EFFECTS OF PREDATION ON FOUNTAIN DARTERS STUDY

HABITAT CONSERVATION PLAN (HCP) 2014 APPLIED RESEARCH

PREPARED FOR:

Edwards Aquifer Authority
900 E. Quincy St.
Antonio, Texas 78215

FINAL REPORT

October 2014

PREPARED BY:

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

BIO-WEST, Inc.
1812 Central Commerce Court
Round Rock, Texas 78664

TABLE OF CONTENTS

List of Tables

List of Figures

1.0 INTRODUCTION

The Edwards Aquifer Habitat Conservation Plan (HCP) is founded on long-term biological goals for the covered species that inhabit the Comal and San Marcos springs/river ecosystems (EARIP 2011). To support the long-term biological goals, flow management objectives (flow regimes) were established that are presumed to be protective of the threatened and endangered species in these systems. The low-flow conditions (discharge and extended durations) incorporated in the HCP flow regime and projected to occur during severe drought have occurred very infrequently (or not at all) during the historical record. Consequently, complete testing of ecological response(s) to these conditions in the wild is unlikely. Therefore, testing of simulated conditions in laboratory and/or field environments is mandatory to address HCP unknowns.

Section 6.3.4 of the HCP provides an outline for answering key questions and filling in data gaps to test assumptions and ultimately assist with management decisions. The focus in 2013 was on addressing several key questions surrounding physical habitat and food source responses both related to the federally-listed endangered Fountain Darter *Etheostoma fonticola*. In 2014, three additional applied research projects focused on the Fountain Darter were conducted. This report focuses on a test of cascading predation on the Fountain Darter.

A management option designed to reduce predation on Fountain Darters that has been discussed would be to remove piscine predators, especially in modified habitats (Spring Lake in the San Marcos system and Landa Lake in the Comal system). However, an unintended consequence might be a trophic cascade where the removal of the piscine predator would increase crayfish populations that would, in turn, decrease Fountain Darter populations. Therefore, the objective of this study was to quantify consumption of Fountain Darters by crayfish and bass separately and combined with and without aquatic vegetation in a laboratory setting. With a cascading effect, we predict that darter consumption would be greater with bass only or crayfish only than bass and crayfish combined (i.e., bass are consuming the crayfish instead of the darters). With an additive effect, we predict that darter consumption would be greater with bass and crayfish combined rather than bass only or crayfish only.

HCP Ecological Model Parameterization

The 2014 Fountain Darter predation study directly assessed the potential for a cascading effect of predation on the Fountain Darter. Based on the results of this study and complexity of the subject, this particular phenomenon is not currently programmed into the Fountain Darter module of the HCP Ecological model. However, preliminary investigations with rates of Fountain Darter predation in enclosed areas has informed the project team on potential ranges for rates of predation during extensive habitat reduction and organism (predators and prey) clumping that is anticipated to occur during extended low-flow conditions. Mean percentages (±1 SE) of Fountain Darters consumed were 29% (10.0) by crayfish, 29% (11.9) by bass, and 63% (18.0) by crayfish and bass, independent of vegetation presence, though consumption of Fountain Darters by crayfish or by bass did not differ from the control (i.e., without bass or crayfish). Although the report puts forward parameters for consideration in model parameterization, it is emphasized that specific use of any of the 2014 applied research will be determined by the HCP ecosystem modeling team with guidance from the HCP Science Committee.

Recommendations for Future Applied Research
The Fountain Darter predation study was successful in assessing a cascade effect of predation relative to Fountain Darters and in developing preliminary predation rates for incorporation into the ecological model. However, remaining questions exist: Will predations rates increase with predictably warmer water temperatures associated with low flow events during the summer months? Will predation rates decrease with predictably cooler water temperatures associated with low flow events in the winter months? Does plant type, specifically surface area, density or height, influence predation rates?

Acknowledgments

The project team would like to acknowledge the U.S. Fish and Wildlife Service San Marcos Aquatic Resource Center (SMARC) scientists and staff for guidance and contributions. We would also like to thank the HCP Science Committee for their timely input regarding approaches and methods for research activities.

2.0 DATA REVIEW AND AVAILABLE LITERATURE

Optimum Foraging Theory states that piscine predators are likely influenced by prey item availability and energy spent in search of prey (MacArthur and Pianka 1966; Stephens and Krebs 1986). When in the presence of piscine predators such as bass (*Micropterus* and *Ambloplites*), prey species (i.e., benthic fishes) tend to move less and seek refuge in substrates (Rahel and Stein 1988). Applications of the manipulative laboratory studies to natural systems likely do not adequately estimate predation risks of benthic fishes, such as darters. This is due to the presence of additional predators (Sih et al. 1998; Thomas 2011) and additional prey (pelagic minnows, drifting invertebrates, terrestrial sources; Dahl and Greenberg 1996, Magoulick 2004, Sullivan et al. 2012) that can synergistically increase or decrease predation rates on prey.

Among simple predator-prey relationships (1 predator and 1 or more aquatic prey), bass (*Micropterus* and *Ambloplites*; < 130 mm in TL) alter habitat selection of *Etheostoma* (Magoulick 2004) and consume *Etheostoma* at an average rate of about 4.5 fish per 24 hours (Rahel and Stein 1988) with lower capture rates in shallow water (<10 cm in depth; Angermeier 1992). Darters in the presence of piscine predators move less, relying on chemical and visual cues to reduce movement (Becker and Gabor 2012) and cryptic coloration for concealment (Armbruster and Page 1996), or seek refuge in substrates (Rahel and Stein 1988; Matthews 1998). Crayfish, which only recently has been recognized as a formidable predator of benthic fishes (Taylor and Soucek 2010), prompt darters to move more and avoid refuge among available substrates because the refuge is also habitat for the crayfish (Rahel and Stein 1988, Thomas 2011). Darter densities are inversely related to crayfish densities among 30 streams in Illinois, supporting Taylor and Soucek (2010) findings that crayfish consume, and therefore, directly or indirectly displace substantial numbers of benthic fish. When combining the two predators (bass and crayfish), predation risk on darters is difficult to quantify. Bass might consume more darters because darter movement has to increase to avoid predation by crayfish (Rahel and Stein 1988). Alternatively, bass might consume fewer darters, preferring to consume crayfish, following the Optimum Foraging Theory (e.g., consuming the most calories while expending the least energy; MacArthur and Pianka 1966), which is influenced by densities of prey items and energy spent in search for prey (Stephens and Krebs 1986). Energy expenditures can be mediated by amounts of refuge provided by vegetation. Regardless of the prey selection by bass, a management option designed to reduce predation on Fountain Darters might be to remove piscine predators. However, an unintended consequence might be a trophic cascade where the removal of the piscine predator would increase crayfish populations that would, in turn, decrease Fountain Darter populations. Thus, management recommendations based on an oversimplified understanding of predator-prey relationship could have unintended effects.

3.0 MATERIALS AND METHODS

Preliminary studies were developed to quantify 1) predation rates (i.e., number of darters consumed) and behavior of darters with crayfish and with and without bass, 2) predation rates of darters with bass and with and without crayfish, and 3) effects of vegetation on consumption of darters by crayfish and bass. During the preliminary studies, four Largemouth Bass *Micropterus salmoides* (one for each tank as needed; TL of ~160 mm), five crayfish *Procambarus clarkii* (~26 mm carapace length) for each tank, and three Greenthroat Darters *Etheostoma lepidum* (used as a representative benthic fish; ~37 mm TL) for each tank were used to begin the experiments. Alterations to the number of specimens, body length size, and amount of aquatic vegetation used occurred until crayfish consumed darters and bass consumed crayfish and darters. Predators were withheld of food for a 24-hour period. Duration of experiment was established when ≥75% of prey were consumed.

Once ratios and size of organisms and vegetation were established, 24 mesocosms; each representing an experimental unit, were established in the Freeman Aquatic Building (FAB) - holding house at Texas State University (Figures 1 and 2). Each mesocosm consisted of a 50 gallon plastic container modified to hold 30 gallons of water with an exchange rate of 0.6 gallons per minute (current velocity <0.1 cm/s). The water source for the mesocosms was well water from the artesian well adjacent to the FAB. Water temperature, nitrates, ammonia, nitrite, pH and dissolved oxygen were monitored daily during duration of the study (12 days) (Table 1). The completely randomized design consisted of four Fountain Darters, vegetation or no vegetation (Treatment 1) and predator type [Treatment 2: control (no predator), bass only (N =1), crayfish only (N=6), bass (N = 1) and crayfish (N = 6) combined] with three replications for each level of treatment. Vegetation was artificial plants (Figure 3) secured to the bottom of the mesocosm, covering about three-quarters of the mesocosm bottom and extending about 50 mm in the water column. Treatment 1 and Treatment 2 were randomly assigned to an experimental unit.

Figure 1. Setting up tanks for the predation study.

Figure 2. Laboratory setup using the concrete raceways at the Freeman Aquatic Building.

Table 1. Mean (± 1 SE) of water quality parameters taken from experimental units.

Parameters	Mean	St. Error
Water Temperature (°C)	22.26	0.02
Nitrate (ppm)	8.39	0.10
Conductivity (µm/cm²)	631.11	0.19
pH	7.85	0.01
Ammonia (ppm)	0.03	0.01
Nitrite (ppm)	0.00	
Dissolved Oxygen (mg/L)	8.07	0.07

At the end of the study, bass and crayfish were removed from the experimental units and numbers of Fountain Darters remaining were enumerated. Effects of vegetation and predator type on number of Fountain Darters consumed (i.e., missing from each experimental unit) and percent Fountain Darters consumed (arcsine-transformed) were assessed with two separate two-factor ANOVA (α=0.05), followed by post-hoc Fisher's Least Significance Difference test.

Figure 3. Crayfish and Largemouth bass treatment with artificial vegetation.

4.0 RESULTS AND DISCUSSION

Mean consumption (±1 SE) of Fountain Darters (N = 4 per experimental unit) was 1.7 (0.67) in no vegetation and 0.7 (0.33) in vegetation by crayfish only and was 0.7 (0.33) in no vegetation and 1.7 (0.88) in vegetation by bass only (Figure 4). Mean consumption of Fountain Darters was 3.0 (1.00) in no vegetation and 2.0 (1.15) in vegetation by crayfish and bass. Mortality or loss of fish was not observed in the control experimental units. Interaction term between vegetation and predator treatments was not significant ($P = 0.43$) and subsequently dropped from the model. As such, mean consumption of Fountain Darters was 1.2 (0.40) for crayfish, 1.2 (0.48) for bass, and 2.5 (0.72) for crayfish and bass (Figure 4). Mean consumption of Fountain Darters was 1.7 (0.49) for no vegetation and 1.2 (0.38) for vegetation. Treatment effects of vegetation and predator type on Fountain Darter consumption were detected ($P = 0.03$) in the full model. Numbers of Fountain Darters consumed were not different between vegetation and no vegetation ($P = 0.61$) but differed ($P = 0.02$) among predator type. Crayfish and bass treatment level consumed more Fountain Darters than the control, but did not differ from crayfish only or bass only treatment levels.

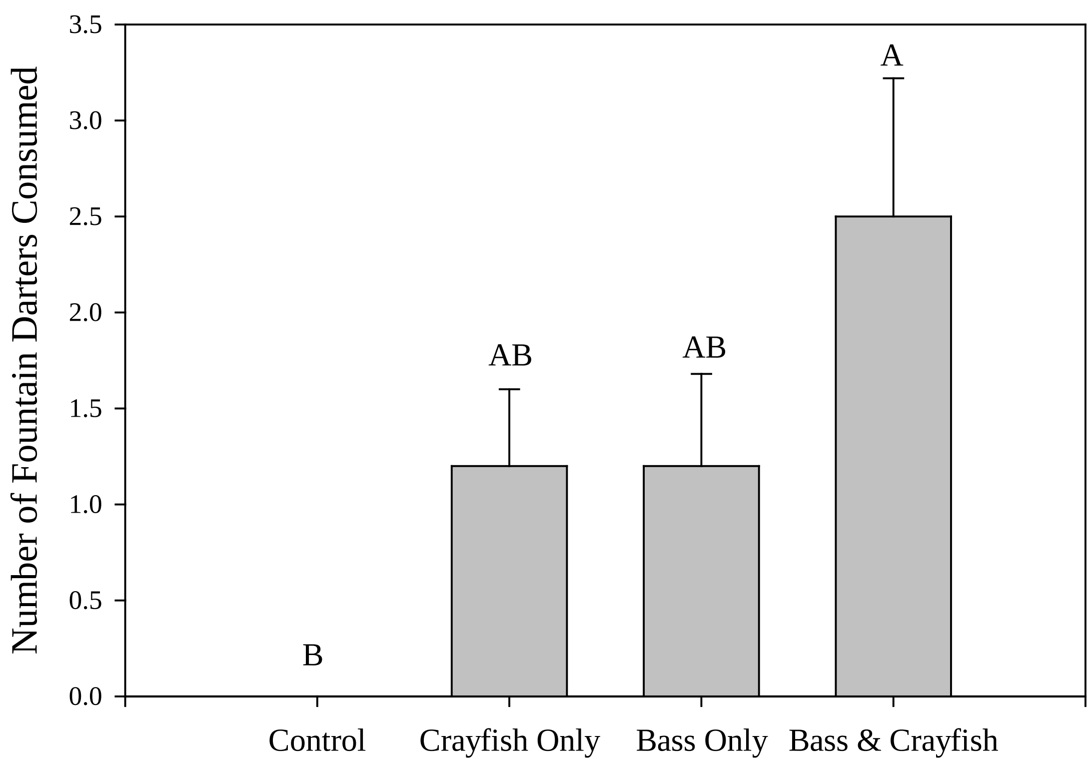

Figure 4. **Number of Fountain Darters consumed at four treatment levels (control: no predator, Crayfish only, Largemouth Bass only, and Largemouth Bass and Crayfish combined) under a laboratory setting. Same upper case letters represent no significant difference ($P > 0.05$).**

Mean percent consumption (±1 SE) of Fountain Darters was 41% (16.7) in no vegetation and 16% (8.3) in vegetation by crayfish only and was 16% (8.3) in no vegetation and 41% (22.0) in vegetation by bass only (Figure 5). Mean consumption of Fountain Darters was 75% (25.0) in no vegetation and 50% (28.9) in vegetation by crayfish and bass. Interaction term between vegetation and predator treatments was not significant ($P = 0.51$) and subsequently dropped from the model. Mean percent consumption of Fountain Darters was 29% (10.0) for crayfish, 29% (11.9) for bass, and 63% (18.0) for crayfish and bass (Figure 5). Mean percent consumption of Fountain Darters was 44% (12.3) for no vegetation and 29% (9.8) for vegetation. Treatment effects of vegetation and predator type on Fountain Darter consumption were detected ($P = 0.01$) in the full model. Numbers of Fountain Darters consumed were not different between vegetation and no vegetation ($P = 0.47$) but differed ($P < 0.01$) among predator type. Crayfish and bass treatment level consumed more Fountain Darters than the control, but did not differ from crayfish only or bass only treatment levels.

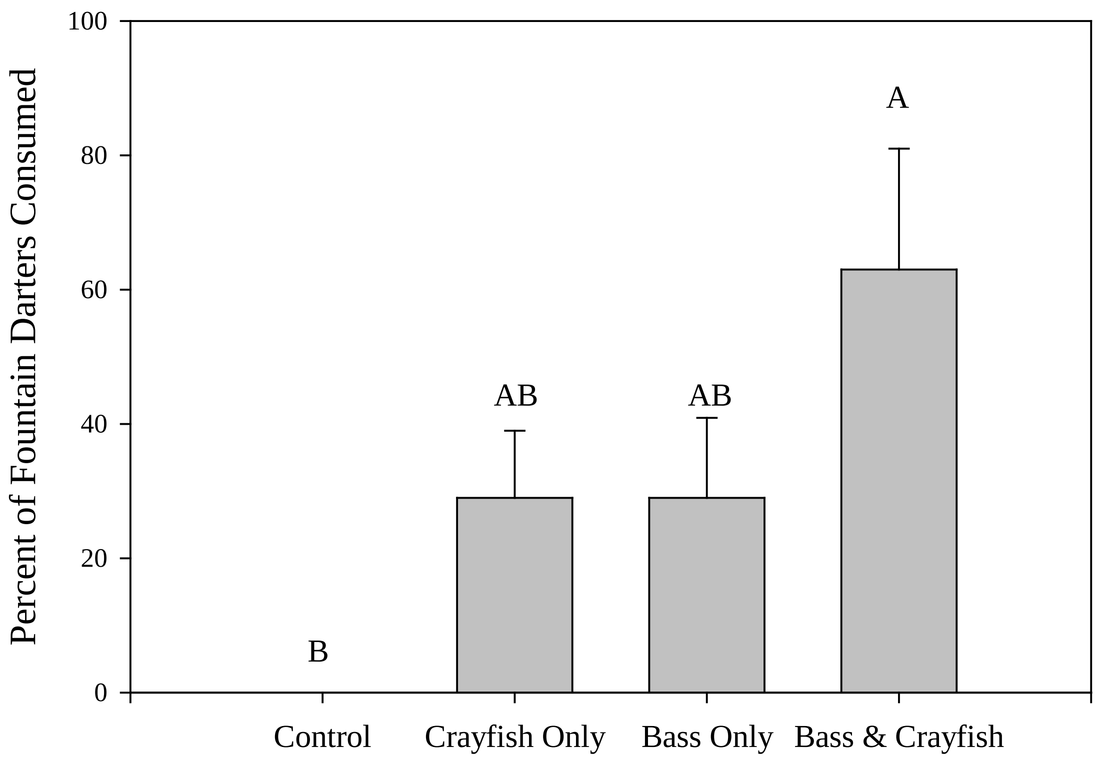

Figure 5. **Percent of Fountain Darters consumed at four treatment levels (control: no predator, Crayfish only, Largemouth Bass only, and Largemouth Bass and Crayfish combined) under a laboratory setting. Same upper case letters represent no significant difference ($P > 0.05$).**

For the number and percent of Fountain Darters consumed, greater numbers of Fountain Darters were consumed in the presence of both predators than the control (without a predator). Consumption and percent consumption of Fountain Darters by crayfish only and bass only did not differ from the control. Predation did not occur in one crayfish with vegetation experimental unit and two bass only treatment experimental units (one with and one without vegetation). Lack of predation in these three experimental units was unexpected given that preliminary studies demonstrated eventual predation by all fish and crayfish. Nevertheless, the study results did allow for an assessment of cascading effects. Cascading effects were not detected, suggesting that presence of two predators do not decrease predation on Fountain Darters. Results of this laboratory experiment have applications to the field in addition to testing for cascading effects. Crayfish and bass will consume Fountain Darters in confined spaces and in the absence of other prey items. The amount of Fountain Darters consumed, however, should be cautiously applied to field situations due to potential differences in predator/prey density, presence of other species, and potential differences in habitat complexity.

For ectotherms, energy demands increase with water temperature. An aspect not tested in this study was the consumption of darters at warmer temperatures, which also will occur with decreases in stream flow. We predict that numbers and percent of darters consumed would increase at warmer water temperatures, since predation and trophic cascading are dependent on water temperature (Kishi et al. 2005). Correspondingly, we would predict that numbers and percent of darters consumed would decrease at cooler water temperatures, which occur during winter months with low flows.

Consumption of Fountain Darters was independent of vegetation as provided in this study. Predator success (e.g., Largemouth Bass) is decreased by vegetation amount and density that obscure predator visual contact with prey (Savino and Stein 1982), so our results with respect to vegetation were unanticipated. We selected artificial vegetation to control amounts of vegetation in each experimental unit. Artificial vegetation resembled a short-growing patch of *Hygrophila*, which is available in the Fountain Darter's natural habitat but one of many plant types available. To further explore the relationship between predation and vegetation type, a similar experimental design could be used but treatments would need to be more representative of the natural environment.

5.0 CONCLUSIONS

Levels of Fountain Darter consumption reported herein suggest that benthic fish consumption by crayfish and bass are additive and not interactive or cascading. Mortality of Fountain Darters was greater, though not statistically different from crayfish only and bass only, than the control. Implications of these findings demonstrate that crayfish and bass will consume Fountain Darters and suggest that both predators will additively increase mortality of Fountain Darters. As such, the removal of Largemouth Bass during low flow periods will predictably reduce Fountain Darter mortalities via predation without causing a trophic release (i.e., greater predation by crayfish).

6.0 REFERENCES

Angermeier, P. L. 1992. Predation by rock bass on other stream fishes: experimental effects of depth and cover. Environmental Biology of Fishes 34:171-180.

Armbruster, J. W. & L. M. Page, 1996. Convergence of a cryptic saddle pattern in benthic freshwater fishes. Environmental Biology of Fishes 45: 249–257.

Becker, L. J. S. and C. R. Gabor. 2012. Effects of turbidity and visual vs. chemical cues on anti-predator response in the endangered Fountain Darter (*Etheostoma fonticola*). Ethology 118:994-1000.

Dahl, J. and L. Greenberg. 1996. Impact on stream benthic prey by benthic vs. drift feeding predators: a meta-analysis. Oikos 77:177-181.

EARIP 2011. Habitat Conservation Plan. Prepared for the Edwards Aquifer Recovery Implementation Program.

Kishi, D., M. Murakami, S. Nakano, and K. Maekawa. Water temperature determines strength of top-down control in a stream food web. Freshwater Biology 50:1315-1322.

MacArthur, R. H. and E. R. Pianka. 1966. One optimal use of a patchy environment. The American Naturalist 100:603-609.

Magoulick, D. D. 2004. Effects of predation risk on habitat selection by water column fish, benthic fish and crayfish in stream pools. Hydrobiologia 527:209-221.

Rahel, F. J. and R. A. Stein. 1988. Complex predator-prey interactions and predator intimidation among crayfish, piscivorous fish, and small benthic fish. Oecologia 75:94-98.

Savino, J. F. and R. A. Stein. 1982. Predator-prey interaction between largemouth bass and bluegills as influenced by simulated, submersed vegetation. Transactions of the American Fisheries Society 111:255-266.

Sih, A. G. Englund, and D. Wooster. 1998. Trends in Ecology and Evolution 13:350-355.

Stephens, D.W. and Krebs, J.R. 1986. Foraging theory. Princeton: Princeton University Press. 464 pp.

Sullivan, M. L., Y. Zhang, and T. H. Bonner. 2012. Terrestrial subsidies in the diets of stream fishes of the USA: comparisons among taxa and morphology. Marine and Freshwater Research 63:409-414.

Taylor, C.A. and D.J. Soucek. 2010. Re-examining the importance of fish in the diets of stream-dwelling crayfish: Implications for food web analyses and conservation. *American Midland Naturalist* 163: 280-293.

Thomas, C. L. 2011. Crayfish: scavenger or deadly predator? Examining a potential predator-prey relationship between crayfish and benthic fish in aquatic food webs. MS Thesis. University of Illinois at Urbana-Champaign.