Journal of the American Mosquito Control Association, 23(1):66–70, 2007
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EFFECTS OF VEGETATION CONTROL ON MOSQUITOES IN SEASONAL FRESHWATER WETLANDS

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ABSTRACT. Wetland restorations benefit wildlife, but wetland vegetation may promote mosquitoes. We studied the effects of removing joint grass (*Paspalum distichum* L.) on the abundance and distribution of immature mosquitoes in seasonal freshwater wetlands in California. In 4 >4-ha plots and one 29.8-ha field, joint grass was removed via herbicide and disking. Four untreated plots and one 4.1-ha field were controls. Fields were sampled every 7–14 days in early autumn by taking 75 dips along levees, plus a 25-dip circular transect 6 m from field edges. Untreated plots had 7 times more larvae and 20 times more pupae than did vegetation-removal plots. Similar densities of mosquitoes were found along edges and within fields.

KEY WORDS *Culex tarsalis*, disking, seasonal wetlands

INTRODUCTION

After decades of wetland losses to agriculture and development, public agencies and private landowners have been restoring or creating seasonal wetlands in many areas (Collins and Resh 1989). Wetlands are being created to support bird migration and overwintering, to restore native plant and animal communities, to provide natural flood control, and to process waste water. However, restored wetlands can provide habitat for mosquitoes that vector disease-causing microbes and that potentially create social problems (e.g., decreased property values, decreased tourism, problems in hiring or retaining outdoor workers, and nuisance issues).

Certain kinds of emergent vegetation are well known to favor mosquito production (Collins and Resh 1989, de Szalay and Resh 2000, review: Jiannino and Walton 2004), whereas expanses of open water are thought to discourage mosquito production (Collins and Resh 1989). Joint grass (*Paspalum distichum* L.; also called "knot grass") is an emergent grass that grows in dense mats that can provide cover for mosquitoes. In addition, some wildlife managers regard areas covered by this grass as poor habitat for waterfowl. This study was undertaken to test whether removing joint grass would reduce mosquito numbers.

The combination of disking and herbicide treatments is a favored method for joint grass control because this grass grows back quickly after mowing and grazing. Disking has long been known as a method for reducing floodwater mosquitoes that lay their eggs in soil (Cooney et al. 1981); however, its efficacy is less well established for mosquitoes that use standing water. While it is generally believed that vegetation control will diminish mosquito production, there are surprisingly few published studies demonstrating this, especially in freshwater systems. Batzer and Resh (1992) showed that mowing vegetation in 7-m strips caused local reductions in mosquitoes in a brackish marsh, but the larvae were merely concentrating in the remaining vegetation. De Szalay et al. (1995) found that disking and mowing decreased *Culex tarsalis* Coq. and *Culiseta inornata* Williston abundances in brackish marshes, at larger scales where behavioral concentration was less likely to be important (16 × 60 m). In contrast, in a study of a wastewater treatment wetland, reducing vegetation by 50% had no effect on mosquito production from the vegetated areas, whether the vegetation was left in 5-m strips or 10-m strips. (Jiannino and Walton 2004). Open marsh water management has a history of success. However, because this technique combines vegetation reduction, pond creation, and tidal flushing in salt marshes (Dale and Hulsman 1990, Wolfe 1996), it is unclear whether vegetation reduction alone is sufficient to decrease mosquitoes.

This study evaluated whether a combined herbicide and disking treatment of joint grass infestations would help reduce mosquito production in seasonal freshwater wetlands in the Yolo Bypass Wildlife Area, a wildlife refuge 5 miles west of Sacramento, CA. Such refuges are typically flooded in late summer or early autumn to provide wetlands for migrating waterfowl.

MATERIALS AND METHODS

We established a large-scale experiment in which a 46-ha field was divided into 8 plots

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>4 ha that either had joint grass removed or were left vegetated; treatments were established in summer 2005. The large scale of the study minimized the chance that mosquito movement would bias results. In addition, we included separate disked and undisked reference fields for comparison. The main experimental field had historically high populations of both larval mosquitoes and joint grass (*P. distichum*). Within this field, we established 4 herbicided and disked areas that alternated in a checkerboard pattern with 4 untreated areas (Fig. 1). Subplots ranged in size from 4.9 to 8.1 ha. The total areas disked and undisked were similar (24.1 ha disked versus 22 ha undisked). The other 2 fields served as references. One 4.1-ha field was left untreated (Field 9). Another 29.8-ha field had been recently 75% disked and graded as part of a restoration project by the California Waterfowl Association; 30% of this field was again disked and sprayed with herbicide before the study (Field 10).

Herbicide was applied July 27 and 28 to treated plots and to the treated reference field. Joint grass was identified and treated with Roundup Pro (Monsanto Company, St. Louis, MO) at 1.14 liters per hectare, by a 757-liter (200 gallon) boom sprayer pulled by a large tractor in the treated plots and by an ATV with a 190-liter boom sprayer in the treated reference field. The majority of sites 1, 3, 5, and 7, minus islands and cattail stands, were treated with Roundup. Approximately 2 weeks after herbicide applications were made and when significant browning was observed, stubble disking was initiated with a Challenger 65E Caterpillar tractor and 4.26-m stubble disk. One to 4 passes with the disk were required depending on the density of the joint grass or other vegetation. The majority of disking was focused on joint grass, while effort was made to avoid emergent rushes, swamp timothy, and water grasses. Sites 2, 4, 6, and 8 and Field 9 were untreated controls.

Fig. 1. Aerial photograph of wetland study plots showing sizes and locations of sites used to collect mosquito immature stages from an experiment on how vegetation removal influences mosquitoes. Field 9 was a control field; another field where vegetation was removed is not shown. Dots show approximate centers of edge transects and the beginnings of circular transects within sites.

After operations were complete, 2 researchers independently walked across each plot or field and performed visual estimates of percentage cover of joint grass and all vegetation to the nearest 10%. Estimates were always within 10% of each other.

Fields were flooded in late September and mosquito larvae were sampled from October 11 to November 17. Samples were taken along the edge and within each study site. Seventy-five dips were taken from the edge at depths of 3–6 cm, spaced ~1 m apart and we left a buffer of >20 m between plots. Twenty-five additional dips were taken within the field, by walking 6 m from the edge and taking a dip every meter in a circular pattern. Depths in these locations were 0.6–0.75 m. Each sample was taken by a standard 0.47-liter dipper and poured into a strainer for collection (mesh size 0.2 mm). A cumulative sample was taken from each edge or within-field transect. Samples were labeled and kept in alcohol until identified to taxon and growth stage. Larvae were identified to species or genus where possible. Pupae and very damaged specimens were identified to family. Edge and within-field samples were tabulated separately, but were combined for all analyses except the spatial analyses. Samples were taken every 1–2 weeks, weather permitting. We recorded pH and water temperature at each location. Temperature ranged from 25.5–10°C, decreasing as the experiment progressed, and pH was 7 in all areas.

All data were analyzed using Systat 11 (Systat Software Inc., Chicago, IL). To meet the assumptions of analysis of variance, abundance data were log transformed and percentage data were arcsin–square root transformed. Full-sized fields were treated as references rather than replicates because of differences in size.

RESULTS

The combination of herbicide application and disking was successful in eliminating joint grass in the treated experimental plots and field, and this also reduced cover of other vegetation to <10% in all sites. In contrast, the control sites had nearly 100% cover. The cover of joint grass in control plots was 20%, 40%, 40%, and 70%, and was 30% in the control field.

There were fewer immature mosquitoes (larvae and pupae of all taxa combined) in plots where joint grass was removed, and this difference increased significantly over time (Fig. 2; repeated-measures analysis of variance [ANOVA], treatment effect, df 1, 6, $F = 27.389$, $P = 0.002$; time × treatment interaction: df 4, 24, $F = 3.567$, $P = 0.021$). Averaging over the experiment, there

were 7 times more immature mosquitoes in control plots than in treated plots. Taxa collected are presented in Fig. 3 and included *Cx. tarsalis, Cs. inornata, Cx. pipiens,* and *Cs. incidens.*

Samples from the larger reference fields also showed that the vegetation-removal field had dramatically fewer mosquitoes than the control field (Fig. 4). These fields were not analyzed statistically due to lack of replication. The data from these fields are nevertheless very important because it shows that the effect of vegetation removal was consistent at the scale of whole fields.

Vegetation-removal plots produced 20 times fewer pupae than vegetated plots (Fig. 5; treatment effect df 1, 6, $F = 15.679$, $P = 0.007$). There was no detectable temporal trend. We did not perform this analysis for the reference fields because of the much smaller number of pupae found there.

Mosquito numbers were not detectably correlated with the estimated percentage of joint grass in control plots (linear regression, 2-tailed test, df 1, 2, $P = 0.48$). However, this was a very low power test since there were only 4 replicates. Vegetation-removal plots were not included in this analysis because in these plots, the absence of joint grass was conflated with the general lack of vegetation. Therefore, it would not have shown if the effect on mosquitoes was due to the exact type of vegetation.

When all taxa were pooled, there were no significant differences in numbers of immature mosquitoes sampled from the edges of plots versus those taken 6 or more meters from the edge, either across or within dates (repeated-measures ANOVA on number per dip, area effect df 1, 13, $F = 0.014$, $P = 0.907$; time by area effect df 4, 10, $F = 0.659$, $P = 0.635$). The mean number of immatures per dip averaged across dates was 0.50 ± 0.21 SE for within-field dips and 0.48 ± 0.19 SE for edge dips.

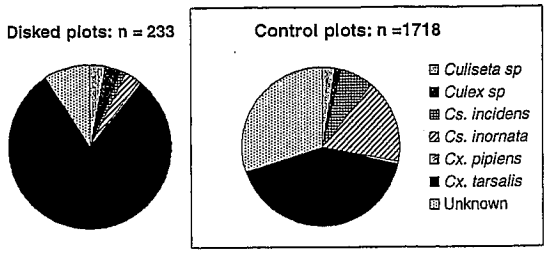

Fig. 3. Proportions of mosquito species collected in plots that either had joint grass removed (disked) or were untreated (control). The total number of immature mosquitoes collected is given as *n*. "Unknowns" were either pupae (73%) or were too small or damaged to identify.

A separate, exploratory analysis of the most abundant species, *Cx. tarsalis,* also showed no spatial effects. However, the second most abundant species, *Cs. inornata,* was approximately 4 times more abundant at the edges of control plots than within the vegetation (ANOVA on log-transformed abundances in edge versus center dips, adjusted for dip number, control plots only, df 1, 6, $F = 8.615$, $P = 0.026$). Note that vegetation-removal plots were not used in this analysis because of low numbers in both center and edge dips. Numbers of other species were too low for this analysis.

DISCUSSION

Control of vegetation through herbicide application and disking was very effective for reducing mosquito larvae in these freshwater wetlands. Vegetation-removal plots had 85% fewer immature mosquitoes and 95% fewer pupae than control plots. Batzer and Resh (1992) showed

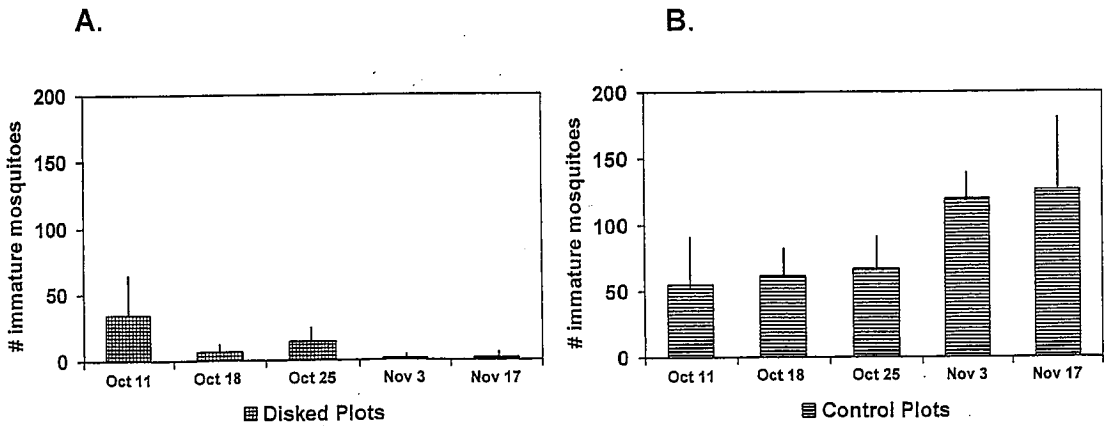

Fig. 2. Immature mosquitoes collected in plots that either had joint grass removed (disked, panel A) or were untreated (control, panel B). Bars show the means of the total numbers of mosquitoes collected in 100 dips in each of 4 plots on each date, and the standard errors of the means.

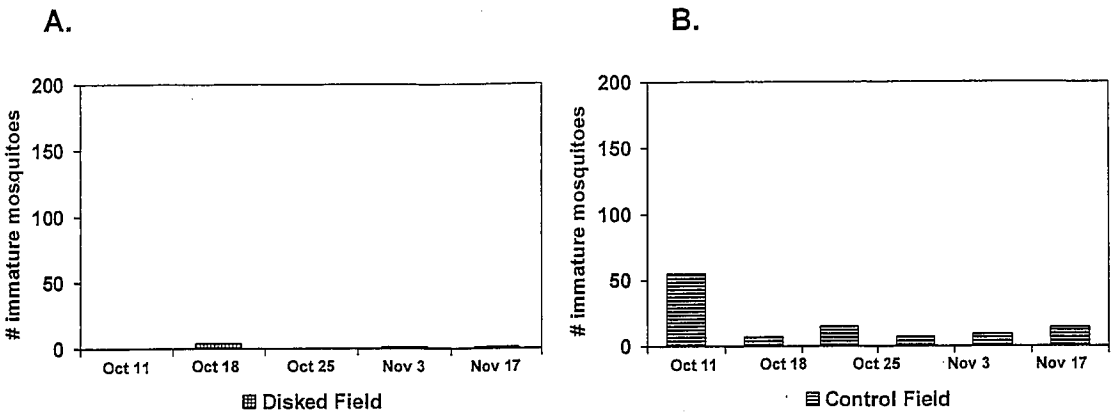

Fig. 4. Abundances of immature mosquitoes in an herbicided and disked reference field and a control field. Bars represent the total numbers of mosquitoes collected in 100 dips per field, per date.

a similar effect in a brackish marsh, where 7-m-wide mown strips of vegetation contained about 92% fewer mosquitoes than unmown strips. However, in that study mosquito numbers were elevated in the vegetated strips compared to fully vegetated plots, suggesting that the strips were concentrating the mosquitoes. The mechanism was unknown, and could have been wind, active movement by larvae, or a preference by ovipositing females for edge habitat. Walton and Mulla (1990) also found more *Cx. tarsalis* in 2-m^2 vegetated plots than in unvegetated plots. In that study plots were separated by window screening with 7 openings per centimeter, which may have restricted movement of at least larger stages. For the study presented here we cannot entirely rule out that mosquitoes were concentrated in the vegetated plots, but the multihectare scale of our study makes concentration through active movement unlikely because mosquito larvae are not strong swimmers. In addition, mosquito populations were similarly low in the reference field with vegetation removed in comparison to the untreated field, which suggests that vegetation removal reduces mosquitoes on a field-wide basis.

Comparable numbers of immature mosquitoes were collected along plot edges and within fields, therefore edge dips are sufficient to indicate overall mosquito population levels in this system. This is of great practical value for 2 reasons: it is easier to collect dips along field edges, and this method may be less disruptive to wildlife than walking out into fields. Of species that were abundant enough to be analyzed separately, *Cx. tarsalis* showed no spatial trends and can therefore be adequately sampled from field edges. However, *Cs. inornata* immatures were more abundant in edge dips in control plots, indicating that edge dips could inflate abundance estimates for this species. Other species were less abundant, and we did not have adequate power to detect their distributions. The distribution of mosquitoes in wetlands vary among species; for example Andis and Meek (1984) found that 93% of *Psorophora columbiae* Dyar and Knab immatures were found near the levees of unharvested rice

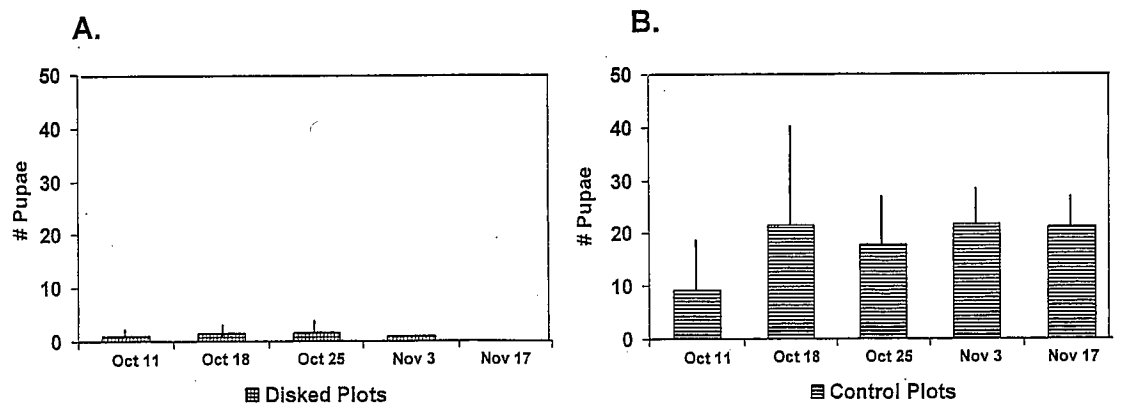

Fig. 5. Mosquito pupae collected in plots that either had joint grass removed (disked, panel A) or were untreated (control, Panel B). Bars show the means of the total numbers of mosquitoes collected in 100 dips in each of 4 plots on each date, and the standard errors of the means.

fields, whereas *Anopheles crucians* Wied. were evenly distributed across fields.

Most of the mosquitoes collected were *Cx. tarsalis,* a vector of several viruses that cause encephalitis. The second most common were *Culiseta* spp., which bite humans but are not thought to be major disease vectors. Thus, vegetation control shows good promise of reducing disease and pest problems. However, Collins and Resh (1989) caution that disking should not be performed when the ground is wet, to avoid causing depressions that might lead to poor drainage and more mosquito problems.

We were unable to establish that joint grass was correlated with mosquito production, possibly as a result of the low power of the analysis. Some studies have shown that mosquitoes may associate with particular vegetation types (Collins and Resh 1989). Pope et al. (2005) found more *Anopheles* mosquitoes in *Typha* L. (cattail) wetlands in Belize, but it was unclear whether the greater numbers were caused by the *Typha* or by agricultural enrichment of the wetlands that was associated with *Typha.* Jiannino and Walton (2004) found significantly more mosquito larvae in bulrush (*Schoenoplectus californicus* CA Meyer) than cattail (*Typha* spp.) in river water treatment wetlands. In their study, water quality was similar between vegetation types; thus, the association likely reflects effects of the vegetation itself on mosquito oviposition or survival. Further work is needed to establish the role of joint grass per se as mosquito habitat.

Although mosquito larvae and pupae were reduced in vegetation-removal areas, significant numbers of mosquitoes matured in adjacent plots. This suggests that effects of vegetation reduction were local. In some situations, local vegetation reductions can lead to greater numbers of invertebrate prey for waterfowl, while also reducing mosquitoes (review: de Szalay and Resh 2000). However, the nontarget effects of large-scale disking are poorly understood. Further research is needed to assess whether the scale of vegetation removal performed for this project affected either beneficial invertebrates, or other functions of the wetland ecosystem. Vegetation drives many ecosystem functions such as productivity, decomposition, and nutrient cycling.

ACKNOWLEDGMENTS

We thank the staffs of Sac-Yolo Mosquito and Vector Control District (MVCD) and California Department of Game and Fish (CDFG) for their hard work, and David Brown and Greg Lanzaro for facilitating the study. We thank 2 anonymous reviewers for helpful comments.

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