

Section 6 (Texas Traditional) Report Review

Form emailed to FWS S6 coordinator (mm/dd/yyyy): [Click here to enter a date.](#)

TPWD signature date on report: 8/27/2015

Project Title: Testing habitat restoration options in the Houston toad (*Bufo houstonensis*) within the aftermath of the Bastrop County Fire Complex

Final or Interim Report? Final

Grant #: E-149-R

Reviewer Station: Austin ESFO

Lead station concurs with the following comments: NA (reviewer from lead station)

Interim Report (check one):

- Acceptable (no comments)
 - Needs revision prior to final report (see comments below)
 - Incomplete (see comments below)
-

Final Report (check one):

- Acceptable (no comments)
 - Needs revision (see comments below)
 - Incomplete (see comments below)
-

Comments:

We appreciate the tremendous amount of work that has gone into this study. It is sound research that will be helpful in designing conservation management strategies for the endangered Houston toad. However, it appears the report could use some additional editing and revision, as we need final section 6 reports to be polished and free from grammatical and formatting errors.

Please make sure that all of the parenthetical references are formatted consistently. See page 52 of the report for an example: (Grueter 2004) and (Vandewege, 2013) are used in the same sentence. One has a comma before the date and one does not.

Please review the entire report for correct punctuation use. A few examples of incorrect punctuation are provided below.

- Commas should follow transitions words at the beginning of a sentence, such as the sentence beginning with “Therefore” on the bottom of page 50 – “Therefore, little evidence that support...”
- Commas should come after dependent clauses. Commas should also come before nonrestrictive clauses. Both comma mistakes can be found throughout the report, as in the sentence at the top of page 50 – “To best utilize the stocking resources we had available we

drew from Harper and Semlitsch (2007) which demonstrated survivorship and growth to be highest in lower stocking densities.” Commas should be placed after “available” and before “which.”

- Commas should come after dates. For example, a comma is needed after “June 6, 2011” in the first sentence of the middle paragraph on page 8 of the report. Check also the dates on the bottom of page 12 for consistency in formatting.

Please review the report for sentence structure problems, such as the sentence at the bottom of page 50 that continues on to page 51. “Therefore, little evidence that support juvenile Houston toads spend any considerable amount of time below the soil surface or implement estivation behavior during summer and winter months as do the adults.” This reads like a fragment instead of a complete sentence.

Please review the report for odd phrases that confuse the reader. For example, the phrase “implement estivation behavior” on the top of page 51 could be revised to say “demonstrate estivation behavior.” Also, the phrase “Undergo inter-pond dispersal” in the last paragraph of page 51 could be revised to say “disperse between ponds.”

In the second sentence of the last paragraph on page 50 of the report, “affect” should be changed to “effect.”

Please consider numerals for numbers greater than nine unless otherwise required by journal formatting guidelines. For example, “sixty zoo raised male Houston toads” on page 8 could be revised to be “60 zoo raised male Houston toads.”

FINAL PERFORMANCE REPORT

As Required by

THE ENDANGERED SPECIES PROGRAM

TEXAS

Grant No. TX E-149-R

F12AP01164

Endangered and Threatened Species Conservation

**Testing habitat restoration options in the Houston toad (*Bufo houstonensis*)
within the aftermath of the Bastrop County Fire Complex**
Prepared by:

Mike Forstner



Carter Smith
Executive Director

Clayton Wolf
Director, Wildlife

27 August 2015

FINAL PERFORMANCE REPORT

STATE: Texas **GRANT NUMBER:** TX E-149-R

GRANT TITLE: Testing habitat restoration options in the Houston toad (*Bufo houstonensis*) within the aftermath of the Bastrop County Fire Complex

REPORTING PERIOD: 1 September 2012 to 31 August 2015

OBJECTIVE(S): To place habitat exclosures, stocked with headstarted Houston toads, within differing fire intensity zones and under alternative habitat recovery options to enable direct measurement of survivorship under those different management strategies.

Segment Objectives:

Task 1: purchase necessary aluminum flashing, verify field locations for all exclosure placement, gather brush and mulch materials as required at each site and install the exclosures, placing Houston toads into the exclosures as soon as all are completed and biosecurity confirmations are met.

Task 2: continue intensive monitoring of the toads and begin processing of results enabling a December 15th delivery data for initial dissemination and discussion of those outcomes with TPWD and USFWS. Jan-May (all remaining years) – field monitoring continues for all exclosures and any alternative treatment options are prepared for establishment (e.g. alternative seed mixes or the projected available loblolly seedlings).

Task 3: June-July (all remaining years) – initial Houston toad cohort is captured and removed from the exclosure system for release after measurement, this will be correspondent to periods of highest activity (ie. heavy rain events).

Task 4: Aug-December (all remaining years) – all exclosures are re-established with a new cohort of male headstart toads, alternative treatments (if any) are established and monitoring continues with new results again slated for interim reporting at the end of the fall activity period (December 15, 2013).

Task 5: August 2015 – all exclosures have been emptied of treatment toads, final results from the tests are compiled and offered as a set of best management practices for habitat recovery after catastrophic fire or other severe disturbances.

Significant Deviations: None.

Summary Of Progress: See Attachment A.

Location: Bastrop County, Texas.

Cost: Costs were not available at time of this report.

Prepared by: Craig Farquhar

Date: 27 August 2015

Approved by: 

Date: 27 August 2015

C. Craig Farquhar

ATTACHMENT A

2015 Final Report

Testing Habitat Restoration Options in the Houston Toad (*Bufo houstonensis*)
Within the Aftermath of the Bastrop County Complex Fire

Project E-149

Submitted to:

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August 27th, 2015

INTRODUCTION

The Lost Pines region of Texas is a loblolly pine (*Pinus taeda*) and oak dominated woodland forest located at the boundary of the Colorado river and the Carrizo-Wilcox aquifer (Brown and Mesrobian, 2005) and currently retains fragments in, Austin, Bastrop, Colorado and Fayette Counties (Taber and Fleenor, 2003). Bastrop County, Texas has historically supported the largest known, and best studied, population of Houston toads (U.S. Fish and Wildlife, 1984; Dixon et al., 1990). First described in 1953 (Sanders, 1953), Houston toad populations quickly became scarce at all of the known localities. The cause(s) of population decline remain unknown, but many speculate the decline was due to the severe drought of the 1950's coupled with the expansion of the city of Houston (U.S. Fish and Wildlife, 1984). Since 1978, Bastrop State Park and the surrounding areas have been designated critical habitat for the Houston toad. Approximately half of the acreage within designated critical habitat was impacted by the Bastrop Complex Fire of 2011, increasing the necessity of understanding Houston toad survivorship within unburned and wildfire modified habitats.

The Houston toad was first described in Houston, Texas in 1953 (Peterson et al., 2004; Sanders, 1953). In 1970, the Houston Toad was the first animal in Texas and the first amphibian federally listed as an endangered species (Peterson et al., 2004). A high correlation has been found between the sandy loam soil that occur in the Lost Pines ecoregion and Houston toad (*Bufo houstonensis*) occurrence (Koepp et al., 2004). It has been suggested the Houston toad is a poor burrower (Bragg, 1960), therefore the sandy soils enable them to bury down and aestivate during the cold winter months. Houston toads are therefore thought to be restricted to areas of sandy loam soils, however not necessarily pine

forests (Brown and Thomas, 1982). Nearly all recovery efforts have centered on the “robust” population remnant in Bastrop County. Houston toad populations have remained in a continual decline consequent of multiple stressors, including habitat fragmentation, continued urban growth of the city of Bastrop, red imported fire ants, fertilizers and chemical run off, agricultural practices, wildfire, and drought. Although all these factors negatively impact toad populations, the Bastrop County Complex fire is of primary concern for the survival of this endangered species.

The aftermath of the Bastrop County Complex fire has left Bastrop County with the need for immediate and active restoration of the plant community on public and private land in order to restore the integrity of the Lost Pines ecoregion. Restoration actions, along with some of the necessary expenses, will require landowner support and involvement and habitat and wildlife management. In order to guide private and public landowners in effective post fire habitat restoration strategies that will in turn improve habitat for the Houston toad, we must determine optimal habitat recovery options for toad survivorship.

Study Areas - This study was conducted on two neighboring properties located within Bastrop Co., TX. The Griffith League Ranch is a 1,900 ha ranch owned by the Boy Scouts of America, and Welsh is a 184 ha ranch held by Bastrop Co. and managed by Texas State University. Both properties are designated by the USFWS as Houston toad critical habitat and are currently managed primarily through habitat restoration efforts. The IACUC permit number for this research is 1011_0501_11.

TESTING SURVIVORSHIP OF ADULT HOUSTON TOADS IN SIMULATED WILD HABITAT ENCLOSURES

Introduction – Many studies have shown that fire can have both positive and negative effects on wildlife populations. These fire effects are driven by multiple factors such as fire intensity, fuel load, wind, and relative humidity (Esque et al., 2003). Moseley et al. (2003) suggests low intensity, low impact prescribed fire has little negative effect on wildlife populations. Fire, in many cases, has been shown to increase population densities (Minshall et al., 1989; Greenberg and Waldrop, 2008; Brown et al., 2011). The natural history and behavior of individual species and how quickly they can adapt to the changing environment will also affect how they respond to fire. Compared with other vertebrates, amphibians have much smaller dispersal and movement capabilities (Sinsch, 1990; Bury et al., 2000; Semlitsch and Bodie, 2003; Bowne and Bowers, 2004), which could increase direct mortality. Furthermore, the moist permeable skin of amphibians would increase their vulnerability to smoke and heat and may lead to dehydration (Stebbins and Cohen 1995; Bury et al., 2000). Habitat type, topography, or the presence of wetlands may create refugia and provide protection to animals during fire (Whelan, 1995). Hossack and Corn (2007) observed a slight increase in amphibian populations at local wetlands post wildfire.

Fewer studies have investigated the effects of catastrophic wildfire on amphibian populations. As fire intensity increases, so may the chances for an increase in direct and indirect mortality of certain species. Direct mortality can be caused by direct exposure to flames, ash, and smoke and occurs immediately as the fire passes across the landscape. Indirect mortality can be caused by changes in habitat, reduction in food, water and shelter, and lower nutrient availability. Brown et al. (2014) reported minimal direct mortality on

amphibians post wildfire, and Greenberg and Waldrop (2008) reported a higher abundance of American toads in burned habitats compared to unburned habitats post wildfire.

The frequency of extirpation of a population or the extinction of a species due to a fire event or other habitat disturbance event is low (Thomas et al., 2004). However, the situation in Bastrop, for Houston toads, is abnormal. The species had only one large population center remaining prior to the fire, which itself was coupled with the ongoing extreme drought. This provides a scenario where the catastrophic fire is paired with extreme drought, affecting the only genetically diverse, large, population fragment of a species that remains. As noted in the 1994 population viability assessment (Seal, 1994), extinction or extirpation risk probabilities, for the Houston toad, are truly a serious concern.

While surveys allowed us to examine amphibian diversity and abundance before and after prescribed fire (Brown et al., 2011; Jones et al., 2006) and wildfire (Brown et al., 2014) in Bastrop County, we still need to continue to test habitat suitability pre and post catastrophic wildfire specifically for the Houston toad. Houston toad response to the newly altered landscape following the Bastrop County Complex fire is unknown and at best speculative.

When this study was originally proposed we sought to test and compare adult Houston toad survivorship within the various different habitat types that existed prior to the fire. During the initial study implementation, the Bastrop Complex Fire occurred and we included the wildfire affected habitats within the modified study design. We sought to gain insight on habitat suitability for the Houston toad given the current conditions found across Bastrop County. These data will hopefully provide suggestions on immediate and ongoing restoration efforts leading toward habitat and population recovery from the 2011 Bastrop

Complex Fire, but also future fire recovery efforts.

Key to enabling this type of evaluation using adult Houston toads, is the availability of improved microchip technology (BioMark) enabling detection even when the toad is buried up to 10 – 12 cm deep. The availability of different habitat types within these study sites, and the coincident availability of adult Houston toads to release into replicate enclosures in those different habitats able us to test survivorship and growth over time. The results from this study seek to inform to the relevant management agencies and Bastrop County stakeholders in order to help guide habitat and wildlife management for public and private lands in Bastrop County and may also be used as a blueprint for management strategies in other fire prone regions of the Southeast United States. Testing habitat recovery options, then establishing the best management practices on public and private lands is the core benefit of the described studies. The studies provide results and benefits for future management options in hopes of offsetting additional mortality to Houston toads during recovery efforts within the altered landscape.

Quantitatively estimating demographic parameters from mark-recapture (henceforth MR) studies have advanced considerably over the last three decades (Lebreton and Pradel, 1992; Burnham and Anderson, 2002). Currently, most MR studies use multi-model analysis in information-theoretic framework to estimate survival (ϕ) and the probability of recapture (p) (Burnham and Anderson 2002, Schmidt et al., 2002). Statistical inference from model selection under an information-theoretic approach requires rigorous attention to selecting the candidate set of models. Briefly, a candidate model set is developed using a priori hypotheses focusing on the relationship between survival and recapture, and covariates, such as treatment effects, environmental parameters, among others. Models are ranked based a

selection criterion, most commonly Akaike's information criterion (AIC_c) (Akaike, 1973), which provides a reliable decision criterion for model selection for both nested and non-nested models (Schmidt and Anholt, 1999; Burnham and Anderson, 2002).

This study used an information-theoretic approach to model selection to choose models that best fit MR datasets collected from enclosure experiments conducted in 2011 – 2012, and 2013 – 2014; where four adult Houston toads were released into large outdoor enclosures representing different habitat types (described above). For each model ϕ and p were estimated.

Material and Methods – Two adult survivorship and habitat suitability trials were conducted between June 2011 and March 2014. Trial 1 compared adult Houston toad survivorship among three habitat types; pine, oak, and juniper dominated habitat patches in Bastrop County prior to the Bastrop County Complex fire. Trial 1 began June 2011 and was completed in March 2012. A second trial (trial 2) was conducted after the Bastrop County Complex fire of 2011. This second trial compared adult Houston toad survivorship among four habitat types, adding a wildfire burned habitat along with the pine, oak, and juniper habitats. These two trials are intended to illustrate habitat suitability in current habitat patches in Bastrop County.

Trial 1 – Prior to the Bastrop County Complex fire we sought to evaluate habitat and adult head-start toad survival in three habitats using a field enclosure experiment. Three habitats were selected for the preliminary study; loblolly pine dominated, oak dominated, and juniper dominated woodlands or forest patches. Five enclosures were built within each habitat for a total of 15 enclosures. Each enclosure is approximately 10 x 10 m², built using

galvanized aluminum flashing. The flashing is buried 10 to 12 cm deep within the soil substrate in order to prevent toads from tunneling under and escaping.

Male Houston toad adults, that were captive raised at the Houston Zoo, were released within each of the 15 exclosures. These adults were raised from wild population eggstrands collected in Bastrop State Park and Griffith League Ranch in Bastrop County, Texas. A total of four toads were placed within each exclosure. Upon release, each toad was implanted with a BioMark Passive Integrated Transponder tag or PIT tag. The BioMark PIT tags can be read at a depth of 10 – 12 cm beneath the surface even when the animal is underground and buried under logs and other debris. These PIT tags enabled us to monitor the location and movement of each toad over time with minimal disturbance using the subsurface detection abilities of the BioMark chip reader.

On June 6, 2011 sixty zoo raised male Houston toads were divided evenly and placed within the 15 exclosures. Upon release, each toad was weighed, snout urostyle length (SUL) and head width (HW) measured, and pit tagged. As toads were released, pit tag numbers were recorded so the location of each toad was confirmed. The first week after initial release, toads were checked every other day. As toads were found, they were flagged and numbered and movement was recorded (Figure 1). Toads were then surveyed 2 to 3 times a month usually following rain events, from June 2011 to March 2012.

Survival estimates – Survivorship estimates were conducted in Program MARK (White and Burnham, 1999) using the Cormack-Jolly-Seber mark-recapture model (Cormack, 1964; Jolly, 1965; Seber, 1965). We assumed that capture probability was at 100% at the conclusion of trial 1. Each exclosure was searched extensively upon and post

completion of trial 1 until no more toads were discovered. All individuals that were captured in subsequent censuses were known to be alive in any previous census.

Canopy Cover – Canopy cover was collected twice during trial 1 (July 2011 and October 2011) for each habitat. Canopy cover was estimated for each exclosure in the three habitats using a spherical crown densitometer. For each exclosure, estimates were taken from nine points arranged in a grid formation with three rows and three points per row. These nine points were then averaged and percent cover reported for all 15 exclosures. Differences among habitats were assessed with a single factor analysis of variance (ANOVA).

Red Imported Fire Ant Counts – Fire ants were trapped, collected, dried, identified, and counted for all 15 exclosures. For each exclosure, nine petri dishes were placed in a grid formation with three rows of three dishes per row. The points used for this procedure were the same nine points used to collect canopy cover. Each dish was baited with one half piece of Vienna sausage link. The bait traps were deployed in each exclosure for 45 minutes then picked up and placed in a Ziploc bag. The bags of ants were put on ice to reduce ant activity while in the field. Ants were then flash frozen, dried, then identified and sorted. Differences among habitats were assessed with a single factor ANOVA.

Trial 2 – In order to test habitat suitability subsequent to the catastrophic wild fire, five additional exclosures were added to the Griffith League Ranch study site in a location that was severely burned (i.e., complete loss of canopy) during the Bastrop County Complex fire, creating a fourth habitat to be tested. These additional exclosures allowed us to test fire habitat suitability as well as replicate our previous exclosure study. For this second study four Houston toad adults were placed into each of the 20 exclosures for a total of 80 adult toads. Each exclosure contained two males and two female toads, allowing us to compare

survivorship among sex. Each toad was implanted with a BioMark PIT tag prior to release. SUL, HW, mass and photographs were collected for each adult toad. Exclosures were monitored 2 to 3 times a month from March, 2013 until March, 2014.

Survival estimates – Survivorship estimates were conducted in Program MARK (White and Burnham, 1999) using the Cormack-Jolly-Seber mark-recapture model (Cormack, 1964; Jolly, 1965; Seber, 1965). We assumed that capture probability was at 100% at the conclusion of trial 2. We justify this presumption, in that, each exclosure was searched extensively upon and post completion of trial 2, iteratively until no more toads were discovered. All individuals that were captured in subsequent censuses were known to be alive in any previous census.

Canopy Cover – Canopy cover was collected twice during the study (July 2013 and January 2014) for each habitat. Canopy cover was estimated for each exclosure in the four habitats using a spherical crown densitometer. For each exclosure, estimates were taken from nine points arranged in a grid formation with three rows and three points per row. These nine points were then averaged and percent cover reported for all 20 exclosures. Differences among habitats in canopy cover were assessed with a single factor ANOVA.

Model selection procedure methods trial 1, trial 2, and males vs females– Using a model selection approach based on information-theoretic methods, Program MARK (White and Burnham, 1999) was used to estimate the probabilities of ϕ and p for adult Houston toads. . Methods followed Cooch and White (2006). Two explanatory factors were used to explore variation in ϕ and p : time and habitat type. Time was considered as constant among sampling periods (\bullet) or variable across periods. Habitat type (ht) was treated as a categorical covariate with three levels (juniper, oak, and pine) and used to determine if habitat type

affected ϕ or p . Based on these factors, eight candidate models were developed, where each model represented a different biologically-based hypothesis that explored the effects of time and habitat type on estimates of ϕ and p . For example, $\phi_t p_t$ represented the Cormack-Jolly-Seber model (CJS) that is fully time dependent for both ϕ and p . Whereas $\phi_{ht} p$ represented a model where survivorship varied among habitat types, and p remained constant among sampling periods.

The amount of support for each of the eight candidate models was evaluated using a correction factor for AIC (AIC_c) which protects against over-fitting the models, especially with small sample sizes (Hurvich and Tsai, 1989). The model with the lowest AIC_c was considered to best fit the data unless the difference in AIC_c values (ΔAIC_c) among competing models was < 2.0 , then the models were considered indistinguishable. Models were ranked from one to eight, with one being the best supported model and eight being the least. If multiple models supported the data, the most parsimonious model was chosen as the best supporting models. Point estimates, standard errors and 95% confidence intervals were recorded for ϕ and p for each model.

In trial 2 we were able to test survivorship and recapture between females and males. Using a model selection approach (described above) we estimated the probabilities of ϕ and p for males and female adult Houston toads. Two explanatory factors were used to explore variation in ϕ and p : time and sex. Time was considered as constant among sampling periods (\bullet) or variable across periods. Sex (s) was treated as a categorical covariate with two levels (males and females) and used to determine if sex affected ϕ or p . Based on these factors, 9 candidate models were developed.

Results

Trial 1

Toad Detection In Exclosures – There is a decrease in total toad detection among all three habitats overtime. Total detection started at 68% during census one decreasing to 2% total detection at the conclusion of trial 1 (Figure 2). Detection percentage was calculated using the total number of toads detected each census divided by the number of toads assumed to be alive at each census.

During the study, PIT tags were found among the debris or under the sandy soils. These lone PIT tags were either the only remnant of a mortality event or, likely a PIT tag that had been shed or expelled out through the skin. It is not uncommon for a toad to expel a PIT tag once it has been implanted. Tags can migrate out of the skin from the tag insertion point, leaving a live toad very difficult to detect. The Houston Zoo reports a 10% expulsion rate for PIT tags implanted into their adult Houston toads (Paul Crump, personal comm. 2012). During trial 1, eight PIT tags have been found within the exclosures. One tag found in juniper habitat, two found in pine habitat and five tags found in the oak habitat. These eight tags represents 13% of all pit tags used in trial 1. To prevent bias, we removed individuals corresponding to the lost tags from detection estimates post date of tag discovery because it is unknown if these toads represent live (undetectable) or dead (mortality event) individuals.

Toad detection was initially high for pine and juniper habitats at 86% and 65% detection for the first census (Figure 3). Detection was at 23% for the first census in the oak habitat, jumping to 50% by census 2, however quickly dropping down to 19% by census 4. Pine was the only habitat where detection remained above 50% until August 16th 2011 (census 11). Detection hit 0% in the juniper and oak habitats by August 25th, 2011 and September 20th, 2011 respectively. On March 25th 2012 we concluded trial 1 and

aggressively searched all 15 exclosures for remaining toads. Duff layer was moved along with debris, rocks, and limbs. A single toad was detected in the pine habitat at the end of trial 1.

Although a decreasing detection trend is noticeable, a slight increase in detection within the pine and cedar habitats was noticed on June 30th, 2011 (census 6) (Figure 3). This increase in detection is positively correlated with a 3.81 cm rainfall event that occurred on June 22.

Toad Survivorship trends and MARK recapture ϕ and p estimates– During the first month of the study, total toad survivorship decreased dramatically. Total survivorship fell below 50% by July 17th, 2011 (census 7) (Figure 4). In the juniper and oak habitats, survivorship decreased by 30% between census 1 and 3. Pine habitat survivorship did not fall below 50% until census 13 (Figure 5). Significant differences between survivorship over time and among habitats were seen in trial 1 (ANOVA: Habitat, $df = 2$, $F=47.159$, $p= <0.001$; Time, $df=1$, $F=291.644$, $p=<0.001$).

Although we found deceased toads during these trials, we were unable to determine the fate of the individuals from missing chips. Therefore we did not account for joint live and dead recaptures in our MARK models. Based on ΔAIC_c , model 1 ($\phi_{ht} p_{ht}$) was the best supported model (Table 1). For this model, ϕ and p varied with habitat type (Table 2), but not across time, with pine having the highest values for both ϕ (0.92) and p (0.79). Juniper had the second highest ϕ (0.84), but had the lowest p (0.6). The other seven models had ΔAIC_c values > 2.0 , which indicates that habitat type had a stronger effect on the data compared to the most parsimonious model (Model 3, $\phi.p$).

Canopy Cover – Average canopy cover during trial 1 was 78.7 % in the pine habitat, 76.7 % in the oak habitat, and 82.6 % in the juniper habitat. Canopy cover did not differ among the habitats (ANOVA: $df = 2, 12, F = 0.223, p = 0.804$). The juniper habitat contain understory of yaupon holly along with oak, increasing canopy cover. The pine and the oak habitats are clear of understory growth allowing more sunlight to penetrate to the forest floor.

Toads and Red Imported Fire Ants – Several deceased toads were found covered in red imported fire ants (RIFA). It is uncertain whether this is the direct cause of mortality or if the ants acted as opportunists once the toads were dead. We tested for the abundance of RIFA within all 15 exclosures to see if ant abundance was correlated with toad mortality within habitats. Ants were sampled in July 2012 after the trial had been concluded, in order to prevent attracting fire ants to the exclosures when toads were present. We sampled during the summer months when fire ant movement is at its highest. Red imported fire ants did not differ among treatments (Table 3) (ANOVA; $df = 2, 12, F = 1.741, p = 0.22$).

Trial 2

Toad Detection In Exclosures – Trial 2 began March, 2013 and was concluded April 2014. There is a decrease in total toad detection overtime and among all habitats (Figure 6). Detection had a sharp initial decrease at the beginning of the study followed by a continual decrease in detection through the summer and early fall. As temperatures rise during the summer months and then fall during the winter months toads will bury down deep to avoid desiccation. This can decrease chances of detection. Although a decrease in detection is expected, as Spring of 2014 approached, detection continued to decline until detection hit 0% on April 6th, 2014.

Among habitats detection was the highest within the oak and pine habitats (Figure 6). Although detection within the juniper habitat had decreased to 13% by the 6th census, detection increased above 30% following the rain events in May. Detection fell to 0% for all habitats by April 2014.

We found detection to be positively correlated with rain events (2013 – 2014 study) (Figure 7). A small increase in detection was observed on May 26th, 2013, July 27th, 2013 and November 22nd, 2013. Two large rain events also occurred December 15th, 2013 and February 9th, 2014. An increase in detection was not observed following these two rain events.

A total of 19 pit tags were found during this study. This represents 24% of the total number of tags used in this study. Two tags were found in the oak habitat, four in the pine habitat, five in the juniper habitat, and eight in the burned habitat. Individuals that corresponded with these tags were not used in detection estimates post date of tag discovery.

Male vs Females – There was no difference in survivorship between male or female toads. Out of 80 total Houston toads, 22 males and 22 females were found deceased or a lost PIT was discovered. Therefore sex ratio (M:F) for detected toads was 1:1 for males to females. In the wild, explosive breeding amphibian sex ratios are commonly male biased (Wells, 1977; Davies and Halliday, 1979) caused by several factors such as, unequal sex ratios at birth, differences in male/female mortality rates, differences in male/female migration rates (Swannack and Forstner, 2007), and delayed maturation (Gibbons, 1990). In a controlled environment without breeding pressures and a decrease in natural predator opportunities males and females are able to survive across the landscape equally.

Based on ΔAIC_c , model 1 ($\phi(\cdot)p(\cdot)$) was the best supported model (Table 4). For this model, ϕ and p were constant across time and between sex (Table 5) with ϕ (0.88) and p (0.62). The other eight models had ΔAIC_c values > 2.0 , which indicates that the most parsimonious model had the strongest effect on the data.

Survivorship trends and MARK data among habitats of trial 2 – We report a 5.0% decrease in survivorship per visitation event for pine, 3.5% in oak, 3.6% in juniper and a 3.2% decrease in survivorship in burned habitat (Figure 8). Pine was the only habitat to sustain survivorship above 50% beyond May of 2013 (census 7). Pine survivorship fell below 50% July 17th, 2013. Pine habitat went from 50% survivorship on July 17th to 0% survivorship by August 20th, 2013 (Figure 8). June 1st marks the end of Houston toad breeding season. Breeding events beyond June 1st have been documented, but are rare. Survivorship on June 1st was at 65% in pine, 46% in oak, 27% in juniper, and 16% in burned habitat. During the breeding season (January 1st – June 1st) Houston toad activity should be at its highest. Total survivorship can be seen in Figure 9.

A total of 17 toads (21 %) were found deceased during Trial 2. Five deceased toads were found in the oak habitat, six were found in pine habitat, four were found in juniper habitat, and two were found in the burned habitat. Deceased toads are collected and transported to the tissue collection at Texas State University under federal permit # TE039544-0.

Based on ΔAIC_c , model 1 ($\phi_{ht}p_{ht}$) was the best supported model (Table 6). For this model, ϕ and p varied with habitat type (Table 7), but not across time, with oak having the highest values for both ϕ (0.91) and p (0.78). Pine had the second highest ϕ (0.89), and second highest p (0.65). The other seven models had ΔAIC_c values > 2.0 , which indicates

that habitat type had a stronger effect on the data compared to the most parsimonious model (Model 4, $\phi.p.$).

Canopy Cover – Average canopy cover during this study was 57.6% in the pine habitat, 72.0% in the oak habitat, 78.9% in the juniper habitat, and 4.0% in the burned habitat. Canopy cover was highest in the juniper habitats. The juniper habitat contains understory of yaupon holly which increases canopy cover. The pine and the oak habitats are clear of understory growth allowing more sunlight to penetrate to the forest floor. The burned habitat contains little to no overstory cover due to the severity of the Bastrop County Complex fire as it moved through areas of the Griffith League Ranch (ANOVA; $df= 3, 16$, $F= 18.39$, $p<0.001$).

Old toad detections – During trial 1, we were monitoring three adult male toads that were part of an initial habitat suitability trial using the 15 original exclosures in three habitats. This initial trial, which began in March of 2010, had complications in detecting Houston toads below the ground. The original PIT tags used were not able to be detected using the biomark pit tag reader, therefore this trial was postponed until stronger chips were received. Toads that could be detected were removed and released in Bastrop County. During trial 1, three adult males from the 2010 attempt were discovered in the exclosures. These individuals had successfully overwintered and were detected the summer of 2011. Two toads were found in the juniper habitat during June of 2011 and the third toad was found in pine habitat in July 2011. Unfortunately these toads had lost the original PIT tags, therefore we cannot compare original SUL and mass. They were identified however as individuals in the original study due to toe clip markings taken as DNA samples before released into the

exclosures. These toads were measured, weighed and released back to their original exclosure.

Discussion – The purpose of this study was to compare survivorship among different habitat types located in Bastrop County. Two trials were conducted in order to replicate the experiment during and after the severe drought and fire of 2011. We were able to include a burned treatment in trial 2 to look at fire affected habitat outcomes. Unburned habitat sites used in this study are sites that have been considered optimal Houston toad occupied and breeding habitat and therefore have been used as Houston toad head-starting release sites. Since 2007 we have been working with the Houston Zoo and with the USFWS on head-starting the Houston toad. Current head-starting strategies have focused on releasing individuals (adults, juveniles, tadpoles and now eggs) in designated “suitable habitat” within Bastrop County. Natural history of the Houston toad has led us to believe these areas of “suitable habitat” contain dominant stands of loblolly pine accompanied by deep sandy loam soils. Not only have these studies helped us confirm many of these designated areas are indeed suitable habitat, we were able to make predictions on how suitable these areas remained post catastrophic wildfire.

The effects of habitat on survivorship and recapture of adult *B. houstonensis* were significant with differences in survivorship and recapture estimates between habitats but not overtime. In both trials pine and oak had the highest survivorship and recapture estimates, therefore further supporting our prior placement of head-start Houston toads in Bastrop County. Based on our data it is difficult to infer if pine or oak is the best habitat. Differences between pine and oak habitats between trial 1 and trial 2 can be due to temporal conditions between each year.

The best supported mark-recapture model for both trials was $\phi_{ht}p_{ht}$ where survivorship and recapture estimates varied among habitats but not over time. Survivorship and recapture estimates constant overtime, suggests temporal conditions were not a driving factor in these estimates. This is also supported by comparing detection and survivorship trend data among the two trials (Figure 10). Trial 1 was conducted in 2011 during an exceptional drought in this region and the majority of the state of Texas. Trial 2 was conducted in 2013 and 2014 where temperatures were cooler and rainfall had increased annually compared to 2011 – 2012. We would expect to see survivorship dependent overtime during 2011 as conditions continued to worsen. Although drought conditions may have some affect on Houston toad these data fit a model where time was not a significant factor.

We did not see a difference in survivorship or recapture rates between males and female Houston toads. In the wild amphibian sex ratios are commonly male biased (Wells, 1977; Davies and Halliday, 1979). The selection of this model matches the detection and survivorship trend data between male and females overtime, therefore supporting its selection. Differences in male and female mortality rates can be caused by the differences in their behaviors. Males can have increased rates of mortality during the breeding season when they are moving across the landscape multiple nights and actively calling at a pond edge. In a controlled environment without breeding pressures and a decrease in natural predator opportunities males and females are able to survive across the landscape equally.

Houston toad survivorship was lowest within the burned treatments ($\phi = 0.84$). It is not surprising to see this result, however with habitat altered in Bastrop State Park and the Griffith League Ranch by catastrophic fire, it has implication to further management strategies. In 2015 very few Houston toads were detected in Bastrop State Park or on any

burned locations in the Griffith League Ranch. Testing head-start survivorship has been difficult since 2010 and the most recent 2015 spring season rainy period have helped create a best case head-start release scenario in the last year of these studies. We will continue to test head-starting in these burned locations, but if trends continue we may need to focus all our concerns on unburned release locations.

Rain events in 2013 and 2014 illustrate Houston toad adults become more active during these rain events due to detection increases correlating with large rain events. These correlations were during early months of this study, however, as survivorship and detection decreased, rain events no longer contributed to an appreciable increase in detection the following spring. We would expect to see an increase in detection during spring rains when Houston toad breeding season is occurring. Cumulatively, we consider the evidence to strongly support that detection is a suitable proxy to survivorship for both studies.

Houston toad mortalities or loss of detections (found PIT tag) were discovered during both trials. In total, 30 out of 140 total toads used in both trials were found deceased and another 27 PIT tags were found in both trials. The numbers of deceased toads for both studies were 20, 6, 10, and 2 and PIT tags were 6, 7, 6, and 8 respectively from pine, oak, juniper and burned habitats. Pine habitat has the highest survivorship during these two trials and counterintuitively, the highest confirmed mortalities. Survivorship is lowest in the burned habitat with the lowest number of confirmed mortalities. We have documentation of toads using shallow burrows in the pine habitat where sand is loose. Toads have been found desiccated while emerging from these burrows (Figure 11). Thus, some of the mortality events are occurring close to the surface and more readily detected. Toads within the burn zone treatment are burrowing deeper sooner, presumably to find cooler soils. It is assumed

mortalities are high in this habitat but are occurring below the surface and therefore not physically detected as carcasses.

We have anecdotal evidence that Houston toads may burrow deeper into these soils during periods of intense drought or suboptimal habitat provided by the three adults from the pilot study. These individuals burrowed beneath our detection threshold of ~20cm. Although detection errors may exist during each census, we are confident support detection as a proxy for survivorship. While early deaths may have occurred as a result of the captive reared toads inability in their initial acclimation to the exclosures, we do not believe subsequent mortality to be failure to thrive, but natural mortality for adult Houston toads.

Red imported fire ants were found in most exclosures. Many deceased toads were discovered covered in fire ants or bones remaining after the tissues had been partially consumed by the ants. Fire ants can drive amphibian mortalities for those individuals emerging from the pond, however are not as commonly linked to adult mortalities. This predation and mortality has been tested in the Houston toad, where predation was linked to newly metamorphosed toadlets (Freed and Neitman, 1988). It is concerning however that fire ants are prevalent in areas that are documented as suitable habitat for the Houston toad. Currently we are managing for red imported fire ants in areas we are conducting head-start releases and will continue to test the effects of these invasive predators.

The results from this study are not encouraging, as adult survivorship is lower in both severe drought and post drought/ post wildfire conditions than predicted from model assessments (Swannack et al., 2009). Our study includes 2011, which was the worst drought in Bastrop County on record (Nielsen-Gammon, 2012). We accept that the realities of the drought, particularly the exceptional severity, may have influenced our results, but the

conditions during the second trial were not as severe as in 2011 and thus the strongly negative trends observed were not expected in 2013 and 2014 during Trial 2. These results improve our understanding of habitat suitability for this species and continue to refine our knowledge of how the current habitats are influencing population persistence. We can only infer from these data that survivorship is low for adult Houston toads and when tested in severe and then optimal conditions, the results are the consistent. Houston toads are utilizing habitats that we have tested to be most suitable and we will continue to release captive propagated toads in these areas. However, given we are releasing them in the habitats with highest survivorship thus far tested in Bastrop County, we need to determine what factor or factors influence these survivorship values.



Figure 1. Example an adult Houston toad (*Bufo houstonensis*) wild mesocosm enclosures in Bastrop County, Texas. This is an example of a replicate enclosure in the pine habitat. The

pink flags represent the last known location of each of the adult male Houston toads. Four adult Houston toads are located within each exclosure.

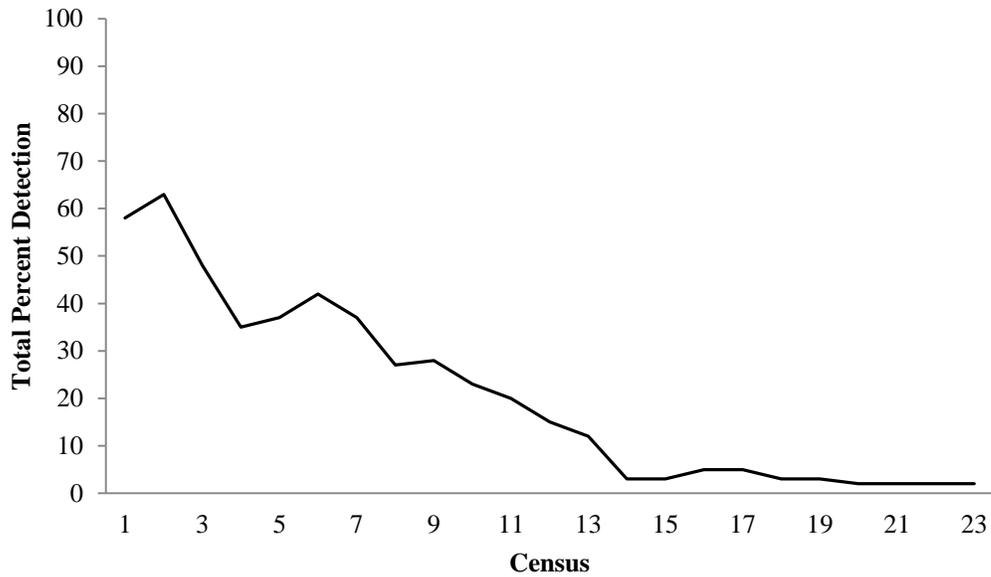


Figure 2. Combined detection over time of adult Houston toads (*Bufo houstonensis*) for all three habitats (juniper, pine and oak). Trial 1 began 11 June 2011 and concluded at census 23 on 25 March 2012.

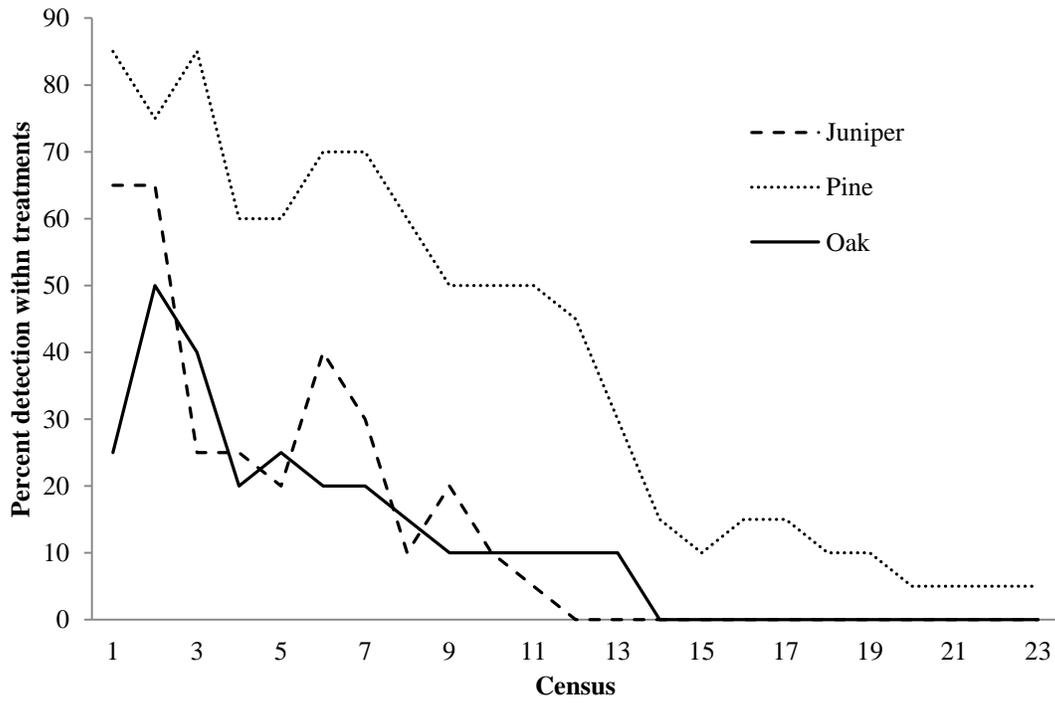


Figure 3. Detection over time of adult Houston toads (*Bufo houstonensis*) within each of the three habitats (juniper, pine and oak). Trial 1 began 11 June 2011 and concluded 25 March 2012.

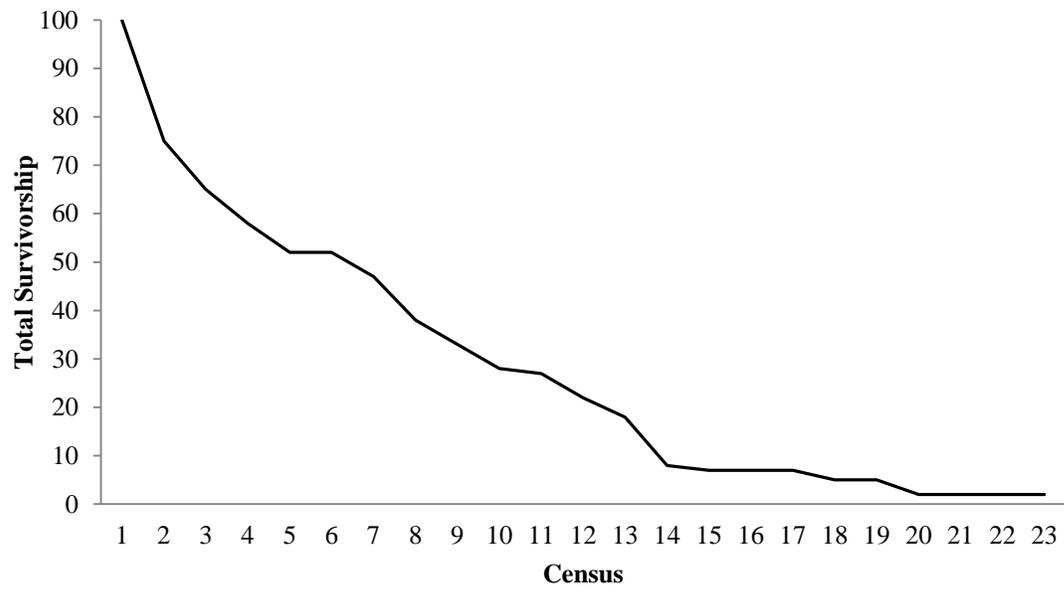


Figure 4. Combined survivorship over time for adult Houston toads (*Bufo houstonensis*) for all habitats (juniper, pine and oak). Trial 1 began 11 June 2011 and concluded 25 March 2012.

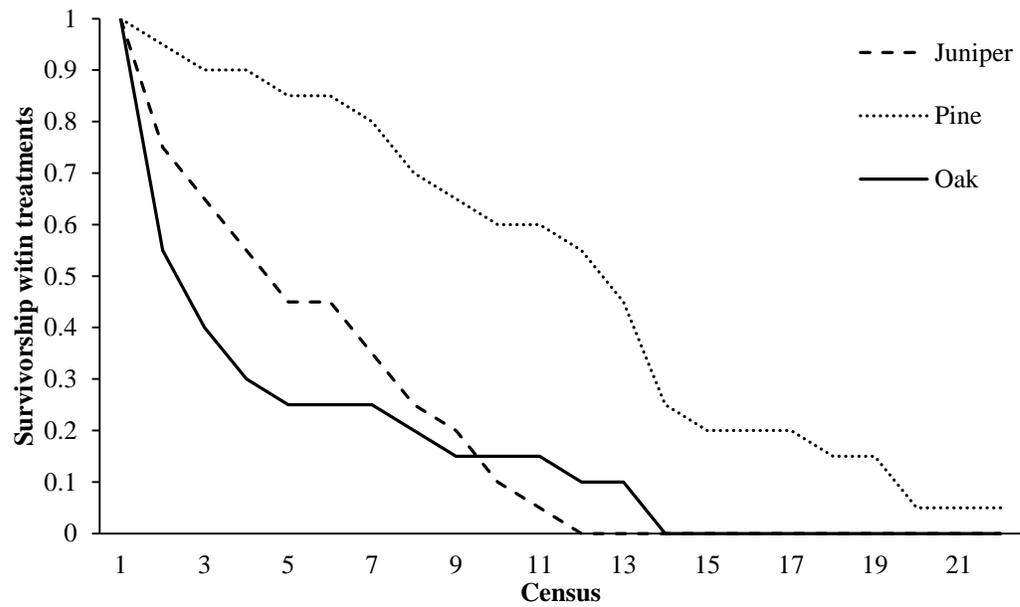


Figure 5. Survivorship over time for adult Houston toads (*Bufo houstonensis*) within all three habitats (juniper, pine and oak). Trial 1 began 11 June 2011 and concluded 25 March 2012.

Table 1. Cormack-Jolly-Seber candidate models and model selection results for trial 1 used for estimating ϕ and p of 60 adult male Houston toads (*Bufo houstonensis*) from a habitat suitability enclosure experiment. Models are listed by most supported to least supported based on AIC_c scores. t represents time-specific estimates (one estimate available for each sampling period), \bullet indicates estimates were constant across time, ht is a covariate representing the habitat type of the treatment (juniper, pine and oak).

Model	AIC_c	ΔAIC_c	AIC_c Weight	Likelihood	# Par.	Deviance
$\phi(ht)p(ht)$	723.21	0.00	0.97	1.00	6	402.12
$\phi(ht)p(\bullet)$	730.26	7.05	0.29	0.03	4	413.30
$\phi(\bullet)p(\bullet)$	738.80	15.58	0.00	0.00	2	425.92
$\phi(t)p(ht)$	750.74	27.52	0.00	0.00	26	385.59
$\phi(t)p(\bullet)$	761.08	37.86	0.00	0.00	24	400.58
$\phi(t^*ht)p(\bullet)$	783.33	60.12	0.00	0.00	50	357.65
$\phi(t^*ht)p(ht)$	807.50	84.29	0.00	0.00	63	344.84
$\phi(t^*ht)p(t^*ht)$	1038.2	315.04	0.00	0.00	135	290.97

Table 2. Trial 1 estimates for ϕ and (p) in adult Houston toads (*Bufo houstonensis*) based on the model supported by the AIC_c selection criterion using program MARK. The model selected was $\phi_{ht} p_{ht}$ where ϕ and p varied with habitats. Lower and upper confidence intervals (CI) are reported.

Estimator	Estimate	Standard Error	Lower CI	Upper CI
Survivorship (ϕ)				
Juniper	0.843	0.032	0.769	0.898
Pine	0.921	0.017	0.88	0.949
Oak	0.786	0.043	0.69	0.859
Recapture (p)				
Juniper	0.600	0.053	0.494	0.699
Pine	0.796	0.028	0.734	0.845
Oak	0.720	0.061	0.587	0.822

Table 3. Total number of Red Imported Fire Ants (*Solenopsis invicta*) found within each of the habitat replicates in trial 1 adult Houston toad (*Bufo houstonensis*) enclosure study in Bastrop County, Texas. Each replicate was baited at nine points and ants were collected after 45 minutes. We compared these totals among the three habitats and toad mortalities.

Habitat	<i>Solenopsis invicta</i>	Total per habitat
Juniper 1	0	
Juniper 2	75	
Juniper 3	0	
Juniper 4	379	
Juniper 5	204	658
Oak 1	0	
Oak 2	0	
Oak 3	75	
Oak 4	0	
Oak 5	0	75
Pine 1	0	
Pine 2	167	
Pine 3	43	
Pine 4	424	
Pine 5	1263	1,897

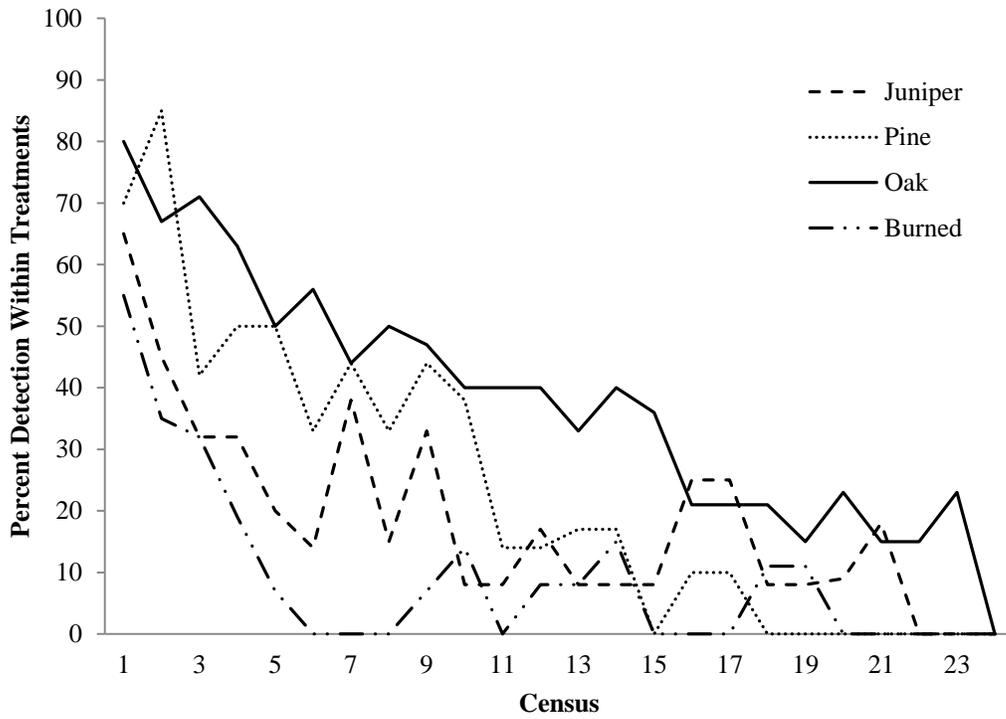


Figure 6. Detection over time of adult Houston toads (*Bufo houstonensis*) among all four habitats during trial 2 (juniper, pine, oak and burned). Trial 2 began March 2013 and concluded March 2014.

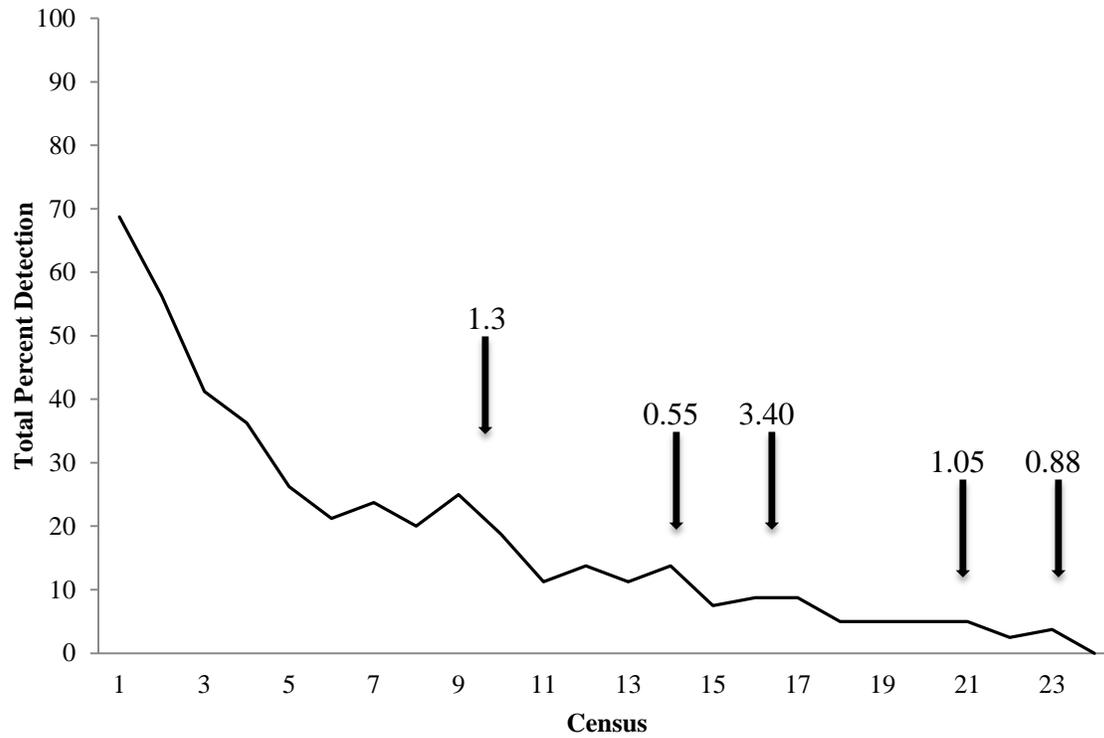


Figure 7. Combined detection over time of adult Houston toads (*Bufo houstonensis*) for all four habitats in trial 2 (juniper, pine, oak and burned). Arrows represent rain amounts from rain events that dropped greater than 0.5 inches of rain in one rain event. Trial 2 began March 2013 and concluded March 2014.

Table 4. Cormack-Jolly-Seber candidate models and model selection results for males and female adult Houston toads (*Bufo houstonensis*) during trial 2 used for estimating ϕ and p from a habitat suitability enclosure experiment. Models are listed by most supported to least supported based on AIC_c scores. t represents time-specific estimates (one estimate available for each sampling period), (\cdot) indicates estimates were constant across time, s is a covariate representing sex.

Model	AICc	$\Delta AICc$	AICc Weight	Likelihood	# Par.	Deviance
$\phi(\cdot)p(\cdot)$	1078.04	0.00	1.00	1.00	2	597.80
$\phi(t)p(\cdot)$	1094.61	16.57	0.00	0.00	25	565.22
$\phi(s)p(t)$	1098.41	20.37	0.00	0.00	26	566.76
$\phi(t)p(t)$	1128.67	50.63	0.00	0.00	48	544.27
$\phi(s)p(s^*t)$	1130.59	52.55	0.00	0.00	50	541.10
$\phi(s^*t)p(\cdot)$	1136.18	58.14	0.00	0.00	49	549.23
$\phi(t)p(s^*t)$	1167.64	89.58	0.00	0.00	72	518.37
$\phi(s^*t)p(t)$	1177.01	98.96	0.00	0.00	72	527.75
$\phi(s^*t)p(s^*t)$	1223.30	145.25	0.00	0.00	95	503.28

Table 5. Trial 2 estimates with males and females for ϕ and (p) in adult Houston toads (*Bufo houstonensis*) based on the model supported by the AIC_c selection criterion using program MARK using the Cormack-Jolly-Seber mark-recapture model. The model selected was $\phi_{(.)}p_{(.)}$ where ϕ and p did not vary among time or sex. Lower and upper confidence intervals (CI) are reported.

Estimator	Estimate	Standard Error	Lower CI	Upper CI
Survivorship (ϕ)				
Males and Females	0.88	0.012	0.852	0.903
Recapture (p)				
Males and Females	0.62	0.022	0.579	0.661

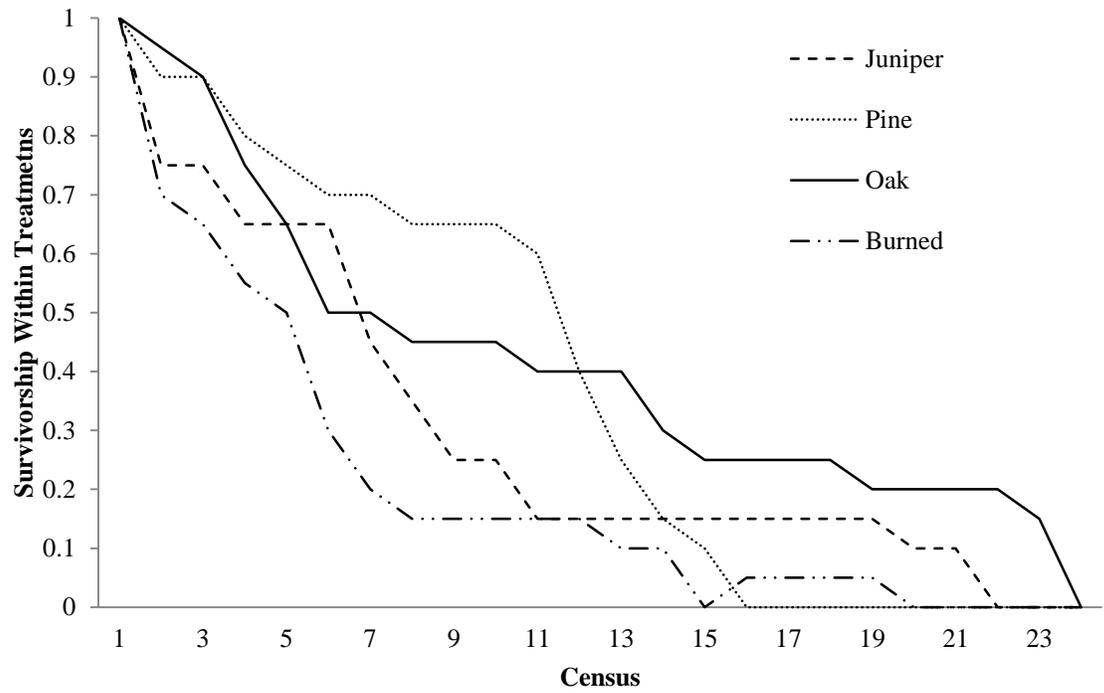


Figure 8: Percent survivorship over time of adult Houston toads (*Bufo houstonensis*) within all four habitats during trial 2 (juniper, pine, oak and burned). Trial 2 began March 2013 and concluded March 2014.

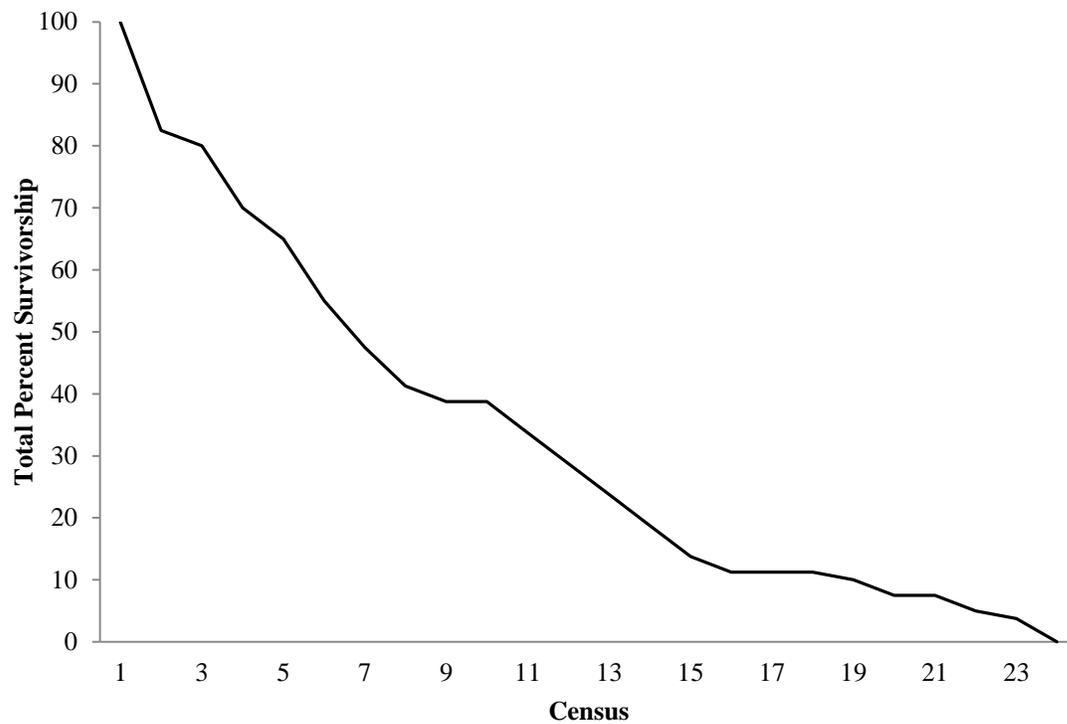


Figure 9. Combined survivorship over time of adult Houston toads (*Bufo houstonensis*) for all four habitats during trial 2 (juniper, pine, oak and burned). Trial 2 began March 2013 and concluded March 2014.

Table 6. Candidate models and model selection results for trial 2 used for estimating ϕ and p of 80 adult Houston toads (*Bufo houstonensis*) from an habitat-suitability enclosure experiment. Models are listed by most supported to least supported based on AIC_c scores. t represents time-specific estimates (one estimate available for each sampling period), \bullet indicates estimates were constant across time, ht is a covariate representing the habitat type of the treatment (juniper, pine, oak, and burned).

Model	AIC_c	ΔAIC_c	AIC_c Weight	Likelihood	# Par.	Deviance
1. $\phi(ht)p(ht)$	1040.43	0.00	1.00	1.00	8	626.79
2. $\phi(t)p(ht)$	1055.39	14.96	0.00	0.00	28	598.09
3. $\phi(ht)p(\bullet)$	1077.15	36.72	0.00	0.00	5	669.71
4. $\phi(\bullet)p(\bullet)$	1078.04	37.61	0.00	0.00	2	676.71
5. $\phi(t)p(\bullet)$	1094.61	54.18	0.00	0.00	25	644.13
6. $\phi(t*\bullet)p(ht)$	1182.38	141.95	0.00	0.00	99	527.97
7. $\phi(t*\bullet)p(\bullet)$	1209.55	169.12	0.00	0.00	95	568.44
8. $\phi(t*\bullet)p(t*\bullet)$	1524.50	484.07	0.00	0.00	191	443.37

Table 7. Trial 2 estimates for ϕ and (p) in adult Houston toads (*Bufo houstonensis*) based on the model supported by the AIC_c selection criterion using program MARK using the Cormack-Jolly-Seber mark-recapture model. The model selected was $\phi_{ht} p_{ht}$ where ϕ and p varied with habitats. Lower and upper confidence intervals (CI) are reported.

Estimator	Estimate	Standard Error	Lower CI	Upper CI
Survivorship (ϕ)				
Juniper	0.870	0.028	0.803	0.916
Pine	0.888	0.024	0.832	0.927
Oak	0.914	0.02	0.866	0.946
Burned	0.841	0.035	0.76	0.899
Recapture (p)				
Juniper	0.537	0.049	0.44	0.631
Pine	0.645	0.041	0.56	0.721
Oak	0.777	0.032	0.707	0.834
Burned	0.351	0.056	0.251	0.466

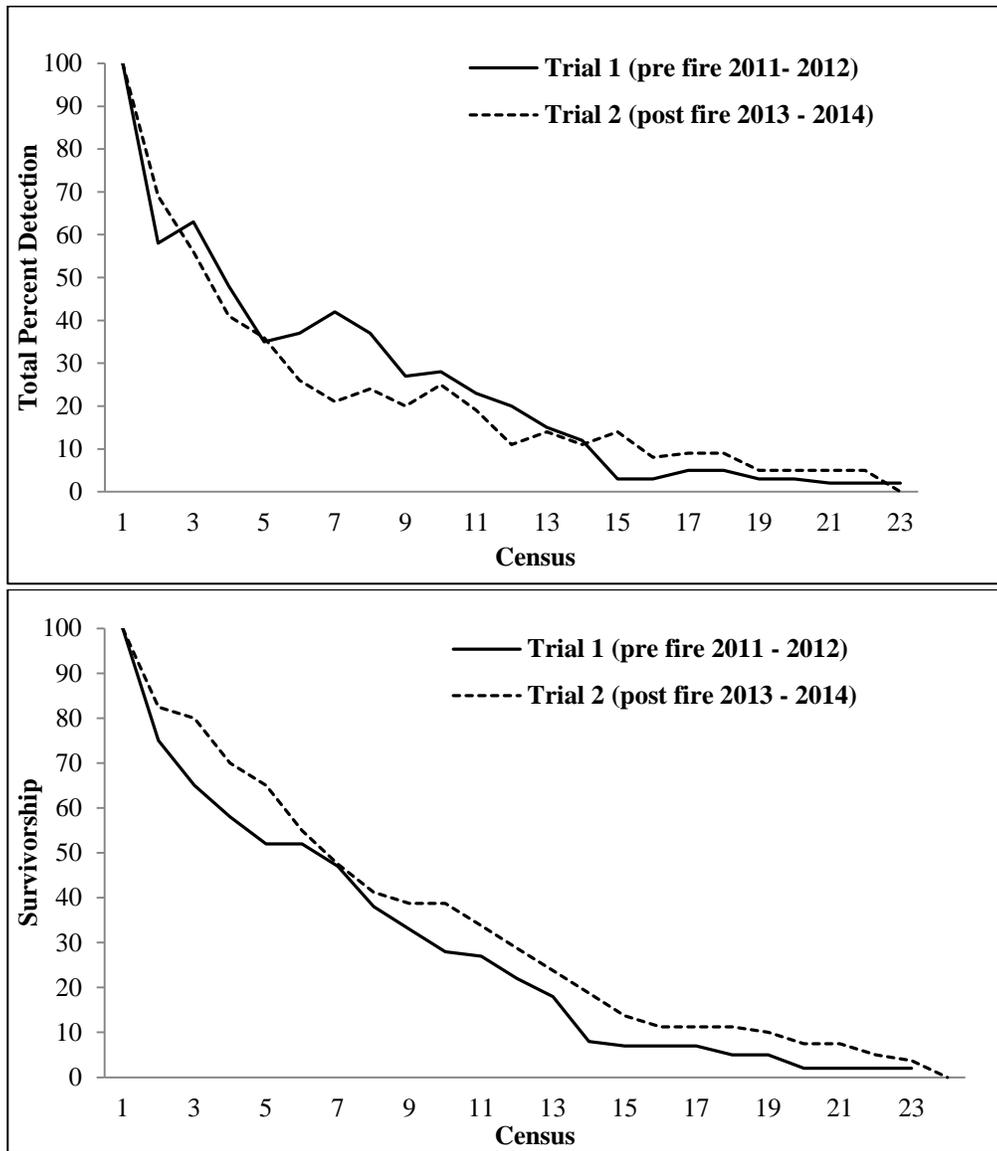


Figure 10. Houston toad (*Bufo houstonensis*) total percent detection and survivorship comparison between trial 1 (2011 – 2012) (top) and trial 2 (2013 – 2014) (bottom). Detection and survivorship trends are comparable across time during the two trials.



Figure 11. Photo of an adult male Houston toad (*Bufo houstonensis*) from trial 1 during the drought of 2011. This individual was found desiccated as it emerges from its shallow burrow. This toad was from pine habitat.

TESTING DENSITY DEPENDENCE IN JUVENILE HOUSTON TOADS (*BUFO HOUSTONENSIS*) IN BASTROP COUNTY, TEXAS

Introduction – Amphibian declines are continuing to accelerate globally. This is in part due to habitat loss, disease, agriculture practices, invasive species, drought, and wildfire. Over the past several years, there has been an increase in the global loss of biodiversity (Griffith et al., 1989). Therefore population supplementation practices such as captive-breeding, head-starting, and translocation programs have increased in necessity (Dodd and Seigel, 1991).

In order to implement effective conservation management practices, we must be able to determine which life stage is the most effective to use in offsetting these declines. Many studies have looked at various life stages to determine which stage is having the most effect on global amphibian declines. It has been hypothesized that mortality at the egg stage could be the leading factor for the continued declines. Vonesh and De la Cruz (2002) tested egg and juvenile mortality within *Bufo* and concluded that mortality occurring at the juvenile life stage may have a greater impact on amphibian declines rather than embryonic life stages. Berven (2009) reported that juvenile population size of the wood frog (*Rana sylvatica*) was the most important factor that impacted juvenile survivorship alongside the age of female at first reproduction. Harper and Semlitsch (2007) showed juvenile density had strong negative effects on survival, growth and reproduction of the American toad (*Bufo americanus*).

Research on density dependence during juvenile terrestrial life stages is still relatively rare, especially research focusing on endangered amphibians. When species populations are low, density dependence may not be a concern because we assume larval or terrestrial densities will not be large enough to have a negative effect. Determining which life stage

drives population regulation can be helpful in developing new or increasing the effectiveness of conservation management strategies.

Houston toad head-starting efforts have focused on the release of thousands of tadpoles, metamorphs (Vandewege et al., 2011) and now eggs onto the landscape onto recently extirpated or current Houston toad locations. Adult toads have been monitored via radio telemetry (Forstner and Swannack, 2004) pit tag, and toe clips (Brown et al., 2011). It is uncertain which Houston toad life stage is most vital to growth rate and species survival. Studies have shown that pre-metamorphic densities have little impact on life factors such as survivorship and growth rates therefore the critical life stage in question may be metamorph and/or juvenile stages. Metamorph or juvenile survivorship and growth rates of the Houston toad have yet been significantly tested.

Preliminary data from the adult toad enclosure experiment was used to determine which habitat (juniper, pine or oak) was the most suitable for Houston toads. Before we can eventually test habitat suitability post catastrophic wildfire, we must determine the optimal juvenile dispersal density for emerging metamorphs. Finding the optimal dispersal density will enable us to eliminate survivorship variables for a post catastrophic fire survivorship study.

Houston toad dispersal of a 50 m radius of the natal pond up to 13 weeks post emergence has been reported in Greuter (2004) and has been used to develop a buffer zone for habitat management. Density of individuals during dispersal and along with conspecifics across a landscape can alter and affect growth rates, resource competition, survival and reproduction (Harper and Semlitsch, 2007). For the Houston toad, conservation management practices have been implemented at various life stages with varying successes.

We are conducting direct assessments of habitat restoration options following the aftermath and recovery efforts from the Bastrop County Complex Fire using juvenile Houston toad enclosure experiments. These enclosures are being applied to assess the density of juveniles required to evaluate future juvenile head-start releases. The density enclosures are within an unburned pine dominated 80% or greater canopy cover habitat. This habitat was chosen based on the adult enclosure study discussed in the previous chapter. These densities are needed to better guide metamorph releases and to enable the eventual repeat of habitat suitability testing for juvenile Houston toads. Once these optimum densities are determined we will also have the ability to test these densities in burned habitats retaining limited canopy with approximately 40% canopy and catastrophically burned habitats with 10% or less canopy cover thus representing 40% of current Houston toad habitat in Bastrop County.

Quantitatively estimating demographic parameters from mark-recapture (henceforth MR) studies have advanced considerably over the last three decades (Lebreton and Pradel, 1992; Burnham and Anderson, 2002). Currently, most MR studies use multi-model analysis in information-theoretic framework to estimate survival (ϕ) and the probability of recapture (p) (Burnham and Anderson 2002, Schmidt et al., 2002). Statistical inference from model selection under an information-theoretic approach requires rigorous attention to selecting the candidate set of models. Briefly, a candidate model set is developed using a priori hypotheses focusing on the relationship between survival and recapture, and covariates, such as treatment effects, environmental parameters, among others. Models are ranked based a selection criterion, most commonly Akaike's information criterion (AIC_c) (Akaike, 1973),

which provides a reliable decision criterion for model selection for both nested and non-nested models (Schmidt and Anholt, 1999; Burnham and Anderson, 2002).

This study used an information-theoretic approach to model selection to choose models that best fit MR datasets collected from a juvenile exclosure experiment conducted in 2014 – 2015 where juvenile Houston toads were released into outdoor exclosures at six different densities. For each model ϕ and p were estimated.

Materials and Methods – Exclosures for juveniles were 2 x 1 m² constructed of 1/8th inch hardware cloth, with covers made from bird netting (preventing immediate bird predation and tree debris in falls) (Figure 1). The exclosures were buried 20 cm deep with walls extending 50 cm above ground. A 10 cm lip was folded along the top and bottom of each pen to prevent toads from tunneling out of the exclosure and prevent toads from scaling the hardware cloth walls and escaping. Each exclosure contained ground cover and woody debris to offer shade and two Tupperware bowl reservoirs filled with sphagnum moss and water to supplement hydration to each exclosure. Moss reservoirs were filled as needed to prevent toad desiccation.

Twenty-seven exclosures were loaded with juvenile Houston toads on August 18th 2014. Densities used for this experiment are 2, 4, 5, 6, 9, and 12 juvenile Houston toads per exclosure. Juvenile Houston toad availability for the study influenced the total number of replicates possible. Replicates for each density were four replicates (density of 2), seven replicates (density of 4), five replicates (density of 5), six replicates (density of 6), three replicates (density of 9) and two replicates (density of 12). A total of 148 juvenile Houston toads were used in this experiment. Upon release each toad was measured (snout urostyle length (SUL) and head width (HW)), weight recorded, and given an individual toe clip

number for easy identification. Average mass and SUL for enclosure toads was 1.15 g and 19.2 mm SUL. Houston toad juveniles weighed 0.25 – 3.0 g (mean = 1.16; SD = 0.5019) and SUL length was 9.4 – 26.3 mm (mean = 19.26; SD 2.9840). Houston toad juveniles were assigned to the 27 enclosures and differences were seen among starting mass and SUL of individuals among the six densities (Mass: ANOVA, $df = 5, 21$, $F = 2.434$, $p = 0.037$) (SUL: ANOVA, $df = 5, 21$, $F = 3.479$, $p < .005$). The five density enclosures had significantly larger juveniles at the start of this study, however did not affect the overall growth outcome for this study. This study began on August 18th 2014 and concluded March 25th 2015. Initially the enclosures were visited once a week in order to document any initial decrease in detection. Once detection had stabilized, enclosures were visited once every two weeks. During each census, SUL, and mass were taken. A total of 13 censuses occurred during this study.

Drought is a common concern for the survivorship of Houston toads. Before juveniles were released into the enclosure, we simulated a rain event in order to increase soil moisture levels. Each enclosure received an initial 15 gallon treatment of water to simulate a two inch rain event. Enclosures were rehydrated daily using a three gallon pump sprayer at dusk with the intent to slow down instant evaporation associated with Texas summer days. Enclosures were checked each evening on a three day rotation of nine enclosures each day. Data was collected for each enclosure once a week until detection stabilized. Enclosures were then checked bi-weekly.

Analysis of Survivorship – Capture probabilities for each census were calculated in the program MARK (White and Burnham, 1999) using a Cormack-Jolly-Seber (Cormack, 1964; Jolly, 1965; Seber, 1965) model to assess the accuracy of our censuses. For this model

all juvenile toads captured in subsequent censuses were known to be alive in all previous census no matter if the individual was not detected. We compared survivorship based on stocking densities then compared to final densities among all six densities. We assumed that capture probability was at 100% at the conclusion of this study.

Model selection procedure in MARK – Using a model selection approach based on information-theoretic methods, Program MARK (White and Burnham, 1999) was used to estimate the probabilities of ϕ and p for juvenile Houston toads. Program MARK methods followed Cooch and White (2006). Two explanatory factors were used to explore variation in ϕ and p : time and density. Time was considered as constant among sampling periods (\bullet) or variable across periods. Habitat type (d) was treated as a categorical covariate with six densities (2, 4, 5, 6, 9, and 12) and used to determine if density affected ϕ or p . Based on these factors, six candidate models were developed, where each model represented a different biologically-based hypothesis that explored the effects of time and density on estimates of ϕ and p . For example, $\phi_t p_t$ represented the Cormack-Jolly-Seber model (CJS) that is fully time dependent for both ϕ and p . Whereas $\phi_d p.$ represented a model where survivorship varied among density, and p remained constant among sampling periods.

The amount of support for each of the six candidate models was evaluated using a correction factor for AIC (AIC_c) which protects against over-fitting the models, especially with small sample sizes (Hurvich and Tsai, 1989). The model with the lowest AIC_c was considered to best fit the data unless the difference in AIC_c values (ΔAIC_c) among competing models was < 2.0 , then the models were considered indistinguishable. Models were ranked from one to six, with one being the best supported model and eight being the least. If multiple models supported the data, the most parsimonious model was chosen as the best supporting models. Point

estimates, standard errors and 95% confidence intervals were recorded for ϕ and p for each model.

Analysis of Growth – To analyze growth we used a linear mixed effects model, repeated measures analysis of variance (ANOVA) to determine differences in SUL among all density treatments overtime. Only exclosures that contained at least one detection each census were used in these calculations. At the close of this study, juvenile Houston toads were not detected in any of the 12 density exclosures. Therefore we ran a repeated measures ANOVA using only data from the five densities that were represented at the conclusion of the study (densities 2, 4, 5, 6, and 9). Fixed factors were density and time with exclosure as the random factor.

Soil moisture monitoring – Soil moisture was monitored and recorded for four months (October – January). Ten Decagon Devices EC-5 soil moisture meters were evenly placed among the 27 exclosures. These soil meters measure the volumetric water content (m^3/m^3 VWC) of the soil and have a ~0.2 L measurement volume. Each meter is placed in the center of the exclosure, approximately 10 cm below the surface of the soil. Data from each soil meter was sent to a Decagon Devices Em50 data logger via 15 m extension cables. Soil moisture was set to record at 6 am, 12 noon, and 6 pm each day for four months. Soil moisture was then averaged daily and graphed. Data logger batteries were replaced once every three weeks.

Results

Juvenile Survivorship – One hundred forty seven juvenile Houston toads were released into 27 exclosures on August 18th 2014 and were monitored until March 25th 2015. Upon completion 46 toads (31%) were detected throughout the entire study. Five out of the six density treatments had toads survive throughout the entire study. Exclosures containing

12 juvenile toads failed to provide a single toad detection after October 2014 (census 7). Average density per enclosure was highest in densities four, five, six and nine (Figure 2). Therefore only enclosures with densities of 2, 4, 5, 6 and 9 had individuals that could overwinter. There was a dramatic decrease of total juvenile survivorship during the first two months of the study (Figure 3). This is most likely due to initial stresses involved with transporting toads and then initial acclimation of the new environment. Survivorship hit a plateau just prior to overwintering (November – February). Individuals who survived up to the onset of freezing temperatures were successful in overwintering and surviving until the spring season. Toads in the highest density enclosures experienced the greatest mortality with 0% surviving in enclosures that held 12 Houston toads. When comparing survivorship among the six densities, survivorship was the highest in enclosures containing four juvenile Houston toads (Figure 4).

Based on ΔAIC_c , model 1 ($\phi_d p$) was the best supported model (Table 1). For this model, ϕ varied by density but not across time and recapture was constant (Table 2). Survivorship was highest in enclosures with five juveniles ($\phi = 0.93$), followed closely by densities of six ($\phi = 0.92$) and four ($\phi = 0.91$) juvenile toads per enclosure. Recapture (p) for densities 4, 5 and 6 toads per census was $p = 0.91$, $p = 0.87$, and $p = 0.92$ respectively. The highest recapture estimate for each census were the 12 density enclosures ($p = 0.98$). Model 2 was closely comparable with Model 1, however we chose to select Model 1 due to the high standard deviance seen in Model 2. The other 4 models had ΔAIC_c values > 2.0 , which indicates that density had a stronger effect on the data compared to the most parsimonious model (Model 3, $\phi \cdot p$).

Growth Analysis – Growth rates were significantly reduced in the 12 density enclosures by census 7 (ANOVA: $df = 6, 19, F = 4.1003; p = 0.0167$). During the last census, growth rates were not significantly different among the remaining five densities (ANOVA: df

= 4, 21, $F= 2.3894$; $p= 0.1044$) (Table 3). Toad SUL and mass was measured during each census. Average SUL at the start of the study was 19.23 mm and average SUL at the end of the study was 25.67 mm. Average mass at the start of the study was 1.15 grams and 1.91 grams at the end of the study (Figure 5) resulting in a positive linear regression correlation among total SUL and mass over time. All but density 12 exclosures saw an increase in SUL and mass throughout the study. Individuals in the 12 toad density exclosures lost mass overtime and survivorship hit 0% by census 7.

Soil moisture data – Soil moisture was measured from October 2014 to January 2015. Soil moisture was positively correlated with rain events (Figure 6). Juvenile toad detection did not increase following large rain events (Figure 7).

Discussion – The effects of density on survivorship and recapture of juvenile *B. houstonensis* were significant with differences in survivorship and recapture estimates between densities but not overtime. Recapture rates were constant among habitats and time. Survivorship estimates were highest in density 5, followed closely by densities of 6 and 4 toads per exclosure. Survivorship was lowest in exclosures containing 12 juvenile toads ($\phi = 0.73$). Detection and survivorship trend data supports the selection of this model.

Recapture estimates were highest in the 12 and 2 density exclosures, both showing the lowest survivorship estimates. By census 7 toads in the 12 density exclosures were no longer detected. Recapture estimates are high because it is reporting the probability of recapturing an individual each census. If toads are no longer being detected then estimates report a high probability that your recapture rate of 0 will occur.

Growth was not significant among densities during this study. Trend data shows that 12 density exclosures were the only density to see a reduction of mass overtime. This high

density was excluded from the repeated measures ANOVA due to lack of detection post census 7. It can be inferred from trend data that significant differences may have occurred if these data were included. Succinctly, the results indicate releasing at lower densities appears to favor the overall health and success of juvenile Houston toads.

Conditions in the exclosures were hospitable, with consistent shade and moisture. Artificially watering was not needed after census 4. Rainfall amounts were enough to maintain moisture within the exclosures. On two occasions large *B. nebulifer* were found within an exclosures. Although removed, there was no detection of juvenile Houston toads in those exclosures. This occurred at census 2 in both occasions and predation by congeneric is likely the cause of detection loss. Unlike the adult exclosure studies, we have little evidence of red imported fire ants present in these exclosures. On occasion ants were seen in the exclosures, however were not exhibiting aggressive mound behavior. Bird netting was effective in keeping birds from preying on the toads, and kept hog nose snakes (*Heterodon platyrhinos*) from entering the exclosures.

Overwintering was not a period of high mortality as seen in Harper and Semlitsch, 2007 with *B. americanus* (American toad). Prior to overwintering total detection stabilized and maintained between 30% and 40% detection until the close of the study. This suggests that at these densities, individuals were not competing for resources and therefore able to maintain body condition before overwinter estivation.

Similar, but not identical stocking densities and replicates were seen in Harper and Semlitsch (2007), testing American toad densities. Due to difficulties in acquiring juvenile Houston toads for this experiment we could not mimic the breadth of their American toad stocking densities. Similarly, our limitations resulted in fewer replicates and could have

influenced our results. To best utilize the stocking resources we had available we drew from the results from Harper and Semlitsch (2007) which demonstrated survivorship and growth to be highest in lower stocking densities. Consequently, we reduced the number of lowest and highest stocking densities, focusing on the proposed optimum densities seen in this study. It would be optimum to replicate this study and increase in replicates for comparison, but this may not be practical given the limitations on juvenile availability for such an experiment.

There was no recovery of any dead juvenile toads during this study making it unclear what proximate causes contributed to the loss of detection for 69% of the juvenile toads. The initial decreases observed in overall detection is likely due to stresses involved in acclimation and time of year of release. These individuals were released in August, one of the hottest months of the year. Luckily, large rain events occurred often and very likely decreased the mortality by desiccation.

Juvenile Houston toads remained actively above ground from August 2014 to March 2015. Rain events did not have an affect on juvenile toad movement as seen in adult Houston toads. We observed juvenile toads above ground throughout the day but as temperatures would rise during the late morning hours, toads were more often observed taking shelter under the provided structure in or near the water pools provided. Toad movement would then increase in the late afternoon hours. Only when temperatures dropped below 0° C did we observe a decrease in above ground movement within the exclosures. During these freezing temperatures toads were found below the provided structure and many were found tucked up under the water pools. Very few toads were observed actually subsurface into the sandy loam soils even during these freeze events. Therefore little evidence that support juvenile Houston toads spend any considerable amount of time below

the soil surface or implement estivation behavior during summer and winter months as do the adults.

If density dependence is regulated at the juvenile life stage for the Houston toad then focusing efforts in improving habitat specifically for Houston toad juveniles could potentially increase survivorship at this level (Halpern et al., 2005). Determining the optimal habitat can be a huge help in future conservation strategies for the Houston toad and amphibians in Bastrop County. We need to focus our release efforts in areas that are conducive to housing this rare amphibian. Head-starting has been our key conservation strategy for the Houston toad. We are now one step closer in maximizing the efficiency and efficacy of head-starting Houston toads.

From these results we can continue to optimize our management practices for this endangered species. We know that at high densities these individuals will not thrive, however stocking densities are not confined to a narrow or specific number of individuals. It can be assumed that current populations will not reach such high densities naturally, therefore this knowledge can be used to guide future head-starting or captive propagated releases.

Above ground activity can be an important factor in juvenile Houston toad ecology. Juvenile amphibian activity increases once individuals emerge and move out onto the landscape. Dispersal across the landscape allows juveniles to seek out upland habitats or undergo inter-pond dispersal. Therefore this need to disperse would increase activity for juvenile individuals. This study gave us the first opportunity to observe juvenile toads post one year since hatching as tadpoles. During this study there was a steady increase in overall juvenile SUL and mass overtime. From this study we conclude that Houston toad juveniles are above ground and active during all months of the year. This should have major

implications on future conservation management practices and plans conducted in Bastrop County.

Conservation and management practices implemented for the Houston toad in Bastrop County have strongly been influenced by data gathered from monitoring adult populations. Few studies have discussed Houston toad emergence behavior (Greuter 2004) or juvenile dispersal (Vandewege, 2013). Understanding juvenile ecology and behavior post emergence to one year has not been documented until now. Differences in adult and juvenile Houston toad ecology are significant and have major management implications.

In the aftermath of the Bastrop County Complex fire, clean-up operations were conducted in cooperation with USFWS in order to decrease or prevent activities that would lead to “take”. Many of these operations were conducted year round in Houston toad habitat. Monitoring for the Houston toad during these operations was most intense during Houston toad breeding season, continuing a few months into the summer capture emergence and then movement of metamorphs. From this study we conclude that juvenile Houston toads are consistently moving across the landscape all months of the year. If density dependence is regulated at the juvenile life stage in Houston toads, management practices need to shift to incorporate avoidance measures for not only breeding adults, but movement and habitat use by juveniles dispersing across the landscape after the initial pond emergence event.

We will be able to use these data collected from this study to look at optimal juvenile densities across various habitats in Bastrop County. As Bastrop County is still undergoing continual habitat management post wildfire, these data will continue to aid in our efforts to manage the habitat and population of this endangered amphibian.

Juvenile Survivorship Within Three Habitat Treatments

Optimum density data collected from the juvenile density study provided the framework for testing juvenile Houston toad survivorship in three different habitat types in Bastrop County. This second study will complete in the fall of 2015 or possibly the spring of 2016 dependent on continuing detections, therefore only results for the first six months can be reported here.

Materials and Methods – Sixty juvenile exclosures were constructed for this habitat study. Three habitat treatments were selected (burned, 30% cover, and control) with 20 exclosures placed within each treatment. Three juvenile Houston toads were placed in each exclosure for a total of 60 toads per treatment (See page 44 for exclosure specifications). Each exclosure contained ground cover and woody debris to offer shade and two Tupperware bowl reservoirs filled with sphagnum moss and water to supplement hydration to each exclosure. Moss reservoirs were filled as needed to prevent toad desiccation.

A total of 120 juvenile Houston toads were used in this experiment. Upon release each toad was measured (snout urostyle length (SUL) and head width (HW)), weight recorded, and given an individual toe clip number for easy identification. Average mass and SUL for exclosure toads was 0.589 g and 17.86 mm SUL. Houston toad juveniles were assigned to one of the three treatments and a week difference was seen among starting SUL of individuals among the three treatments (SUL: ANOVA, $df = 2$, $F = 3.3042$, $p = 0.0485$). This study began on March 30th, 2015 and will run until 2016. Initially the exclosures were visited once a week in order to document any initial decrease in detection. Once detection had stabilized, exclosures were visited once every two weeks. During each census, SUL, and mass were taken. Preliminary results are from 11 census events in 2015.

Analysis of Survivorship – Capture probabilities for each census were calculated in the program MARK (White and Burnham, 1999) using a Cormack-Jolly-Seber (Cormack, 1964; Jolly, 1965; Seber, 1965) model to assess the accuracy of our censuses. For this model all juvenile toads captured in subsequent censuses were known to be alive in all previous census no matter if the individual was not detected. We assumed that capture probability was consistent at 100%.

Model selection procedure in MARK – Using a model selection approach based on information-theoretic methods, Program MARK (White and Burnham, 1999) was used to estimate the probabilities of ϕ and p for juvenile Houston toads. Program MARK methods followed Cooch and White (2006). Two explanatory factors were used to explore variation in ϕ and p : time and treatment. Time (t) was considered as constant among sampling periods (\bullet) or variable across periods. Treatment type (g) was treated as a categorical covariate with three treatments (burned, 30% cover, control) and used to determine if treatment affected ϕ or p . Based on these factors, 11 candidate models were developed, where each model represented a different biologically-based hypothesis that explored the effects of time and treatment on estimates of ϕ and p .

The amount of support for each of the 11 candidate models was evaluated using a correction factor for AIC (AIC_c) which protects against over-fitting the models, especially with small sample sizes (Hurvich and Tsai, 1989). The model with the lowest AIC_c was considered to best fit the data unless the difference in AIC_c values (ΔAIC_c) among competing models was < 2.0 , then the models were considered indistinguishable. Models were ranked from one to 11, with one being the best supported model and 11 being the least. If multiple models supported the data, the most parsimonious model was chosen as the best supporting models. Point estimates, standard errors and 95% confidence intervals were recorded for ϕ and p for each model.

Analysis of Growth – To analyze growth we used a linear mixed effects model, repeated measures analysis of variance (ANOVA) to determine differences in SUL among all three treatments overtime. Only exclosures that contained at least one detection each census were used in these calculations. Fixed factors were treatment and time with exclosure as the random factor.

Results

Juvenile Survivorship – One hundred twenty juvenile Houston toads were released into 60 exclosures on March 30th, 2015 and will be monitored until 2016. To date, 25 toads (20.8%) were detected during the 11th census event; 7 toads (11.7%) in burned), 7 toads (11.7%) in 30% cover, and 11 toads (18.3%) in the control treatment. When comparing survivorship among the three habitat treatments, survivorship was the highest in the burned treatment, however was not significantly different from the 30% cover and control treatments (Figure 8).

Based on ΔAIC_c , model 1 ($\phi(g)p(t*g)$) was the best supported model (Table 4). For this model, ϕ varied by habitat treatment and p was dependent on time and treatment. Survivorship was highest in the burned exclosures ($\phi = 0.97$), followed closely by the control treatment ($\phi = 0.96$) and then 30% cover ($\phi = 0.90$) (Table 5).

Growth Analysis – Growth rates were significantly different among the three habitat treatments and over time (Table 6) (ANOVA: $df = 2$, $F = 12.153$; $p = <.0001$). Toad SUL and mass was measured during each census. Average SUL at the start of the study was 17.86 mm and average SUL at census 11 was 33.99 mm (Figure 9). Average mass at the start of the study was 0.589 grams and 4.19 grams at census 11.

Survivorship, although higher in the burned habitat treatment, was not significantly different among the three habitat treatments. Snout urostyle length was significantly different

among the three habitat treatments with larger toads being found in the burned treatment. These preliminary results suggest the burned unit (although does not increase survivorship) contains factors that allow toads to grow significantly larger than the 30% cover and control treatments. Visual surveys of the burned treatment reveal a large abundance of termite larvae which is not seen in either the 30% cover and control treatments. The fire altered the fuel load within the burned treatment resulting in a shift in the invertebrate composition, diversity and abundance. Further studies should explicitly seek to compare invertebrate diversity and density among the different habitat treatments to see if these differences are as we speculate which could result in larger individuals in the burned treatment consequent of overall invertebrate composition or abundance.

This study may well be able to continue until the spring of 2016 in order to monitor juvenile overwinter survivorship among the three treatments. Starting September 2015, exclosures will be checked only once a month until spring 2016.



Figure 1. Example of a juvenile Houston toad (*Bufo houstonensis*) wild mesocosm exclosures in Bastrop County, Texas. This represents one of 27 exclosures located in pine habitat in Bastrop County. Toad densities of 2, 4, 5, 6, 9 or 12 toads are located within each exclosure.

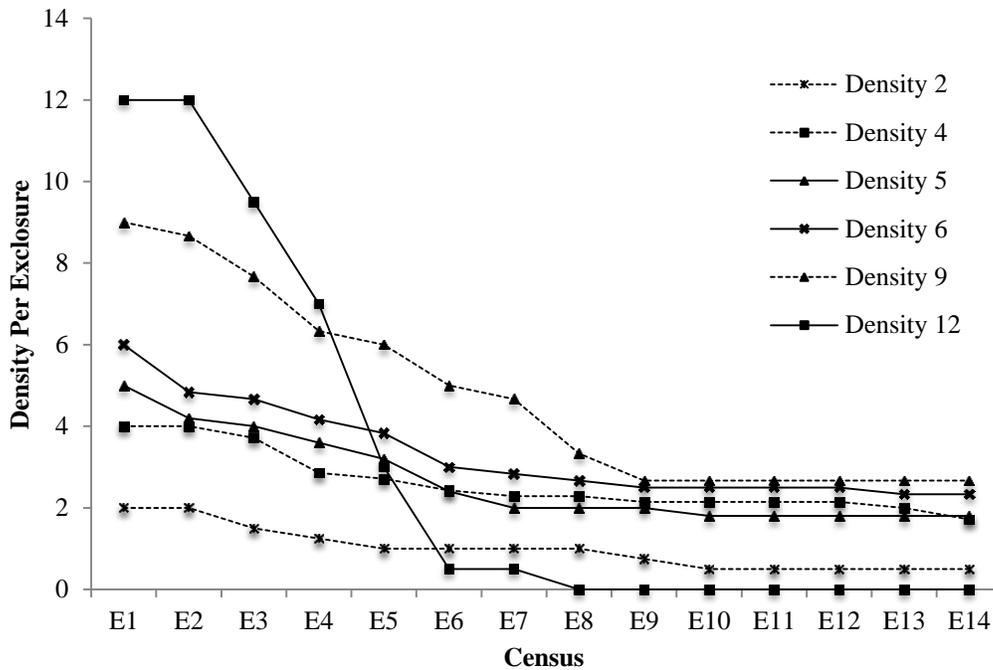


Figure 2. Average density per enclosure of juvenile Houston toads (*Bufo houstonensis*) detected in 26 1 x 2 m² outdoor enclosures in Bastrop County, Texas. Each enclosure contains one of six densities of juvenile Houston toads (2, 4, 5, 6, 9 and 12 individuals per enclosure). Initial release was 15 August 2014 (E1) and final census was 25 March 2015 (E14).

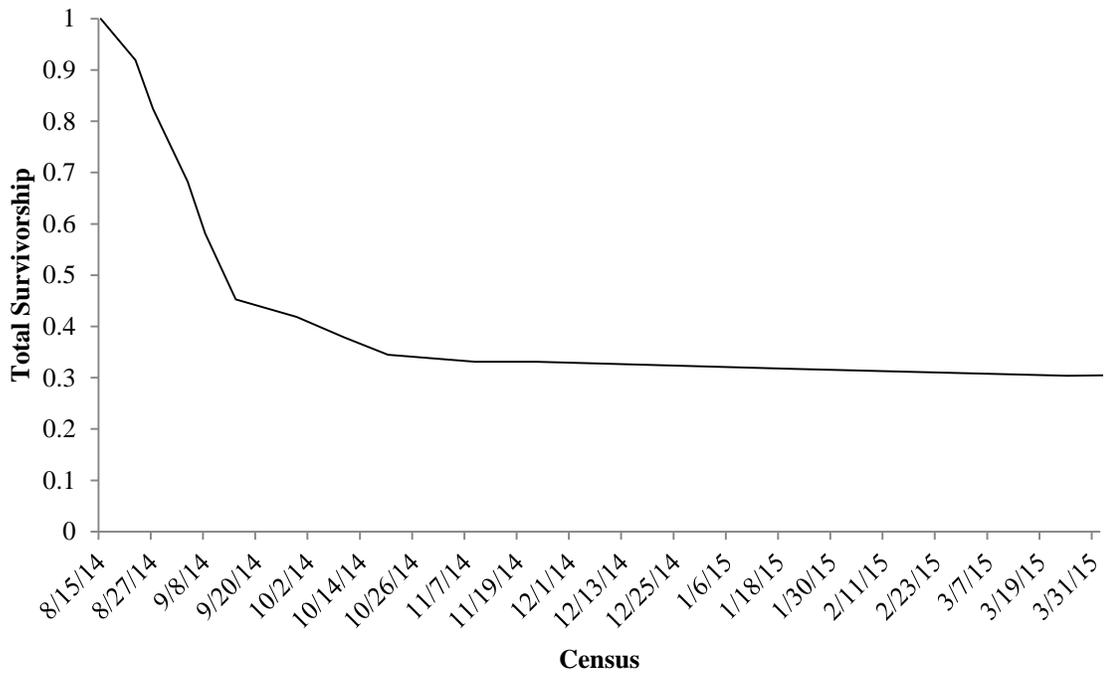


Figure 3. Total survivorship over time of 148 juvenile Houston toads (*Bufo houstonensis*) detected in 26 1 x 2 m² outdoor exclosures. Initial release of juvenile Houston toads was 15 August 2014 and final census was 25 March 2015

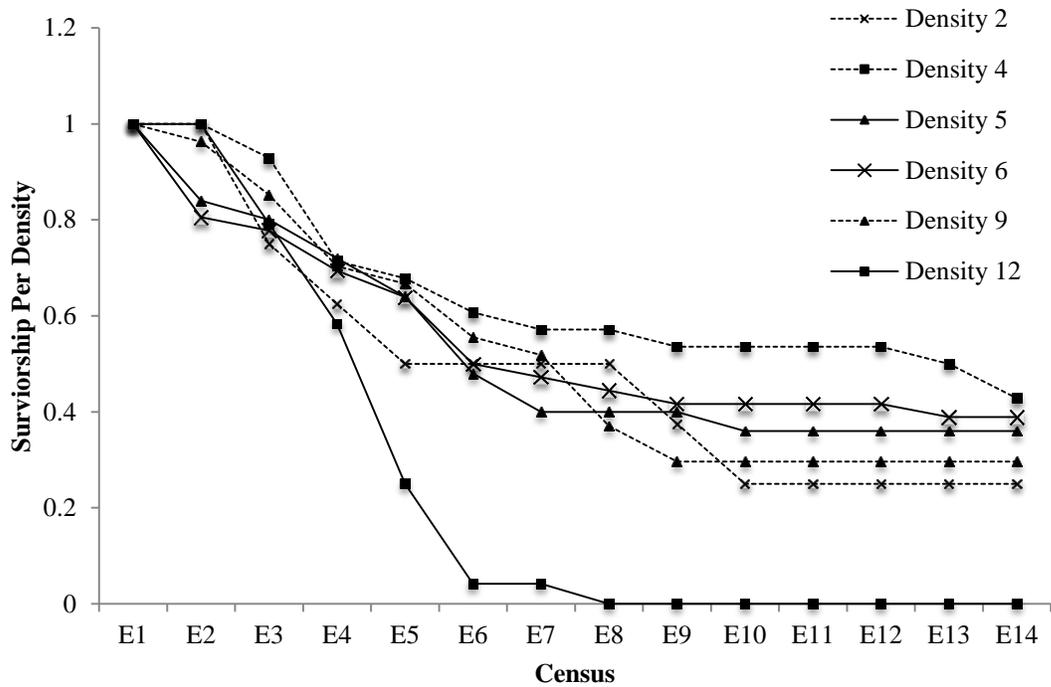


Figure 4. Survivorship per density over time of juvenile Houston toads (*Bufo houstonensis*) detected in 26 1 x 2 m² outdoor exclosures. Each exclosure contains one of six densities of juvenile Houston toads (2, 4, 5, 6, 9 and 12 individuals per exclosure). Initial release was 15 August 2014 (E1) and final census was 25 March 2015 (E14).

Table 1. Program MARK Cormack-Jolly-Seber (CJS) model AIC_c comparison for Juvenile Houston toads (*Bufo houstonensis*) in a juvenile density enclosure study. Density is represented by (d). AIC_c model chosen reflects survivorship parameter (ϕ) dependent by density and recapture parameter (p) is constant ($\phi(d)p(\cdot)$).

Model	AIC_c	ΔAIC_c	AIC_c Weight	Likelihood	# Par.	Deviance
$\phi(d)p(\cdot)$	1136.43	0	0.57	1	7	44.06
$\phi(d)p(d)$	1137.01	0.59	0.43	0.75	12	434.43
$\phi(\cdot)p(\cdot)$	1152.65	16.23	0	0	2	470.39
$\phi(t*d)p(\cdot)$	1180.55	44.12	0	0	78	332.1
$\phi(t*d)p(d)$	1182.38	45.95	0	0	83	321.97

Table 2. Survivorship estimate of juvenile Houston toads (*Bufo houstonensis*) from six stocking densities; 2, 4, 5, 6, 9, and 12 toads per 1 x 2 m² enclosure. $\phi(d)p(\cdot)$ was the Cormack-Jolly-Seber (CJS) model used for each treatment. Lower and Upper confidence intervals (CI) are reported.

Estimator	Estimate	Standard Error	Lower CI	Upper CI
Survivorship (ϕ)				
Density 2	0.889	0.043	0.774	0.949
Density 4	0.911	0.016	0.9	0.963
Density 5	0.939	0.021	0.06	0.945
Density 6	0.918	0.017	0.879	0.946
Density 9	0.902	0.021	0.851	0.936
Density 12	0.725	0.048	0.623	0.809
Recapture (p)				
Density 2	0.977	0.023	0.852	0.997
Density 4	0.905	0.02	0.857	0.938
Density 5	0.871	0.028	0.807	0.916
Density 6	0.915	0.019	0.87	0.945
Density 9	0.923	0.021	0.869	0.956
Density 12	0.978	0.022	0.861	0.997

Table 3. Linear mixed effects model, repeated measures analysis of variance (ANOVA) between SUL among five of the six toad densities (2, 4, 5, 6, 9, and 12) overtime using juvenile Houston toads (*Bufo houstonensis*). Only exclosures that contained at least one detection each census were used in these calculations. At the end of the study juvenile Houston toads were not detected in any of the 12 density exclosures. SUL was the measured variable with density and census as factors and exclosure as the random factor.

ANOVA	Degrees of Freedom	F-value	P-value
Density	4	2.3894	0.1044
Census	12	2.2671	0.0112
Density:Census	48	1.2249	0.1779

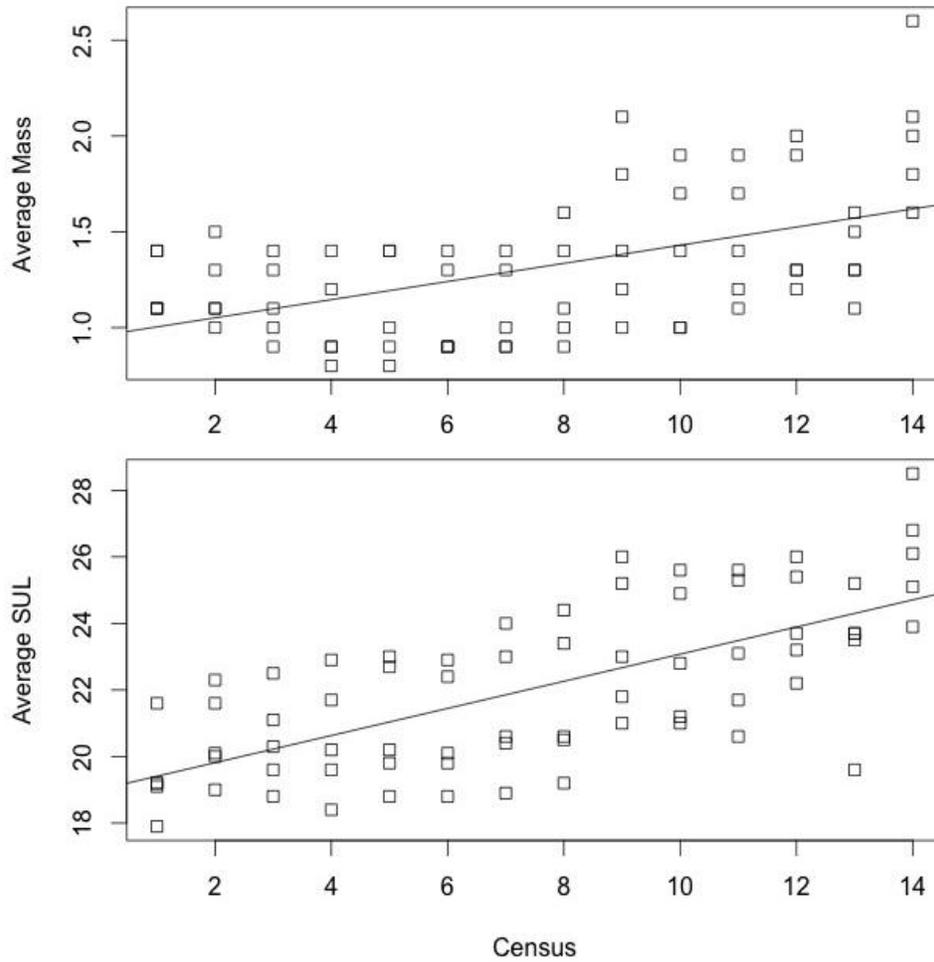


Figure 5. Linear regressions of 1) Total juvenile Houston toad (*Bufo houstonensis*) SUL over time (top) and 2) Total juvenile Houston toad mass over time (bottom) for all individuals who were detected during the final census.

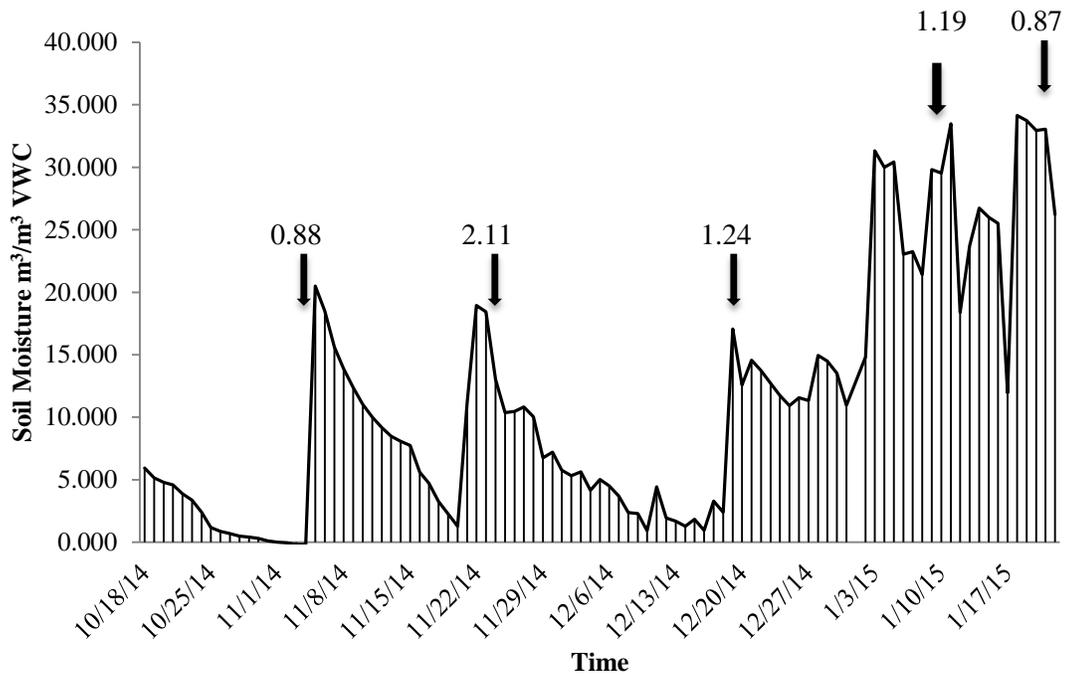


Figure 6. Changes in soil moisture at the juvenile Houston toad (*Bufo houstonensis*) pine habitat density exclosures in Bastrop County, Texas during October 2014 and January 2015. Soil moisture is measured by m^3/m^3 VWC and was recorded every 30 min, 24 hours a day. Rain events (more than 0.5 inches) are indicated by downward arrows.

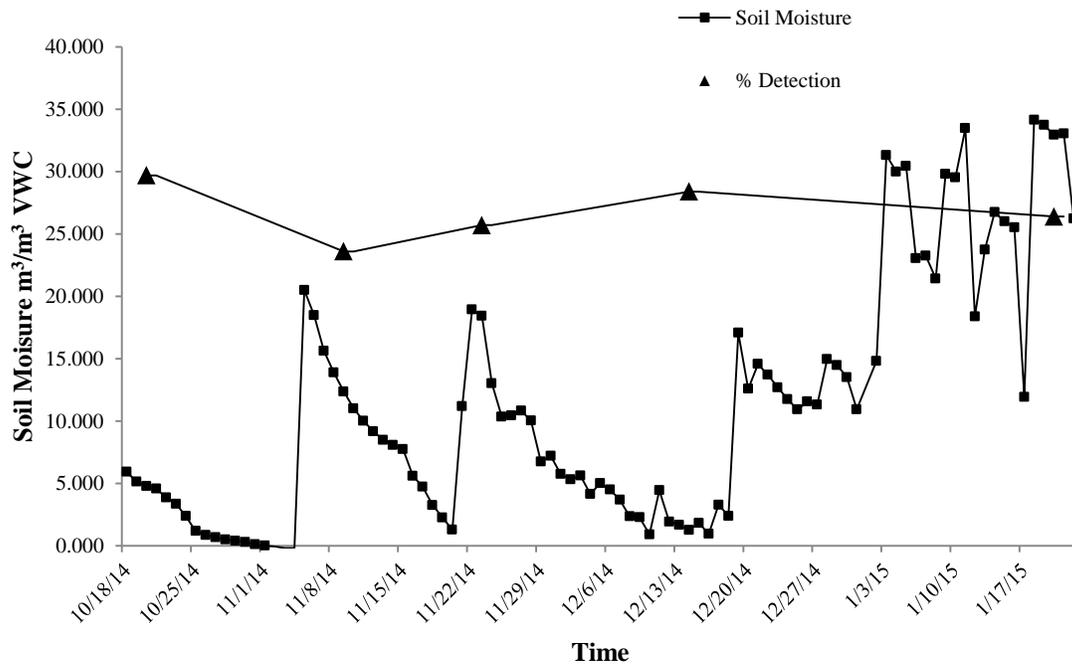


Figure 7. Changes in soil moisture with juvenile Houston toad (*Bufo houstonensis*) detection over time from October 18th 2014 until January 17th 2015. Juvenile Houston toad detection did not change as soil moisture changed overtime.

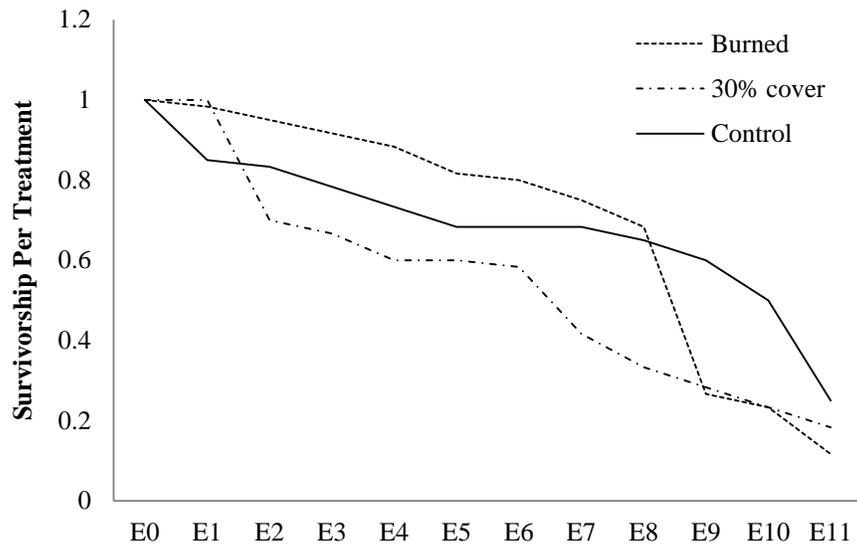


Figure 8. Survivorship per treatment over time of juvenile Houston toads (*Bufo houstonensis*) detected in 60 1 x 2 m² outdoor exclosures. Three treatments (burned, 30% cover, and control) contained 20 exclosures. Each exclosure contained 3 juvenile Houston toads. Initial release was March 30th, 2015 (E0).

Table 4. Program MARK Cormack-Jolly-Seber (CJS) model AIC_c comparison for Juvenile Houston toads (*Bufo houstonensis*) in a juvenile density enclosure study. Treatment is represented by (*g*) and time is represented by (*t*). AIC_c model chosen reflects survivorship parameter (ϕ) dependent by treatment and recapture parameter (*p*) is dependent on time and treatment ($\phi(g)p(t^*g)$).

Model	AICc	Delta AICc	AICc Weights	Model Likelihood	# Par	Deviance
$\phi(g)p(t^*g)$	2054.0915	0	0.99995	1	36	783.1474
$\phi(t)p(t^*g)$	2074.0906	19.9991	0.00005	0.0001	44	787.5884
$\phi(t^*g)p(t^*g)$	2082.9150	28.8235	0	0	66	752.6506
$\phi(.)p(t^*g)$	2118.8749	64.7834	0	0	24	873.9296
$\phi(t^*g)p(g)$	2157.7081	103.6166	0	0	36	886.7640
$\phi(t)p(g)$	2186.4074	132.3159	0	0	14	962.5039
$\phi(g)p(g)$	2207.4390	153.3475	0	0	6	999.9763
$\phi(t^*g)p(.)$	2264.4512	210.3597	0	0	34	997.8985
$\phi(t^*g)p(t)$	2265.6410	211.5495	0	0	44	979.1389
$\phi(.)p(t)$	2302.3468	248.2553	0	0	12	1082.5857
$\phi(.)p(.)$	2304.7817	250.6902	0	0	2	1105.4124

Table 5. Survivorship estimate of juvenile Houston toads (*Bufo houstonensis*) from three treatments; control, 30% cover, burned with three juvenile toads per enclosure with a total of X enclosures. $\phi(g)p(t^*g)$ was the Cormack-Jolly-Seber (CJS) model used for each treatment. Lower and Upper confidence intervals (CI) are reported.

Estimator	Estimate	Standard Error	Lower CI	Upper CI
Survivorship (ϕ)				
Control	0.9623880	0.0104129	0.9357316	0.9782452
30% Cover	0.9040133	0.0174735	0.8638862	0.9332251
Burned	0.9739031	0.0089074	0.9494338	0.9866975

Table 6. Linear mixed effects model, repeated measures analysis of variance (ANOVA) between SUL among three treatments (control, 30% cover, burned) overtime using juvenile Houston toads (*Bufo houstonensis*). Only exclosures that contained at least one detection each census were used in these calculations. SUL was the measured variable with treatment and census as factors and exclosure as the random factor.

ANOVA	Degrees of Freedom	F-value	P-value
Treatment	2	12.153	<.0001
Census	11	97.218	<.0001
Density:Census	22	2.962	<.0001

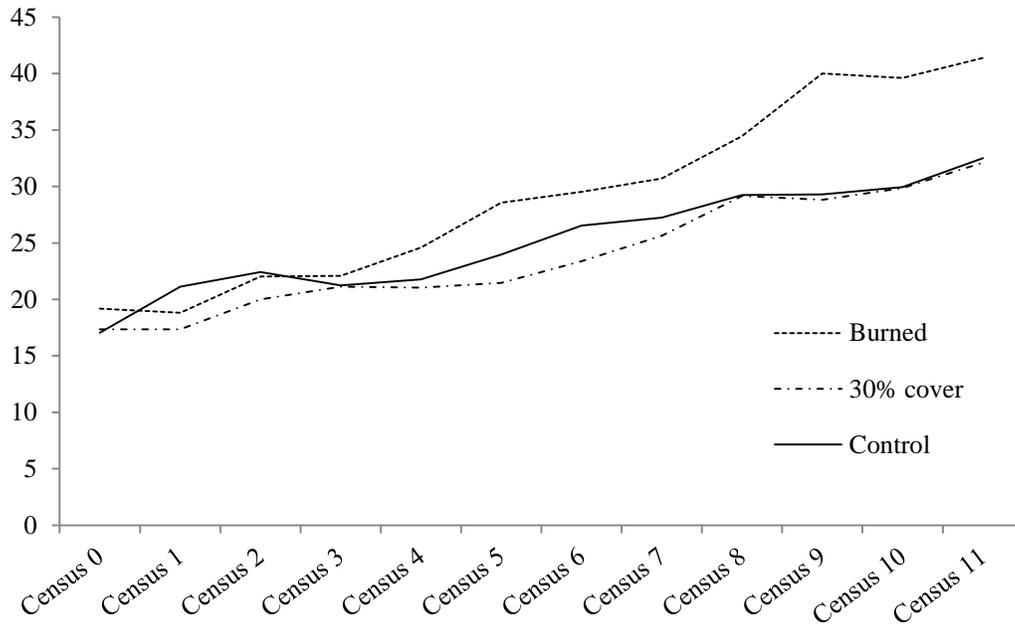


Figure 9. Average snout urostyle length (sul) over time among the three habitat treatments (burned, 30% cover, control). Average sul was significantly larger in the burned habitat treatment.

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