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ABUNDANCE, DISTRIBUTION, AND HABITAT OF THE THREATENED MINNOWS CAMPOSTOMA ORNATUM AND NOTROPIS CHIHUAHUA IN THE TRANS-PECOS REGION OF TEXAS

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ABSTRACT—The Rio Grande and its tributaries in the Trans-Pecos region of Texas have been impacted by a variety of anthropogenic activities such as dewatering and the introduction of nonnative species. These environmental manipulations have negatively affected the native fishes leading to extirpations and declining populations throughout the region. Campostoma ornatum and Notropis chihuahua inhabit tributary streams of the Rio Grande in the Trans-Pecos region and are considered as state-listed threatened species. Little is known about their status and ecological requirements in the region. We hypothesized that the distribution and abundance of these threatened minnows in these spring-fed habitats can be modeled by two primary factors: local environmental conditions that are maintained by spring flow and the abundance of introduced species such as the plains killifish, Fundulus zebrinus. We used classification and regression trees to analyze variation in abundance and incidence of the target species from Alamito, Terlingua, and Tornillo creeks, as well as the Rio Grande proper based on local environmental factors (e.g., size of stream and water quality), abundance of nonnative species, season, and distance from the Rio Grande. The analyses indicated that distance from the Rio Grande, maximum depth, and composition of substrate were the most important predictors for the abundance and occurrence of the target species in the region.

RESUMEN—El río Bravo y sus tributarios en la región Trans-Pecos de Texas han sido afectados por una variedad de actividades antropogénicas tales como la disecación y la introducción de especies no nativas. Estas manipulaciones ambientales han afectado negativamente a los peces nativos, llevándolos a la extirpación y a la disminución poblacional a través de la región. Campostoma ornatum y Notropis chihuahua habitan arroyos tributarios del río Bravo en la región Trans-Pecos y son considerados como amenazados por el estado. Se sabe poco sobre su estado y requerimientos ecológicos en la región. Planteamos que la distribución y abundancia de estos peces amenazados en estos arroyos de manantiales pueden ser modelados por dos factores principales: 1) condiciones ambientales locales que son mantenidas por el flujo del manantial, y 2) la abundancia de las especies introducidas como los Fundulus zebrinus. Usamos la clasificación y árboles de regresión para analizar la variación en la abundancia e incidencia de las especies focales de Alamito, Terlingua y Tornillo Creeks, así como también del río Bravo, basándonos en factores ambientales locales (por ejemplo, tamaño del arroyo y calidad del agua), abundancia de especies no nativas, la estación del año, y la distancia del río Bravo. Los análisis indicaron que la distancia del río Bravo, la profundidad máxima, y la composición del sustrato fueron los indicadores más importantes para la abundancia y la presencia de las especies focales en la región.

The Rio Grande system in the Trans-Pecos region of Texas contains many unique aquatic species and environments. This aquatic system has been impacted by a variety of anthropogenic activities, including dewatering of the mainstem Rio Grande and the introduction of nonnative species (Edwards et al., 2002). These environmental manipulations have negatively impacted native fishes, leading to extirpations and declining populations throughout the region (Hubbs, 1990).

Campostoma ornatum (Mexican stoneroller) and Notropis

chihuahua (Chihuahua shiner) inhabit tributary streams of the Rio Grande in the Trans-Pecos region and are considered as state-listed threatened species by the Texas Parks and Wildlife Department (Hubbs et al., 1991; Miller, 1972). Both species are on the Watch List of the Texas Organization for Endangered Species (1988) and listed as special concern by Williams et al. (1989). Little is known about the status and ecological requirement of either species, yet both are native components to desert stream ecosystems that are under considerable stress from

declining water quality and quantity (Hubbs and Wauer, 1973).

We hypothesized that the distribution and abundance of the target species in spring-fed habitats in the Trans-Pecos region can be modeled by two primary factors: local environmental factors and the abundance of nonnative species. Campostoma ornatum prefers riffles and pools with gravel substrate and clear, cool water (Contreras-Balderas, 1974; Burr, 1980a). Notropis chihuahua tends to inhabit springs with gravel-sand substrates and clear, cool water (Burr, 1980b; Burr and Mayden, 1981). We predicted that depth of pools, water clarity, and gravel substrate in the tributaries should be positively related to the incidence and abundance of both species. Hubbs and Wauer (1973) hypothesized that Fundulus zebrinus, a nonnative species, was impacting native species in Tornillo Creek in the Trans-Pecos region. We predicted that the abundance and distribution of both species would be negatively related to the abundance of F. zebrinus.

MATERIALS AND METHODS—The Rio Grande in the Trans-Pecos region is diverted to irrigate fields south and east of El Paso, Texas, and tends to be slow-moving, shallow, channelized, and heavily silted until it receives discharge from the Rio Conchos in Mexico (Fig. 1), its primary tributary (Hubbs et al., 1977; Edwards et al., 2002). The discharge of the Rio Conchos downstream of their junction significantly changes the habitat characteristics of the Rio Grande in comparison to upstream reaches (Bestgen and Platania, 1988). The Rio Grande downstream of the Rio Conchos has deeper runs, larger substrate (e.g., cobble and rubble), and lower conductivity and salinity than does the Rio Grande upstream of the Rio Conchos (Bestgen and Platania, 1988). The Rio Grande in the Trans-Pecos region also receives water from a series of tributaries including Cibolo, Alamito, Terlingua, and Tornillo creeks. We conducted seasonal monitoring of fish and their environment at 3–5 month intervals at Alamito (1 site), Terlingua (3 sites), and Tornillo (1 site) creeks, and the Rio Grande (5 sites) in Presidio and Brewster counties (Fig. 1, sites 1–10), Texas, from August 2009–June 2011 (total of 79 samples). Additionally, we conducted regional collections of fish at 27 sites in tributaries (Fig. 1, sites 11–37) from October 2009–May 2011. We used the 79 monitoring samples to examine the temporal distribution and patterns of abundance of both species. We used 32 sites in tributaries (5 monitored sites and 27 additional sites in tributaries throughout the region) to examine spatial distribution and patterns of abundance of both species in the tributaries.

We sampled fishes from each site by seine (4.2 m by 1.7 m, 5 mm mesh), dragged by hand for 30–60 min per site. We converted abundance data into catch rate (catch per one seining hour) for statistical analyses. We sampled all available types of habitat (i.e., riffles, pools, and runs) within a stream reach. We identified, counted, and returned to the water fishes >25 cm in total length. We fixed smaller fishes in 10% formalin and brought them to the laboratory for identification and then preservation in 50% ethanol. We curated all collections of fishes into Texas Natural History Collection (TNHC). For each locality where we sampled fishes, we also collected data on habitat. We used a Multiparameter Meter (Hanna Instruments, Schertz,

Texas, USA) to measure pH and specific conductance and used an Aquafluor Handheld Fluorometer and Turbidimeter (Turner Designs, Sunnyvale, California, USA) to measure chlorophyll-a concentration and turbidity. Within each site, we made 5–8 transects perpendicular to flow of the stream and spaced at 10– 20 m intervals along the sampled stream reach. We then calculated mean width and depth of the stream and composition of substrate across transects. We categorized substrate according to Taylor and Lienesch (1996a, 1996b) and measured maximum current velocity of the sampled reach according to Taylor et al. (2008). We measured distance of the water course between the Rio Grande and its tributary localities with Google Earth (http://www.google.com/earth/index.html).

We used classification and regression trees (CART) to describe variation in catch per hour and incidence of the target species based on distance from the Rio Grande, local environmental factors (maximum depth, mean depth, mean width, pH, turbidity, specific conductance, chlorophyll-a concentration, substrate composition, and maximum current velocity), the catch per hour of nonnative fishes (F. zebrinus and Lepomis cyanellus), and season. The methodology of CART recursively splits a matched dataset of categorical variables (for classification trees) or continuous variables (for regression trees) into progressively smaller mutually exclusive groups, using binary splits based on single independent or predictor variables (De'ath and Fabricius, 2000; Prasad et al., 2006). Models from CART have advantages over parametric statistical analyses (e.g., multiple regression models) because of the applicability to cases in which the relationships between variables are strongly nonlinear or involve high-order interactions (Rahel and Jackson, 2007). Additionally, CART can admit a mix of categorical and continuous variables and is insensitive to monotonic transformations of the predictor variables because they rely on the rank ordering of variables (McCune and Grace, 2002; Rahel and Jackson, 2007). We used incidence of fish for classification trees (fitting method: Gini index; minimum split index value: 0.05; minimum improvement in the proportion of reduction in error: 0.05; minimum count allowed in each node: 5) and abundance of fish for regression trees (fitting method: least squares; minimum split index value: 0.05; minimum improvement in the proportion of reduction in error: 0.05; minimum count allowed in each node: 5). We conducted CART analyses for the 79 monitoring samples and the regional dataset (5 monitored sites in tributaries and 27 additional sites throughout the region) separately. We used SYSTAT 11 (SYSTAT Software, Inc., Richmond, California, USA) to perform CART analyses.

RESULTS—We caught C. ornatum ($n = 2,590$) from 14 of the 37 sampled sites and N. *chihuahua* ($n = 1,534$) from 12 of the 37 sampled sites (Fig. 1). Campostoma ornatum occurred in Alamito Creek (mean abundance $= 115.7$), Terlingua Creek (mean abundance $= 30.6$), and the Rio Grande (mean abundance $= 0.1$). Notropis chihuahua occurred in Terlingua Creek (mean abundance = 38.6), Tornillo Creek (mean abundance $= 0.1$), and the Rio Grande (mean abundance $= 0.6$).

A classification tree using distance from the Rio Grande and season explained 62% of the variation in incidence of C. ornatum at the 10 monitored sites. The

FIG. 1-Maps of 37 sampled sites and distribution of (a) Campostoma ornatum and (b) Notropis chihuahua in the Rio Grande and tributaries in Trans-Pecos region, Texas, 2009–2011. Squares and circles (solid = present, open = absent) indicate monitored sites and additional regional sites, respectively.

first split in the classification tree of incidence for C. ornatum was based on distance from the Rio Grande at a value of 3.7 km (Fig. 2a). In other words, the mean incidence of C. ornatum at monitored sites on and near the Rio Grande \langle < 3.7 km, sites in Alamito and Tornillo creeks) was considerably lower (mean incidence $= 0.05$) than at monitored sites further from the Rio Grande (sites 4, 5, and 9 in Terlingua Creek; mean incidence $=$ 0.62), which were well upstream of the confluence with the river. The second split in the classification tree of incidence of C. ornatum (Fig. 2a) at the monitored sites also was based on distance from the Rio Grande at a value of 20.8 km. Thus, occurrences of C. ornatum were more prevalent at the monitored sites most upstream in Terlingua Creek (mean incidence $= 1$). The third split in the classification tree for C. ornatum occurrence (Fig. 2a) was based on season. The mean incidence of C. ornatum at monitored sites in Terlingua Creek from August-November was lower (mean incidence $= 0.12$) than from March-June (mean incidence $= 0.75$). A

FIG. 2—Results of classification and regression trees for incidence and abundance of *Campostoma ornatum* (a, b) and *Notropis* chihuahua (c, d) collected in 10 monitored sites in the Rio Grande and tributaries in Trans-Pecos region, Texas, 2009–2011 (total of 79 samples). Numbers in parentheses indicate the percentage of variation explained by environmental variables.

regression tree (based on abundance rather than incidence) using distance from the Rio Grande (split occurred at 20.8 km) explained 42% of the variation in abundance of C. ornatum in the 10 monitored sites (Fig. 2b). The split divided the monitored sites upstream in Terlingua Creek from all others. Campostoma ornatum had a mean abundance of 184.7 individuals at this site, which was considerably higher than at the other monitored sites where the species occurred (mean abundance $= 4.8$). To summarize, C. ornatum occupied Terlingua Creek throughout the study but was more prevalent at the monitored site most upstream and during the spring season. Campostoma ornatum occurred minimally in the sites on the Rio Grande.

For N. chihuahua, a classification tree using distance from the Rio Grande (3.7 km; Fig. 2c) explained 78% of the variation in incidence of N. chihuahua across the 10 monitored sites. Thus, as with C. ornatum, the incidence of N. chihuahua was highest at the monitored sites in Terlingua Creek (mean incidence $= 0.95$) and was considerably lower elsewhere (mean incidence $= 0.05$). A regression tree using maximum depth (split at 1.8 m) explained 14.5% of the variation in abundance of N. chihuahua across the 10 monitored sites (Fig. 2d). In other words, mean abundance of N. chihuahua was highest where deep-pool habitat was found (mean abundance $= 138.9$. To summarize, N. *chihuahua* occupied monitored sites in Terlingua Creek throughout the study but was most abundant in the deeper, spring-fed pools of Terlingua Creek.

The regional analyses for C. ornatum were more complex than analyses for monitored sites. For incidence of C. ornatum, the first split was again based on distance from the Rio Grande (Fig. 3a). However, this split largely separated the occurrences in Terlingua Creek (right split, all in the lower one-half of the drainage) from those in Alamito Creek (left split, all in the upper one-half of the drainage). This odd pattern of distribution seems confusing until habitat is considered. Gravel substrates were prevalent in upper Alamito Creek and corresponded to a relatively high level of occurrence of C. ornatum (mean incidence $= 0.4$). The branch of Terlingua Creek was further split by distance such that lower 3.9 km of stream above the confluence with the Rio Grande contained a relatively low level of incidence (mean incidence $= 0.2$) compared to the localities between 3.9 and 28 km upstream of the confluence (mean incidence $= 0.8$). Based on the amount of mud substrate present, the final split separated the localities in Terlingua Creek with high occurrence; high levels of mud substrate corresponded to lower levels of occurrence. Much of the upper Terlingua Creek watershed had muddy or bedrock substrates that were lacking in high percentages of gravel and in occurrence of C. ornatum. A regression tree based on abundance and using distance from the Rio Grande and gravel substrate explained 25% of the

FIG. 3—Results of classification and regression trees for incidence and abundance of Campostoma ornatum (a, b) and Notropis chihuahua (c, d) collected in 32 tributary localities of the Rio Grande in Trans-Pecos region, Texas, 2009–2011. Numbers in parentheses indicate the percentage of variation explained by environmental variables.

variation in abundance of C. ornatum across the 32 regional sites. The first split in the regression tree of abundance of C. ornatum was based on gravel substrate at a value of 38.8% (Fig. 3b). In other words, the mean abundance of C. ornatum at regional sites with less gravel (percentage of gravel substrate <38.8; mean abundance $= 16.7$) was considerably lower than for sites with more gravel substrate (percentage of gravel substrate ≥ 38.8 ; mean abundance $= 229.7$. The second split in this regression tree was based on distance from the Rio Grande at a value of 23.4 km. Thus, C. ornatum was more abundant upstream in Alamito Creek at sites with greater gravel substrate (\geq 23.4 km; mean abundance = 463). To summarize, C. ornatum was more prevalent in lower reaches of Terlingua Creek where muddy substrates were lacking but was more abundant in upper reaches of Alamito Creek where more gravel substrate was present.

For N. chihuahua, a classification tree using distance from the Rio Grande (split at 23.4 km) explained 36% of the variation in incidence across the 32 regional sites (Fig. 3c). Incidence of N. chihuahua was limited to only one tributary (Tornillo Creek) outside of Terlingua Creek, and all occurrences in Terlingua Creek were in the lower one-half of the drainage. The second split in the classification tree also was based on distance from the Rio Grande at values of 3.9 km and indicated that the localities near the Rio Grande confluence held fewer occurrences than localities further upstream (>3.9 km). A regression tree using distance from the Rio Grande and maximum depth explained 34% of the variation in abundance of N. chihuahua across the 32 regional sites. The first split was based on distance from the Rio Grande (21.3 km), and the second was based on maximum depth (0.76 m; Fig. 3d). Thus, localities in the lower reaches of Terlingua Creek that had deep-pool habitats contained the most N. chihuahua.

DISCUSSION—Our results indicated that C. ornatum and N. chihuahua primarily inhabited tributary systems to the Rio Grande but responded differentially to measured environmental factors. The result of CART analyses indicated that C. ornatum constantly occurred through much of the lower Terlingua Creek system and were more prevalent in the spring (March–June). Hubbs and Wauer (1973) reported that young and breeding adults were present in January, and half-grown young in May and June

in Tornillo Creek, suggesting that the breeding season of C. ornatum was winter to spring. Because most C. ornatum we collected in the downstream localities of Terlingua Creek were young of year, C. ornatum appeared to have spawned in Terlingua Creek in winter to spring and might have used the downstream localities for nursery sites. Conversely, season was not a predictor for the incidence of N. chihuahua, and we collected N. chihuahua throughout most of Terlingua Creek. Maximum pool depth was positively related to the abundance of N. chihuahua, suggesting that N. chihuahua might persist in the deep tributary habitats in Terlingua Creek.

Substrate was an important predictor for the incidence and abundance of C. ornatum in the regional analysis. Incidence and abundance of C. ornatum were positively related to the percentage of gravel substrate. Campostoma ornatum is an herbivorous, bottom feeder (Contreras-Balderas, 1974) and constructs spawning pits (Johnston, 1999). Notropis chihuahua is an invertivore (Burr and Mayden, 1981) and is likely a broadcast spawner (Johnston, 1999). Campostoma ornatum might rely on substrate for their foraging and spawning more than N. chihuahua in our system.

Historical records of assemblages of fishes (1977–1989) showed that C. ornatum and N. chihuahua occurred at our downstream locality (monitored site) in Alamito Creek (Hubbs et al., 1977; Bestgen and Platania, 1988; Linam et al., 2002). However, we did not collect either species at that locality. Our analysis indicated that pool depth and the percentage of gravel substrate were important variables in our study system. These habitats were lacking at the monitored site in Alamito Creek. Furthermore, this site has been strongly impacted by cattle, possibly affecting the ability of either species to persist at this locality.

Hubbs and Wauer (1973) hypothesized that F. zebrinus might be replacing C. ornatum in Tornillo Creek. In 1954, C. ornatum was the dominant species in Tornillo Creek (Hubbs and Wauer, 1973), occurring in 5 of 11 samples from 1967–1970. We did not collect any C. ornatum from Tornillo Creek. Conversely, F. zebrinus has expanded their distribution and abundance in our study system since 1956 (Hubbs and Wauer, 1973). Hubbs and Wauer (1973) reported that F. zebrinus occurred in 9 of 11 samples and was abundant in the creek in the spring months from 1967–1970. We found F. zebrinus in all of our samples from Tornillo Creek; thus, they appear to be persisting indefinitely in the creek. Although the abundance of F. zebrinus was not negatively related to the abundance and incidence of either of the threatened species in our CART analyses, F. zebrinus is now widespread and abundant in the region and may have impacts on native species that are yet to be seen. Additionally, red shiners (Cyprinella lutrensis) are now a dominant species in lower Alamito and Tornillo creeks, and their impact on the target

species in this system is unknown and should be further studied.

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