

## Notes

# Fire Ants in Houston Toad Habitat: Annual Activity and Responses to Canopy Cover and Fire

Donald J. Brown,\* Bei DeVold, Weston H. Nowlin, Michael R.J. Forstner

*D.J. Brown, B. DeVold, M.R.J. Forstner*

Department of Biology, Texas State University, 601 University Drive, San Marcos, Texas 78666

*W.H. Nowlin*

Department of Biology, Texas State University, Aquatic Station, San Marcos, Texas 78666

## Abstract

The red imported fire ant *Solenopsis invicta* (RIFA) is an invasive species found throughout the southern and southeastern United States. Since its introduction, RIFA has been shown to negatively affect a wide range of native vertebrate and invertebrate species. The purposes of this study were to delineate the annual RIFA activity pattern, investigate the association between overstory canopy cover and RIFA captures, and evaluate the effects of low-intensity prescribed fire around pond edges on RIFA in the Lost Pines ecoregion of Texas, a region that provides habitat for most of the remaining endangered Houston toads *Bufo houstonensis*. We found that annual RIFA activity followed a quadratic curve, with above-average activity between May and October. We found an inverse relationship between mean percentage of canopy cover near pond edges and mean number of RIFA captured. We found that low-intensity prescribed burning had no significant influence on RIFA captures during our study period. However, strong spatial and temporal capture variability was apparent, and thus a strong impact would have been necessary to detect an effect. Although this study provides evidence that should decrease concerns that wildlife managers, conservation biologists, and landowners in the Lost Pines ecoregion may have about exacerbating RIFA abundance when using fire as an ecosystem management tool, we recommend additional work be conducted using a greater sample size, greater sampling effort, and longer study duration. We found that RIFA activity was highest during the time frame in which juvenile Houston toads emerge from ponds. Thus, RIFA control may be a useful Houston toad recovery tool where breeding ponds are not within dense canopy habitats.

Keywords: Houston toad; Lost Pines; prescribed fire; red imported fire ant; Texas

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\* Corresponding author: db1300@txstate.edu

## Introduction

The red imported fire ant *Solenopsis invicta* (RIFA) is an invasive species in the United States, predominantly in the south and southeast. Since its introduction into Alabama in the 1930s, the distribution of RIFA has expanded east to North Carolina and west to central Texas (Callcott and Collins 1996), and the species has been introduced into California (Ward 2005). When introduced, RIFA has been shown to negatively affect a

wide range of native vertebrate and invertebrate species (Porter and Savignano 1990; Allen et al. 1994, 2004; Stuble et al. 2009; Diffie et al. 2010; Epperson and Allen 2010). Vertebrates are particularly vulnerable to RIFA predation during early life stages (Landers et al. 1980; Freed and Neitman 1988; Pedersen et al. 1996). Abundance and diversity of native arthropods also can be negatively affected by RIFA competition and predation (Morrison 2002; Plowes et al. 2007; Epperson and Allen 2010).



In the United States, RIFA prefers environments characterized by disturbance and can use roads and powerline cuts as expansion corridors (Tschinkel 1988; Stiles and Jones 1998; Todd et al. 2008). In primarily forested habitats, RIFA tends to be more abundant in forest gaps (Colby and Prowell 2006). Red imported fire ants are adapted to periodically flooded habitats (Allen et al. 1974) and have been found to prefer edges of ponds and other water bodies (Lyle and Fortune 1948; Tschinkel 1988; Stuble et al. 2009; Vogt et al. 2009). Because RIFA can be more flood tolerant than some native ant species, it may have a competitive advantage over these taxa in flood-prone environments (Vogt et al. 2009). The potential for high RIFA density around ponds is of concern in amphibian management, given the potential vulnerability of juvenile amphibians to RIFA predation when they enter the terrestrial landscape (Freed and Neitman 1988). Red imported fire ants prefer canopy gaps; thus, ponds with less canopy cover or near the edges of forested fragments may have higher RIFA abundances and may subsequently have higher RIFA predation pressure on juvenile amphibians.

Red imported fire ants arrived in Bastrop County, central Texas, between 1973 and 1977, and they are now well established in the Lost Pines ecoregion (Cokendolpher and Phillips 1989; Taber and Fleenor 2003). This ecoregion is a 34,400-ha remnant of a pine-dominated forest that occurred in east and east central Texas approximately 10,000–14,000 y ago (Bryant 1977). The Lost Pines houses the majority of the remaining breeding aggregations of the endangered Houston toad *Bufo (Anaxyrus) houstonensis* (Gottschalk 1970), a species that has declined rapidly in the last half-century due primarily to habitat loss and degradation (U.S. Fish and Wildlife Service 1984; Brown and Mesrobian 2005). Red imported fire ants are known to prey upon Houston toads after their emergence from ponds as terrestrial juveniles (Freed and Neitman 1988).

We conducted this investigation as part of a larger study on the use of prescribed fire for Houston toad habitat recovery and conservation initiatives. The majority of the Lost Pines has been fire suppressed for the past century, resulting in heavy fuel loads and dense thickets of fire-intolerant shrubs, primarily yaupon holly *Ilex vomitoria*. Currently, there is limited published information on RIFA response to prescribed burning, and RIFA population responses to fire are unclear. Forbes (1999) documented short-term (i.e., 5-mo postburn) negative effects on RIFA because of prescribed burning in a Texas coastal prairie, presumably due to reduced soil moisture and food availability. Conversely, Norton (2003) found no effect of burning on RIFA abundance in habitats similar to those of Forbes (1999). In contrast, Hanula and Wade (2003) found that RIFA abundances increased with burn frequency in a Florida longleaf pine *Pinus palustris* forest. Thus, the results of the aforementioned studies present an unclear picture of whether the use of prescribed fire as a recovery tool in pine-dominated ecosystems may increase invasive potential for RIFA or whether fire may be an effective method to reduce RIFA in invaded pine forests.

The purposes of this study were to delineate the annual RIFA activity pattern in the Lost Pines ecoregion of Texas and to investigate RIFA responses to overstory canopy cover and low-intensity prescribed fire. We were interested in temporal activity to better understand RIFA activity patterns in relation to Houston toad activity patterns and to delineate optimal time intervals for RIFA control measures. Percentage of canopy cover is an easily quantifiable variable that could be used to determine which Houston toad ponds probably contain large RIFA populations, and thus where emergent Houston toads are most vulnerable to RIFA predation. Prescribed fire is a common habitat management tool used across the southern and southeastern United States, and thus it is important to know the impacts of fire on this invasive species.

## Study Site

We conducted this study on the 1,900-ha Griffith League Ranch (GLR) in Bastrop County, Texas. The GLR is primarily forested, with vegetation typical of the Lost Pines ecoregion. The overstory is dominated by loblolly pine *Pinus taeda*, post oak *Quercus stellata*, blackjack oak *Quercus marilandica*, and eastern red cedar *Juniperus virginiana*, and the understory is dominated by yaupon holly *Ilex vomitoria*, American beautyberry *Callicarpa americana*, and farkleberry *Vaccinium arboreum*.

The GLR contains 17 ponds, 10 of which were used for this study. Three of the ponds were ephemeral (i.e., typically dry several times per year), six were semipermanent (i.e., typically dry several times per decade), and one was permanent (i.e., has not dried in at least 11 years). Four of the pond edges were primarily covered by pine and oak leaf litter (>80% ground cover), with the remaining ponds having edges dominated by grasses and forbs (>80% ground cover). We chose these ponds because they spanned the distribution of size, hydroperiod, and canopy cover variability on the GLR, they were known Houston toad breeding ponds, and, when applicable, they were located in units projected to be burned. Houston toad calling activity occurred at all ponds used in this study within the previous 2 y (Duarte et al. 2011).

## Methods

We conducted this study over a 1-y period between 26 September 2009 and 10 October 2010, sampling the area around each of the 10 ponds 27 times in total. Days between sampling ranged from 0 (i.e., consecutive sampling days) to 73, with a mean of 15 d between samples. We used a standard bait cup method for sampling ants (Porter and Tschinkel 1987; Mueller et al. 1999) that consisted of half a Vienna sausage placed in a 9-cm-diameter round plastic dish. We placed between 3 and 15 dishes around the perimeter of each pond, depending on pond size (i.e., perimeter length). During each sampling event, we placed dishes 1–2 m from the pond edge at 3–4-m intervals, and then we allowed them to attract ants for 30 min. After 30 min, we collected ants from each dish and euthanized them by freezing. We

removed native ants from samples, and RIFAs were dried at  $\sim 50^{\circ}\text{C}$  for at least 48 h. We then counted RIFAs collected in each bait cup at each pond on each sampling date. Alternately, we weighed the cumulative number of ants in each bait cup at each pond on each sampling date, and sample weight was estimated to the nearest  $1 \times 10^{-4}$  g. We estimated RIFA abundances in these samples using an empirically derived relationship between a known number of RIFAs and the weight of the sample. This relationship was derived from samples collected from around study ponds on three randomly selected sampling dates ( $n = 109$  samples,  $y = 2140.8 \times \text{weight [g]}$ , coefficient of determination [ $r^2 = 0.92$ ; Sokal and Rohlf 1995]). We used the estimated RIFA abundances as count data in statistical analyses.

Four of the ponds were subjected to prescribed burning, with all ponds sampled before and after burning. However, because ponds were spread over multiple burn units, areas around individual ponds were burned at different times of the year. The area around one pond was burned on 13 November 2009 (pond 10), one pond on 10 January 2010 (pond 13), and the remaining two ponds were burned on 21 August 2010 (ponds 14 and 15). This burn scenario created the ability to examine the effect of burning in winter (November and January) vs. in summer (August). All fires were low-intensity, fuel reduction burns; burns removed the upper portion (i.e., 1–2 cm) of the leaf litter layer and charred but typically did not consume live woody vegetation.

To assess whether RIFA counts were higher in open canopy areas, between 20 July 2008 and 16 April 2010, we estimated percentage of canopy cover around each pond between 4 and 24 times, depending on hydro-period status at the time of sampling (i.e., estimates were not taken when ponds did not contain water). For each sample, we estimated percentage of canopy cover using a spherical densiometer (Forestry Suppliers Inc., Jackson, MS) at two to six randomly selected points at the pond edge, with higher numbers of estimation points corresponding to larger ponds. We then averaged the estimates at each pond per sampling date. For this study, we included measurements taken during the leaf-off period (December–March) because ponds with consistently high canopy cover at our study site were dominated by loblolly pine, which retains needles throughout the year.

### Statistical analyses

We used a generalized additive model (Hastie and Tibshirani 1990) to summarize the annual activity pattern based on our sampling results. We used the captures-per-unit-effort (CPUE) at each pond on each day sampled (i.e., 27 samples with 10 observations per sample), included day of year (DOY) as a predictor, and we fit a smoothing curve to the data (cubic regression spline), with the optimal amount of smoothing determined using a cross-validation algorithm (Zuur et al. 2009). Thus, the smoothing curve represented the model-fitted relationship between RIFA CPUE and DOY. Generalized additive models assume normality and homoscedasticity, and we investigated these assumptions using graphical

diagnostics plots (Zuur et al. 2009). Because data seemed to violate the assumption of homoscedasticity, we transformed CPUE using the arcsinh (i.e., inverse hyperbolic sine) transformation (Fowler et al. 1998), which was effective. We performed this analysis using the program R (R Version 2.10.1; [www.r-project.org](http://www.r-project.org)), with the *mgcv* package (Wood 2004).

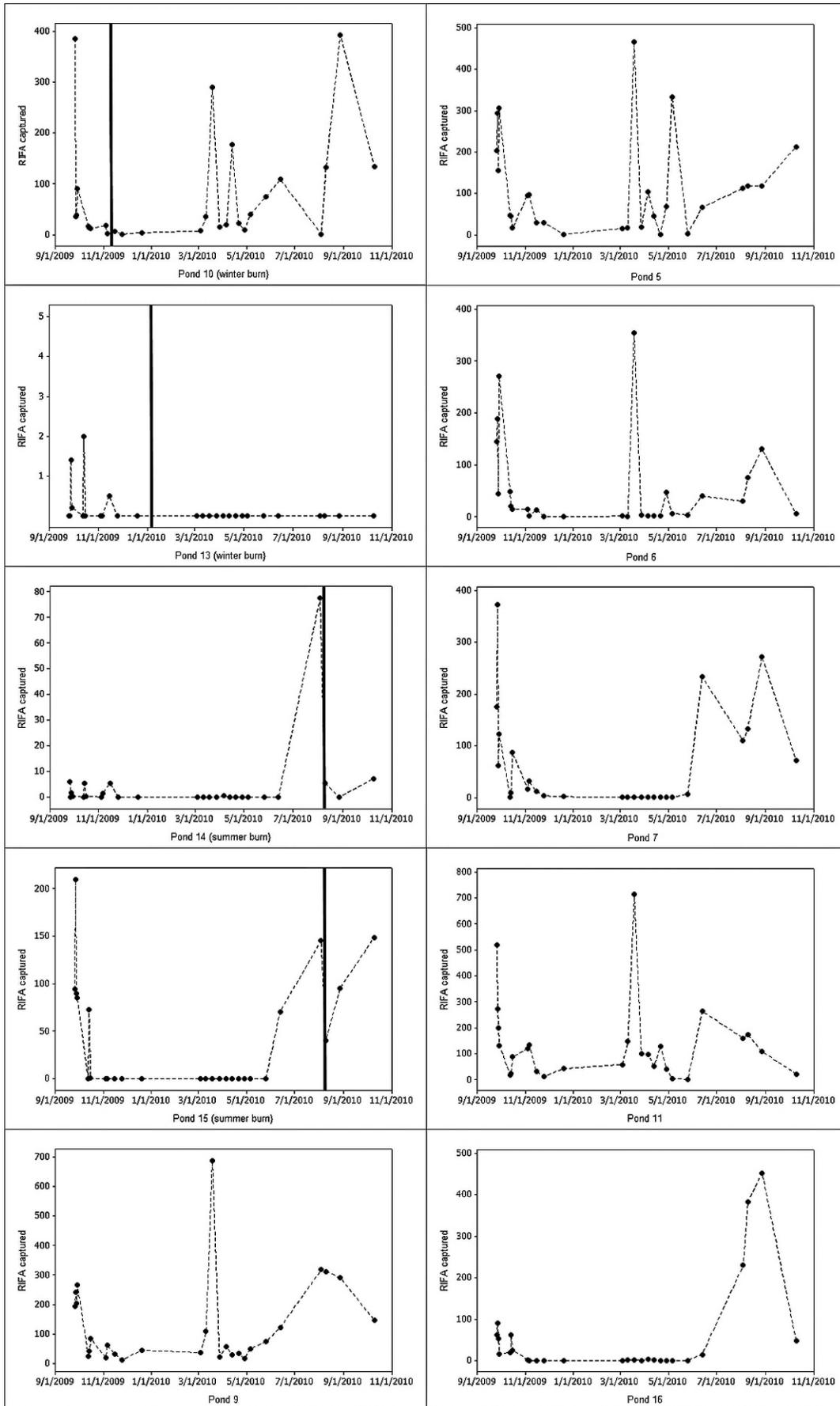
To examine the influence of canopy cover on RIFA CPUE, we used the RIFA capture and canopy cover data from all of the study ponds ( $n = 10$ ), and we calculated the mean percentage of canopy cover of each pond over the study period. We regressed mean RIFA CPUE on mean percentage of canopy cover for all ponds using ordinary least squares linear regression, and we gauged model fit using  $r^2$  (Sokal and Rohlf 1995). We performed this analysis using program R (R Version 2.10.1; [www.r-project.org](http://www.r-project.org)).

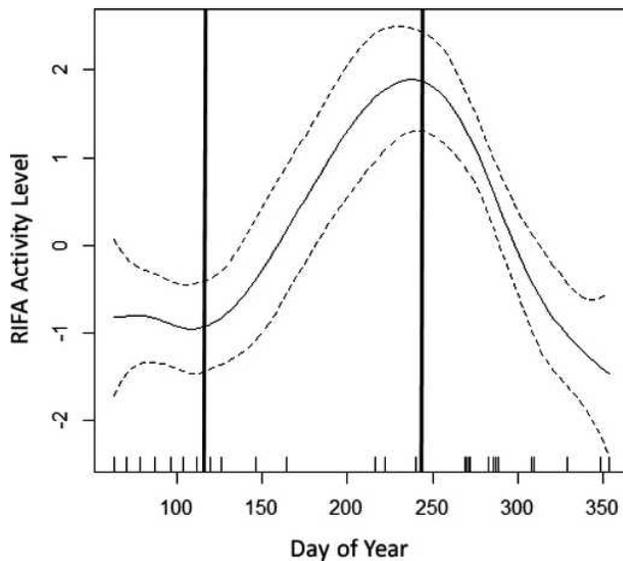
To assess whether prescribed fire influenced RIFA CPUE around ponds, we used the before-after control-impact approach described by Stewart-Oaten et al. (1986, 1992). One burn pond (pond 13) was not included in the statistical analyses because we captured RIFAs on very few sampling events, including none after the burn. We paired treatment (i.e., burned) ponds with control ponds that were similar in preburn temporal CPUE dynamics. For each pair, we computed the difference in CPUE between the control and treatment pond on each sampling occasion, and we used a standard *t*-test to determine whether the mean difference between the control and treatment ponds changed after the prescribed fire. An important assumption with this analysis is that effects are additive (Stewart-Oaten et al. 1986). We tested this assumption using Tukey's test of additivity (Tukey 1949), and two of the three pond pairs indicated nonadditive effects ( $P < 0.001$ ). We transformed CPUE for all ponds using the arcsinh transformation, which was effective for all pairs ( $P = 0.904$  [ponds 5 and 10],  $P = 0.392$  [ponds 9 and 14],  $P = 0.407$  [ponds 16 and 15]), and we used the transformed data for the *t*-tests. Levene's test for equality of variances indicated that variances were equal for all comparisons; thus, unequal variance *t*-tests were not used. However, results from unequal variance *t*-tests were equivalent. We performed this analysis using SPSS Statistics (Version 20).

## Results

We captured an estimated 18,050 RIFAs during this study, with estimated number of captures among ponds ranging from 4 (pond 13) to 3,642 (pond 11; Figure 1; Table S1, *Supplemental Material*, <http://dx.doi.org/10.3996/012012-JFWM-010.S1>). The generalized additive model fit a quadratic curve to the CPUE–DOY relationship, with above-average activity between May and October (Figure 2). We found a strong negative relationship between percentage of canopy cover and mean RIFA CPUE (Figure 3;  $t_8 = 11.08$ ,  $P < 0.0001$ ,  $r^2 = 0.82$ ). We did not detect a prescribed fire effect on RIFA CPUE for ponds burned in the winter (pond 10:  $t_{25} = 1.338$ ,  $P = 0.193$ ; pond 13: number of preburn captures = 4, number of postburn captures = 0). We also did not





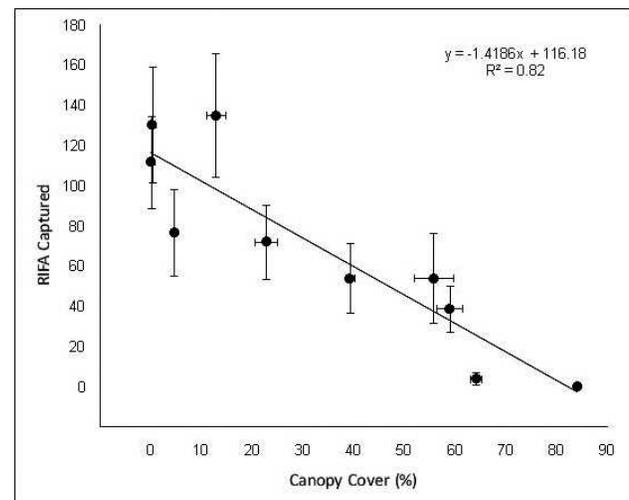


**Figure 2.** Annual activity pattern of red imported fire ants *Solenopsis invicta* (RIFA) in the Lost Pines ecoregion of Texas based on captures-per-unit-effort (CPUE). We sampled RIFA 27 times at 10 ponds between 26 September 2009 and 10 October 2010. Sampling dates are shown with hash marks inside the x-axis. For this analysis, we used CPUE at each pond on each day sampled (i.e., 27 samples with 10 observations per sample). We modeled CPUE as a function of day of year (DOY) using a generalized additive model. The y-axis shows the predicted activity level (with point-wise 95% confidence bands) based on results from days we sampled (inner tick marks) and relative to the observed mean CPUE for all observations (0). The vertical lines enclose the time period in which terrestrial juvenile Houston toads *Bufo houstonensis* may be present near ponds.

detect a prescribed fire effect on RIFA CPUE for ponds burned in the summer (pond 14:  $t_{25} = -0.316$ ,  $P = 0.755$ ; pond 15:  $t_{25} = -0.630$ ,  $P = 0.534$ ).

## Discussion

The results of this study indicate that disturbance from low-intensity prescribed burning did not affect RIFA abundance around ponds in our study area. This finding is in contrast to other studies reporting that disturbance from livestock use or vegetation clearing can lead to increases in RIFA abundance (Lofgren et al. 1975; Stiles and Jones 1998; Todd et al. 2008; Vogt et al. 2009). In the present study, the number of RIFAs captured at the two summer burn ponds decreased immediately after burning, but it returned to background levels within weeks. The winter burns were conducted during a period of low RIFA activity at all ponds, and no short-term changes were apparent. These results indicate that, at least in the short-term, prescribed burning did not benefit RIFA. However, each experimental pond was burned only once, and burn frequency may increase RIFA



**Figure 3.** Relationship between mean percent canopy cover ( $\pm$  SE) and mean red imported fire ant *Solenopsis invicta* (RIFA) captures ( $\pm$  SE) at 10 ponds in the Lost Pines ecoregion of Texas. We sampled RIFA at ponds 27 times between 26 September 2009 and 10 October 2010 and used the mean of the samples for this analysis. For percentage of canopy cover, we used the mean of 4 to 24 samples taken between 20 July 2008 and 16 April 2010. We found a strong inverse relationship between mean RIFA captures and percentage of canopy cover.

abundance (Hanula and Wade 2003). Additional research is needed concerning effects of burn frequency and intensity on RIFA in both forest and nonforest systems. Furthermore, due to strong spatial and temporal capture variability a strong impact would have been necessary to detect an effect. Future work on this topic would benefit from greater sample sizes, greater sampling effort, and longer study durations. We also note that RIFA were present at our study ponds before burning; thus, we did not address the potential for habitat invasion after disturbance through fire.

Percentage of canopy cover was a viable predictor of RIFA CPUE in our study area. This result was not surprising, because this species is known to select for open and edge habitats (Stiles and Jones 1998; Colby and Prowell 2006). We recommend that future studies seek to determine whether substrate-type (i.e., litter-dominated or vegetation-dominated) is an important RIFA CPUE predictor in forested environments, because fire can increase understory herbaceous vegetation density by removing litter and sunlight-capturing woody vegetation (Hodgkins 1958; Sparks et al. 1998; Donato et al. 2009). Because of constantly shifting pond borders due to deposition of precipitation and evapotranspiration, we were unable to test substrate-type in this study, because substrate-type shifted at several ponds. Quantifying substrate relationships would potentially increase predictive power for estimating RIFA

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**Figure 1.** Red imported fire ant *Solenopsis invicta* (RIFA) captures at pond edges in the Lost Pines ecoregion of Texas. We sampled each pond 27 times between 26 September 2009 and 10 October 2010. The solid vertical bars delineate preburn and postburn samples at burn ponds. Captures decreased immediately following a summer prescribed burn at ponds 14 and 15 but returned to background levels within weeks.

densities around ponds, as well as increase our understanding of the causal mechanisms behind fire effects on RIFA.

Monitoring RIFA over the course of a year showed that activity varied seasonally, with highest activity between May and October. This includes the period where juvenile Houston toads are vulnerable to predation near ponds. Houston toad emergence from ponds in the Lost Pines typically occurs between March and June, and juveniles remain near the pond for the first 1–2 mo after emergence (Greuter and Forstner 2003; Greuter 2004). Given that they are active, RIFA abundance differences could potentially significantly affect survivorship rates of terrestrial juvenile Houston toads. Thus, future research on juvenile Houston toad predation by the RIFA is warranted.

Based on the results of this study, low-intensity prescribed burning did not decrease RIFA CPUE around pond margins, suggesting that this kind of fire is not a useful management tool for eliminating RIFA. Chemical control has been shown to be locally effective, but insecticides can have severe impacts on nontarget wildlife, particularly amphibians (Lofgren et al. 1975; Boone and James 2003; Relyea 2003). If chemical control is necessary for managing RIFA in the Lost Pines ecoregion, we hypothesize that fall or winter application may have the lowest amphibian impact because amphibian activity is low relative to spring and summer (Brown et al., in press). Furthermore, although broadcast application is the most effective way to control RIFA, strategic bait placement (e.g., on mounds) also can be effective (Williams et al. 2001; Allen et al. 2004). Because amphibians do not seem to actively avoid soils contaminated with harmful chemicals (Hatch et al. 2001; Storrs Méndez et al. 2009), and these chemicals can be readily absorbed through the skin, strategic bait placement is a more attractive management option. Biological controls, such as the fire ant decapitating fly *Pseudacteon curvatus* and the disease-causing protozoan *Thelohania solenopsae* may eventually prove to be effective broad-scale control agents (Brinkman and Gardner 2001; Porter 2010).

### Supplemental Material

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**Table S1.** Data collected for this study evaluating red imported fire ant *Solenopsis invicta* (RIFA) activity around ponds on the Griffith League Ranch, Bastrop County, Texas, USA. Included in the file are mean percentage of canopy cover, burn treatment, burn status, and RIFA captures-per-unit-effort for each pond.

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