 1992). Outbreaks of these viruses have been correlated with populations of *Culex* mosquitoes. Therefore, accurate monitoring of *Culex* abundance is critical in forecasting the risk of arboviral disease outbreaks. A number of sampling methods commonly are used for this purpose, including Centers for Disease Control (CDC) light traps, New Jersey light traps, gravid female traps, egg raft collection pans, and resting boxes (Vaidyanathan and Edman 1997). Sampling methods that preferentially attract previously bloodfed *Culex* females are particularly beneficial for surveying the prevalence of arbovirus infection in mosquito populations because this segment of the population is most likely to have contacted an infected host.

Many *Culex* share a preference for oviposition site waters containing organic detritus and a high level of microbial activity (Reiter 1986). This feature has been exploited by using fermented infusions of organic matter such as grass, hay, and manure as baits for gravid female and egg raft collection traps (Reisen and Meyer 1990). The sen-

sitivity of this sampling method is related directly to the attractiveness of the infusions to the particular target species. Therefore, gravid *Cx. quinquefasciatus* are known to be strongly attracted to fermented infusions of both plant material or animal excreta (reviewed in Bentley and Day 1989; Reisen and Meyer 1990). In contrast, *Cx. tarsalis* does not seem to be strongly attracted to these infusions (Reisen and Meyer 1990), limiting the effectiveness of these infusions as baits for this important vector species. A more effective bait for gravid *Cx. tarsalis* would be of considerable benefit to public agencies charged with mosquito and arbovirus surveillance and control.

Recent research has provided new leads for investigation. For example, Berkelhamer and Bradley (1989) found that *Cx. tarsalis* and *Cx. erythrorhax* Dyar were associated with floating mats of decaying cattails (*Typha* spp.) and bulrushes (*Schoenoplectus californicus* (Meyer) Soják). These mats provided both a refuge from predators and a source of nutrition for the mosquito larvae. Furthermore, Walton and Workman (1998) demonstrated that infusions of decaying bulrushes were attractive to *Culex* spp. The goal of the work described herein was to determine whether bulrush infusions have potential as baits for *Culex* mosquito sampling. Our specific objectives were to determine the effects of bulrush infusions as oviposition attractants and stimulants for *Cx. tarsalis* and *Cx. quinquefasciatus*, using both laboratory and field bioassays; to examine the effects of infusion concentration on mosquito oviposition behavior; and to examine the effects of different methods of infusion preparation on mosquito oviposition behavior.

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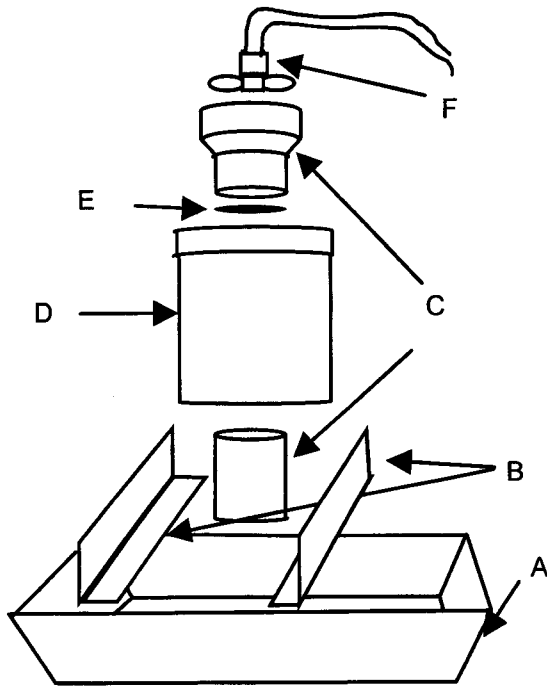


Fig. 1. Diagram of a gravid mosquito trap redesigned from Reiter (1983), using all metal or disposable parts. (A) Black plastic pan (20 × 15 × 7 in.) lined with black trash bag; (B) angle-iron support frame; (C) galvanized stovepipe; (D) cardboard ice-cream tub; (E) screen to retain mosquitoes; (F) fan powered by 4 D-cell batteries.

MATERIALS AND METHODS

Insects: The *Culex* mosquito colonies were maintained in the laboratory as previously described (Kramer and Mulla 1979, Millar et al. 1992, Isoe 1994). In particular, the colonies of the 2 species were maintained in facilities in separate buildings because *Cx. quinquefasciatus* rapidly displace *Cx. tarsalis* under the rearing conditions used in our laboratories. Freshly emerged females were allowed to mate and were bloodfed overnight (7 days after emergence) on White Leghorn chicks (University of California Animal Use Protocol A-M9412044-3). Gravid females were used in bioassays 5–10 days after bloodfeeding.

Preparation of bulrush infusions: The bulrush infusions used in this study were prepared in April–May, 1996. The standard infusion was prepared from 360 g of dried, chopped bulrushes (*Schoenoplectus acutus* (Muhl. ex Bigel) Löve and Löve), collected from a constructed wetlands close to Hemet, CA, steeped outdoors in the shade in 80 liters of irrigation canal water in a covered, 250-liter fiberglass tub for 7 days. Concentrated infusion was prepared from 450 g of dried bulrushes steeped in 20 liters of water. In the bulrush + lactalbumin hydrolysate + yeast infusion (bulrush + LHY; after Reiter 1983), 90 g of dried bulrushes, 5.4 g of lact-

albumin hydrolysate, and 5.4 g of yeast were fermented with 20 liters of water. The resulting infusions were filtered through a mesh screen to remove large debris, transferred to polyethylene bags, and stored frozen until needed.

Laboratory bioassays: For dual-choice bioassays assessing attraction, or combined attraction and oviposition, 30 × 30 × 45-cm wooden frame cages covered with muslin and with a muslin sleeve for access were used. Two glass oviposition cups (~120 ml) were placed in each of the back corners, with 1 containing the test material (100 ml), and the other containing distilled water (100 ml) as a control. The positions of treatment and control cups were alternated between replicates. Eight replicates of 20 gravid female mosquitoes per cage were used, with cages placed side by side for each bioassay, with a 12:10 h light : dark photoperiod, and 1-h simulated dawn and dusk periods. Assays were run overnight (~1700 to ~0800 h) in a well-ventilated room, at temperatures of 27 ± 2°C and a relative humidity of 30–40%.

The sticky-screen method (Isoe et al. 1995) was used to assess the attractiveness of test materials. Sticky screens were prepared about 3 h before the beginning of each bioassay by dipping galvanized hardware cloth disks (6.5 mm mesh, 8 cm diameter) in a solution of Tanglefoot® (Grand Rapids, MI) diluted with hexane. The screens were placed in a fume hood for 3 h for the hexane to evaporate. A sticky-screen disk then was placed over the mouth of each oviposition cup. Bioassays were run overnight as described above, and scored the next morning by counting the number of females stuck on each screen.

Egg-raft-counting bioassays were used to assess the combined effects of attraction and oviposition stimulation. These bioassays were conducted exactly as described above except that the mouths of the oviposition cups were not covered with sticky screens, so that mosquitoes had free access to the oviposition waters. Bioassays were evaluated the following morning by counting the numbers of egg rafts laid in treatment and control cups.

Multiple-choice bioassays were used to compare several treatments simultaneously, using 4 glass oviposition cups placed in each of the 4 bottom corners of 45 × 45 × 45-cm screen cages. The placement of cups was randomized between replicates. Each test was replicated 4 times using 50 gravid mosquitoes per replicate. Other conditions were as described above.

Field tests: Field tests were carried out at the Aquatic Insect Research Center, Agricultural Operations, University of California, Riverside, during August–October, 1996. Oval, black enamel turkey roasting pans (18 × 12.25 × 7.5 in. [45.7 × 31.1 × 19.1 cm]) were used as egg raft traps. Traps were placed along the treeline in a clearing with a number of small experimental ponds. Tests were set up at about 1700 h, and run consecutively for 4 nights.

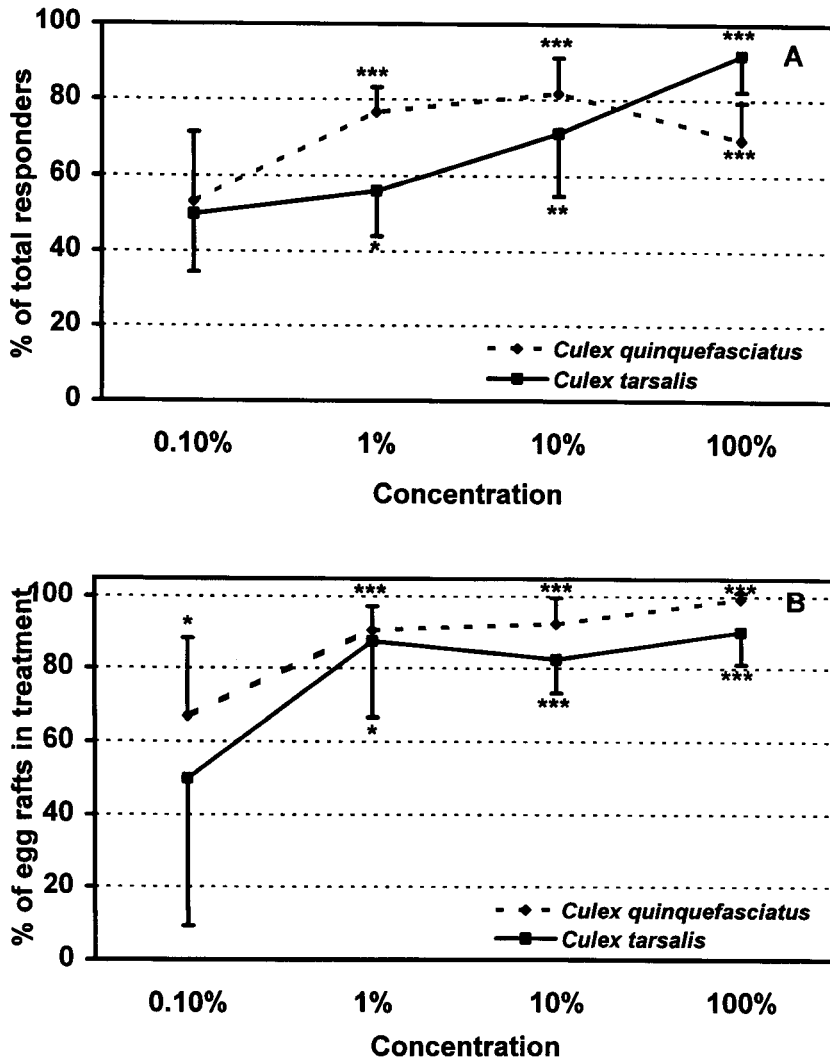


Fig. 2. Responses of gravid *Culex quinquefasciatus* and *Culex tarsalis* in (A) sticky-screen bioassays measuring attraction and (B) egg-raft-counting bioassays measuring oviposition, to dilutions of bulrush (*Schoenoplectus acutus*) infusion. Data are expressed as the number of mosquitoes responding to the treatment as a percent of the total number responding to treatments + controls. Each test was replicated 8 times, with 20 females/replicate. Differences between treatments and controls were determined with *t*-tests (* = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$).

Traps were divided into 3 groups of replicates, with each group consisting of a control (5 liters of irrigation water) and 1 to 3 infusion treatments. Infusion treatments were diluted as required with irrigation water. Pans within a group were placed ~3 m apart, and groups were separated by at least 20 m. Egg rafts laid in each pan were counted each morning, and each egg raft was reared individually until the 4th stage for identification to species using the characters described in Bohart and Washino (1978). Each experiment was repeated twice.

Field tests also were carried out at the same location during July 1997, with gravid female traps (Fig. 1) modified from Reiter (1983) so that all parts were metal or disposable, to prevent cross

contamination between trials. Briefly, black plastic pans (20 × 15 × 7 in. [50.8 × 38.1 × 17.8 cm]) lined with black trash bags were fitted with a metal frame to which the trap assembly was fitted. The trap assembly consisted of a piece of galvanized stovepipe (7.6 cm inner diameter; from a local hardware store) topped with a 1-gal (3.8-liter) cardboard ice-cream tub. The top of the tub was fitted with a 2nd piece of galvanized stovepipe, with a screened entrance. A small 6-V DC fan (John W. Hock Co., Gainesville, FL) powered by 4 D-cell batteries connected in series was fitted into the top piece of pipe to provide suction. Tests were set up at about 1700 h and run for 4 consecutive nights. The numbers of females trapped were counted each

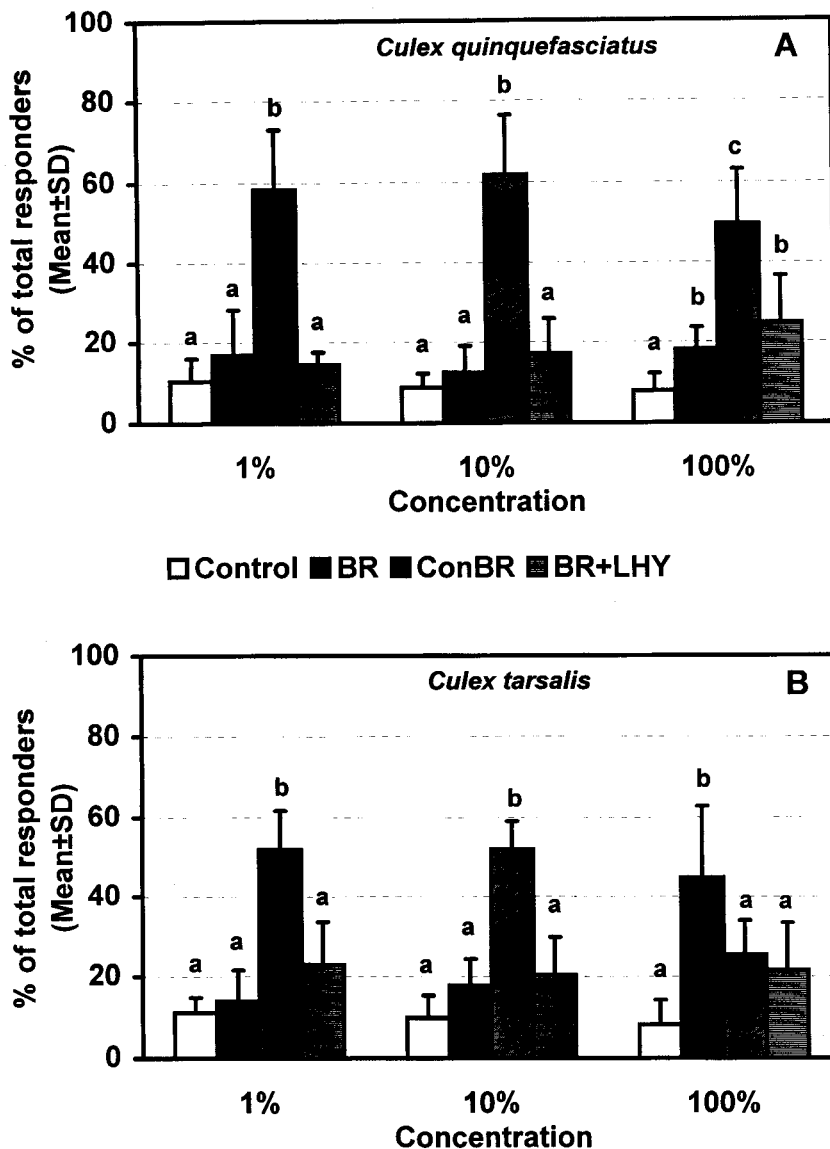


Fig. 3. Attraction of gravid *Culex quinquefasciatus* (A) and *Culex tarsalis* (B) to dilutions of infusions in multiple-choice sticky-screen bioassays. Treatments were standard bulrush infusion (BR), concentrated bulrush infusion prepared with 5 times as much bulrushes (ConBR), and bulrush infusion fermented with lactalbumin hydrolysate and yeast (BR + LHY). Each test was replicated 4 times, with 50 gravid mosquitoes/replicate. Treatments surmounted by the same letter are not significantly different (Student–Newman–Keuls test, $P > 0.05$).

morning. Traps were divided into 3 groups, with 1 control (16 liters of irrigation water) and 3 treatments per group. Traps within a group were placed 3 m apart, and groups were separated by at least 20 m. Gravid females captured were transferred to plastic bags and then freeze-killed for counting and species identification. Each experiment was repeated twice.

Data analysis: Data from dual-choice tests were analyzed by *t*-tests, and 1-way analyses of variance followed by Student–Newman–Keuls tests were

used to determine differences between treatments in multiple-choice tests.

RESULTS

Laboratory bioassays

The standard bulrush infusion strongly influenced oviposition by both mosquito species in laboratory bioassays. In sticky-screen bioassays measuring only attraction, gravid *Cx. tarsalis* and *Cx.*

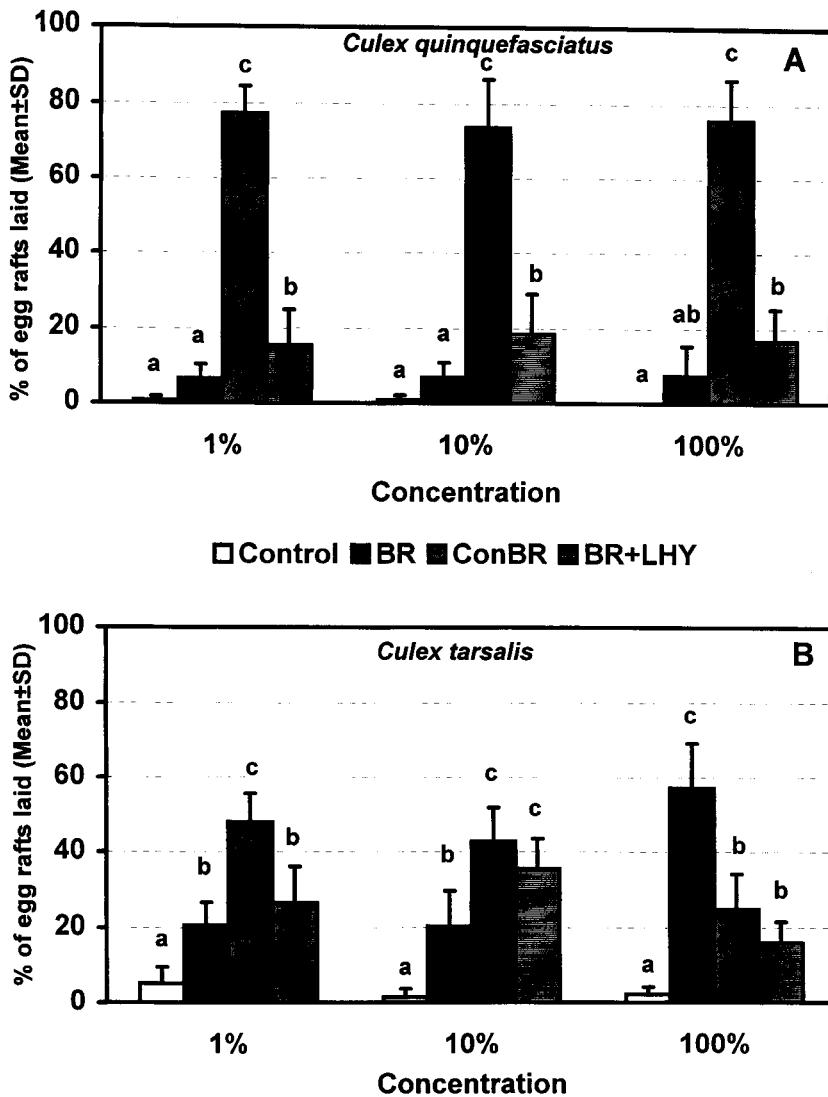


Fig. 4. Oviposition responses of gravid *Culex quinquefasciatus* and *Culex tarsalis* to dilutions of infusions in multiple-choice egg-raft-counting bioassays. Treatments were as in Fig. 3. Each test was replicated 4 times, with 50 gravid mosquitoes/replicate. Treatments labeled with the same letter are not significantly different (Student-Newman-Keuls test, $P > 0.05$).

quinquefasciatus were significantly more attracted to the undiluted infusion, and to the 10 and 1% dilutions than to water controls (Fig. 2A). The 0.1% dilution was not significantly more attractive than the distilled water controls to either species.

This pattern was repeated in oviposition bioassays, in which gravid females of both species laid significantly more egg rafts in the 100, 10, and 1% infusion solutions (Fig. 2B). *Culex quinquefasciatus* but not *Cx. tarsalis* females also laid significantly more egg rafts in the 0.1% infusion solution than in the control.

The effects of bulrush infusions prepared with different ingredients or different amounts of ingre-

dients were tested in multiple-choice, sticky-screen bioassays. At all 3 concentrations tested (100, 10, and 1% infusion solutions) the concentrated bulrush infusion, prepared with 5 times more bulrushes than the standard infusion, was significantly more attractive to gravid *Cx. quinquefasciatus* than the other 2 treatments, which, except at the undiluted concentration, were not different from the controls (Fig. 3A). The results were somewhat different with *Cx. tarsalis*; for the 1 and 10% dilutions, the concentrated bulrush infusion was significantly more attractive than the other treatments, which were not significantly different from the controls (Fig. 3B). However, for the undiluted infusions, the standard

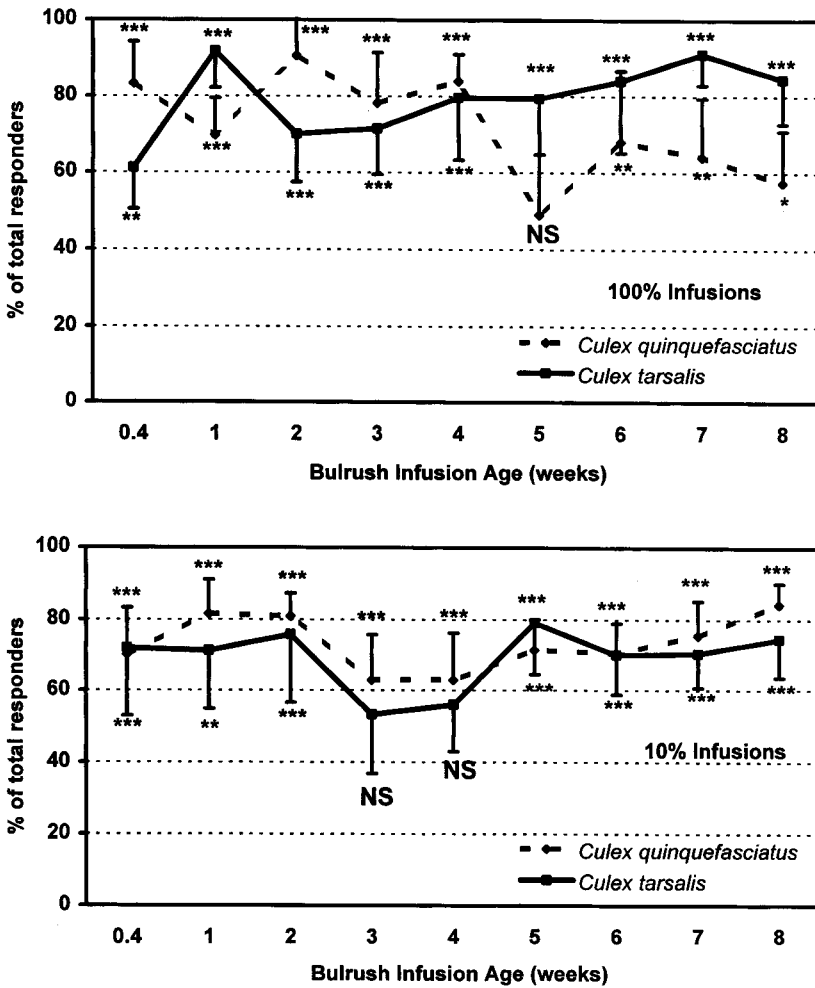


Fig. 5. Percentage of gravid *Culex quinquefasciatus* and *Culex tarsalis* attracted to undiluted (A) and diluted (10% dilution) (B) standard bulrush infusion aged for increasing periods of time, in dual-choice tests with water controls. Each test was replicated 8 times with 20 mosquitoes/replicate. Differences between treatments and controls were determined with *t*-tests (* = $P < 0.05$; ** = $P < 0.01$; *** = $P < 0.001$).

bulrush infusion was the most attractive treatment, with the other treatments and the control not being significantly different from each other.

The results of oviposition bioassays followed similar general trends (Fig. 4A). At all concentrations tested, *Cx. quinquefasciatus* laid the overwhelming majority of their egg rafts in the concentrated bulrush treatments. Differences also were found among the other treatments, with the bulrush + LHY infusion receiving significantly more egg rafts than the standard bulrush treatments, which were not different from the distilled water controls.

The results of oviposition bioassays with *Cx. tarsalis* (Fig. 4B) were not as straightforward. All treatments at all concentrations received more egg rafts than the water controls. In the 1% dilution series, the concentrated bulrush treatment was significantly better than the other 2 treatments, which were not different from each other. In the 10% di-

lution series, the concentrated and bulrush + LHY treatments were equivalent, and significantly better than the standard infusion. However, in parallel with the attraction bioassays described above, for the undiluted infusions, the standard bulrush infusion received significantly more egg rafts than the concentrated and bulrush + LHY treatments, again suggesting that more concentrated infusions are less attractive and stimulatory to *Cx. tarsalis*.

To assess changes in attractiveness with duration of fermentation, the attractiveness of standard bulrush infusion fermented for increasing periods of time was tested in dual-choice sticky-screen assays versus distilled water controls. Overall, both the undiluted infusion and a 10% dilution of infusion generally were significantly more attractive to gravid females of both species than the water controls, for infusion aged for 0.4–8 wk (Fig. 5).

Table 1. Numbers of egg rafts laid in oviposition traps in multiple-choice bioassays using 2 concentrations (10% dilution and undiluted) of different bulrush infusions. Results represent the mean number of egg rafts laid per trap over the 4 nights of each experiment, using 3 replicates of each treatment. Experiments were carried out from Sept. 3 to Oct. 10, 1996. Values followed by the same letters are not significantly different (Student–Newman–Keuls, $P > 0.05$).

Stimulus	Egg rafts laid	
	Trial 1	Trial 2
10% dilutions		
Standard bulrush infusion	2 ± 2.6 a	2.7 ± 3.1 a
Concentrated bulrush infusion	11.3 ± 2.9 b	15.7 ± 14.2 a
Bulrush + lactalbumin hydrolysate + yeast infusion	2.3 ± 2.3 a	4.7 ± 4.0 a
Irrigation water control	0.3 ± 0.6 a	0.7 ± 0.6 a
Undiluted infusions		
Standard bulrush infusion	10.7 ± 8.5 ab	7 ± 3.6 b
Concentrated bulrush infusion	22.7 ± 7.5 b	68.3 ± 5.5 c
Bulrush + lactalbumin hydrolysate + yeast infusion	13.0 ± 9.2 ab	27.0 ± 17.1 b
Irrigation water control	1.0 ± 1.0 a	0.3 ± 0.6 a

Field trials

A preliminary field trial with oviposition traps determined that the standard bulrush infusion strongly influenced mosquito oviposition, with infusion traps receiving 24.3 ± 8.4 egg rafts/trap-night (mean ± SD), compared to 1.0 ± 1.7 egg rafts/trap-night in water controls ($P < 0.01$, t -test). Multiple-choice tests, repeated twice, were used in follow-up tests of responses of mosquitoes to the different infusion "recipes." In the 1st set of experiments, using infusions diluted 10-fold, in the 1st trial the concentrated bulrush infusion received significantly more mosquito egg rafts than the other treatments, which were not significantly different from each other or from the controls (Table 1). Differences between treatments in the 2nd trial were obscured by the small number of replicates and the high variance among replicates. In the 2nd set of experiments, using undiluted infusions, the concentrated bulrush infusion received the most egg rafts, particularly in the 2nd trial, in which it was significantly better than the other 2 treatments and the control (Table 1).

The species compositions of the egg rafts laid in each of the trials are shown in Figs. 6 and 7, respectively. Most of the egg rafts collected in all 4 experiments were laid by *Cx. quinquefasciatus*, with lesser numbers of *Cx. stigmatosoma* Dyar, *Cx. tarsalis*, and *Culiseta incidens* (Thomson) egg rafts. The majority of the *Cx. quinquefasciatus* egg rafts were laid in the treatments prepared from the largest amounts of fermentable organic material.

The relative attractiveness of the infusions prepared from different mixtures of ingredients was tested in multiple-choice experiments with gravid female traps. Trap catches decreased with time during the 4-night course of the experiment (Table 2). All treatments were significantly more attractive

than water controls on the 1st 3 nights, but by the 4th night, only the concentrated infusion was significantly better than the control. On the 1st night, trap catches with the concentrated and bulrush + LHY treatments were not significantly different, but the activity of the bulrush + LHY infusion decreased on subsequent nights. Summed over the 4 nights, the concentrated treatment was best overall. *Culex quinquefasciatus* comprised the majority of the mosquitoes caught, but significant numbers of *Cx. stigmatosoma* and *Cx. tarsalis* were also attracted, along with a few *Cs. incidens* (Fig. 8).

DISCUSSION

Fermented infusions of organic matter are used widely as baits for gravid female and oviposition traps by public agencies charged with mosquito control. Infusions have been prepared from a diverse array of substrates, including alfalfa, grass, leaves, sod, various types of manure, tree bark, food pellets for laboratory animals, and fish oil (reviewed in Isoe 1994; Bentley and Day 1989). Although many of these infusions are attractive to *Cx. quinquefasciatus* (Reisen and Meyer 1990), their biological activity for *Cx. tarsalis* varies from weakly attractive to repellent (Reisen and Meyer 1990, Isoe 1994). Although Walton et al. (1990) found that *Cx. tarsalis* larval abundance was not significantly correlated to bulrush (*S. californicus*) stem density in freshwater marshes, recent studies have suggested that bulrush infusions were indeed quite attractive to gravid *Cx. tarsalis* (Walton and Workman 1998; Reisen et al., 1999). Surprisingly, this seems to be the 1st report of mosquito oviposition attractant activity associated with bulrush infusions, despite bulrushes being common in marshes and other *Culex* oviposition sites. Our laboratory

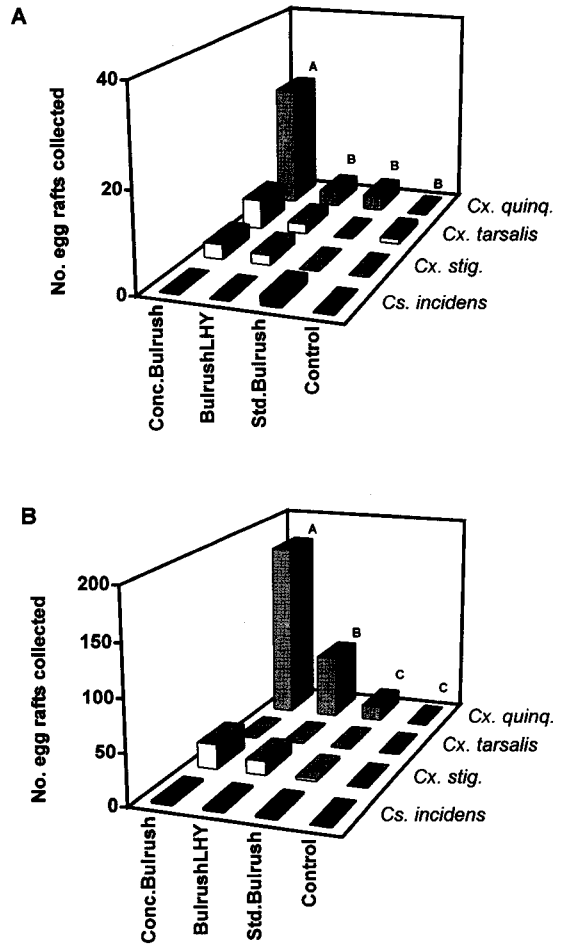
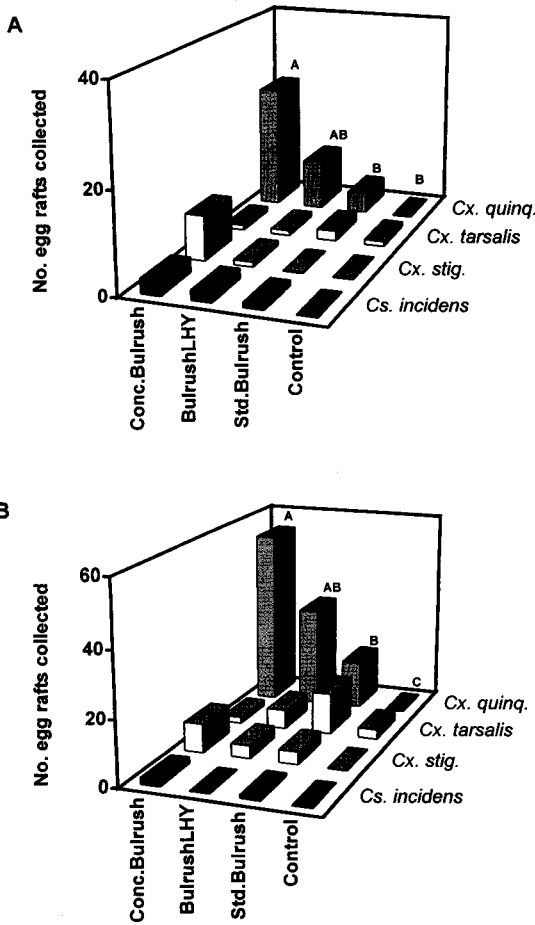


Fig. 6. Species composition of egg rafts laid in oviposition traps in multiple-choice bioassays using 2 concentrations (A, 10% dilution; B, undiluted) of different bulrush infusions. Results represent the total number of egg rafts laid in the 1st of 2 consecutive trials, each carried out for 4 nights. Treatment abbreviations are as in Fig. 3. Within a species, treatments labeled with different letters are significantly different (Student–Newman–Keuls test, $P < 0.05$).

Fig. 7. Species composition of egg rafts laid in oviposition traps in multiple-choice bioassays using 2 concentrations (A, 10% dilution; B, undiluted) of different bulrush infusions. Results represent the total number of egg rafts laid in the 2nd of 2 consecutive trials, each carried out for 4 nights. Treatment abbreviations are as in Fig. 3. Within a species, treatments labeled with different letters are significantly different (Student–Newman–Keuls test, $P < 0.05$).

Table 2. Numbers of mosquitoes trapped in a multiple-choice field bioassay using gravid traps baited with bulrush infusions prepared in different ways (described in the Materials and Methods). Each treatment was replicated 3 times, and traps were left in place for 4 consecutive nights. Values followed by the same letters are not significantly different (Student–Newman–Keuls, $P > 0.05$).

Stimulus infusion	Number of mosquitoes trapped (mean \pm SD)				
	1st night	2nd night	3rd night	4th night	Total
Standard bulrush	11.7 \pm 7.2 b	6.0 \pm 2.0 b	4.0 \pm 0.0 b	3.7 \pm 2.1 a	25.3 \pm 8.4 ab
Concentrated bulrush	29.7 \pm 8.1 c	32.0 \pm 15.5 b	18.3 \pm 6.5 c	16.7 \pm 17.7 b	96.7 \pm 28.2 c
Bulrush + lactalbumin hydrolysate + yeast	27.7 \pm 9.1 c	18.3 \pm 14.6 b	5.7 \pm 1.5 b	3.3 \pm 2.5 a	55.0 \pm 26.7 b
Water control	0.3 \pm 0.6 a	0.0 a	0.3 \pm 0.6 a	0.0 a	0.7 \pm 0.6 a

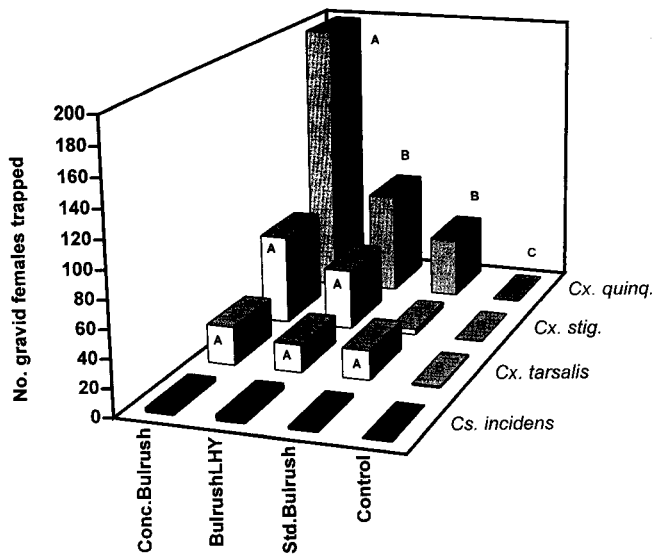


Fig. 8. Species composition of females caught in multiple-choice bioassays with gravid mosquito traps baited with different bulrush infusions. Results represent the total number of females caught in the 4-night experiment period. Treatment abbreviations are as in Fig 3.

and field results corroborate this report, and clearly indicate that bulrush infusions are attractive and stimulatory to gravid *Culex* mosquitoes. However, the field bioassays demonstrated subtle differences between the responses of *Cx. tarsalis* and *Cx. quinquefasciatus*, with the former species responding to a narrower range of concentrations of infusions than the latter. In particular, in choice bioassays testing the undiluted standard infusion versus the undiluted concentrated infusion, *Cx. tarsalis* demonstrated a preference for the standard infusion containing less organic matter than the concentrated infusion, whereas *Cx. quinquefasciatus* uniformly preferred infusions prepared from the highest content of organic matter. These results could be crucially important in the optimization of infusions as baits for trapping of gravid *Cx. tarsalis*. It also was interesting to note that the infusions prepared with bulrushes, lactalbumin hydrolysate, and yeast, in analogy to the widely used preparation developed by Reiter (1983), in general were not significantly better than infusions prepared from the same quantity of bulrushes alone, and at full strength, were significantly less attractive and stimulatory to *Cx. tarsalis*.

Distinct differences also were found in the behavioral responses measured by the 2 laboratory bioassay methods, as has been noted in previous studies of chemical cues mediating *Culex* mosquito oviposition (Isoe 1994, Isoe et al. 1995). The 1st method, using sticky screens placed over the mouths of cups containing test solutions, prevented mosquitoes from contacting the solutions, and thus measured only attraction to the odor of the test treatment. In these bioassays, the water controls often trapped 10–20% of the mosquitoes. In contrast,

differences between treatments and controls in the egg-raft-counting bioassays, which measure the end result of the whole series of consecutive behavioral steps culminating in oviposition (i.e., orientation to a potential oviposition site, arrestment and landing, stimulation to oviposit) tended to be more extreme, with very few egg rafts being laid in water controls in the presence of infusions. Overall, these results indicate that gravid females use volatile cues to orient to a potential oviposition site, but once at a site, the same or additional cues in the water also stimulate oviposition. In field tests, these differences disappeared, with the control gravid female traps (measuring attraction) and oviposition traps (measuring oviposition), baited with irrigation water alone, catching few or no mosquitoes, and receiving very few egg rafts. This difference may reflect the fact that laboratory bioassays were carried out in small screen cages, and the interior of the cages may have become permeated with infusion odors, presenting mosquitoes with a less clear-cut odor plume than in the field tests carried out in the open air, with the treatments separated by at least 3 m.

The field bioassays demonstrated that the bulrush infusions were attractive and stimulatory to several mosquito species, and that species exhibited preferences for different baits. The majority of mosquitoes caught were *Cx. quinquefasciatus*, with lower numbers of *Cx. stigmatosoma* and *Cx. tarsalis* also being trapped. However, no conclusions can be drawn with regard to the relative numbers of each species caught because the relative proportion of each species in the overall mosquito population could not be determined. Thus, the low numbers of *Cx. tarsalis* and other species attracted may

reflect low abundance of gravid females at the trapping site rather than poor attraction.

In summary, infusions prepared by fermenting dried bulrushes and water, with no other ingredients added, seem to have considerable potential as baits for use in mosquito surveillance programs. In particular, these infusions were attractive and stimulatory to *Cx. tarsalis*, an important disease vector in western North America, which is poorly attracted to other types of infusions. However, our data indicate that the infusion concentration must be regulated in order to produce an optimally attractive infusion because gravid *Cx. tarsalis* seem to prefer intermediate concentrations of infusions. In contrast, several other species attracted to bulrush infusions, including *Cx. quinquefasciatus* and *Cx. stigmatosoma*, seem to prefer more concentrated infusions. Chemical analysis of the bulrush infusions to identify the components influencing mosquito oviposition behavior is in progress.

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