

Diet of Nonnative *Hyla cinerea* in a Chihuahuan Desert Wetland

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ABSTRACT.—*Hyla cinerea* recently introduced to a Chihuahuan Desert wetland in Big Bend National Park, Texas consumed a variety of terrestrial arthropods. We investigated its diet to analyze a component of colonizing potential for this species. Of 105 individuals analyzed, we found a total of 194 prey items representing 13 unique categories. The most important prey, determined with a quotient that combines volume and count with frequency, were beetles, cockroaches, grasshoppers and crickets, ants, and spiders. We documented the first occurrence of *Scorpiones* in the diet of *H. cinerea*, which demonstrates this invader's ability to exploit novel resource types outside of its native range. In comparison to other dietary studies conducted on *H. cinerea*, this nonnative population feeds on a similar diversity of prey in equal proportions. This analysis represents a first step into evaluating ecological effects of this nonnative population of *H. cinerea*.

Populations existing in geographically distinct habitats may show variability in their diet because of their environmental surroundings or phenotypic plasticity (Paine, 1980; Zerba and Collins, 1992). Understanding this variability in resource use is especially important to the study of invasion biology where novel species interact in new surroundings and potentially with new prey species. Impacts caused by nonnative organisms feeding in their new surroundings span the spectrum from ecologically devastating (Fritts and Rodda, 1998; Phillips et al., 2003; Wiles et al., 2003) to innocuous (Saenz, 1996). Too often we perceive the impacts of a species invasion after it is too late to gather baseline ecological information and plan management interventions (Enserink, 1999). As such, the earliest possible opportunity should be taken to assess potential impacts of the organism's arrival (Byers et al., 2002).

Hyla cinerea, a treefrog native to southeastern North America was discovered in September 2006 near a warm spring-fed pond in the Chihuahuan Desert of Big Bend National Park (BBNP) in west Texas (Leavitt et al., 2007). This location, near a campground, suggests the arrival was either as a stowaway on a vehicle or an intentional release. There are two documented instances of natural range expansions of Green Treefrogs, in South Carolina (Platt et al. 1999) and Illinois (Redmer et al. 1999). A natural range expansion to the Trans-Pecos region of the Chihuahuan Desert is unlikely because of the harsh conditions and distance of at least 320 km between the closest known breeding populations and this pond. No matter how *H. cinerea* arrived at this location, their local population size increased rapidly since its discovery (DJL, pers. obs.).

Desert wetlands are known hotspots for biodiversity and endemism because of their isolation and island-like qualities (Tiner, 2003). Many endemic inhabitants

of desert wetlands are endangered because of habitat loss and pressures from nonnative predators and competitors (Minckley and Deacon, 1968). For this reason, any invasion into these locations is of concern (Lovei, 1997; Vitousek et al. 1997). The site where this new population of *H. cinerea* is established is the only habitat remaining for the endemic fish, *Gambusia gaigei*, which is federally endangered because of its limited range and precarious history (Hubbs et al. 2002). In fact, *G. gaigei* was near extinction in the 1950s (reduced to only three individuals) but has now rebounded and persists in two ponds (Hubbs and Brodrick, 1963). It is prudent to understand what ecological effects *H. cinerea* may have in this system.

Existing dietary studies on *H. cinerea* include analysis of 100 frogs from Okefenokee Swamp, Georgia (Haber, 1926), 497 individuals from Gainesville, Florida (Kilby, 1945), 163 more frogs from Gainesville (Freed, 1982), and 130 frogs from Everglades National Park, Florida (Meshaka, 2001). These analyses indicate some variability exists between and among populations of this rather indiscriminant and opportunistic feeder. As such, it is valuable to conduct a dietary analysis on this new *H. cinerea* population to uncover further variability that might exist in this organism's diet and to analyze a component of its colonizing potential. Furthermore, empirical data on the prey *H. cinerea* is consuming at this location are necessary to inform management plans for dealing with this nonnative population. It is especially important that this analysis be conducted early in the establishment process. This baseline may prove valuable to maintaining the biological diversity of this isolated wetland.

MATERIALS AND METHODS

Collections were made in and around two small ponds in Rio Grande Village, Big Bend National Park, Brewster County, Texas. These ponds (Beaver Pond and Spring 4 Pond) are currently the only known locations for *H. cinerea* in BBNP and near the only localities for *G. gaigei*. Dominant plants that surround these ponds are *Arundo donax*, *Phragmites australis*, and *Vitis arizonica*. All individual frogs seen or heard were pursued by as many as four searchers on eight evenings between 15

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TABLE 1. Dietary composition of nonnative Green Treefrogs from Big Bend National Park, Texas ($N = 105$).

Prey category	Frequency (F)		Volume (V)		Count (N)		Importance (I)
	No.	%	mm ³	%	No.	%	(% F + % V + % N)/3
Araneae	19	12.03	689.39	6.15	20	10.31	9.49
Blattodeae	21	13.29	3,623.99	32.31	27	13.92	19.84
Coleoptera	49	31.01	3,372.84	30.07	68	35.05	32.05
Diptera	1	0.63	61.89	0.55	1	0.52	0.57
Formicidae	22	13.92	96.28	0.86	31	15.98	10.25
Hemiptera	7	4.43	589.02	5.25	7	3.61	4.43
Hymenoptera (other)	2	1.27	28.22	0.25	2	1.03	0.85
Insect Larvae	2	1.27	46.67	0.42	2	1.03	0.90
Neuroptera	1	0.63	106.28	0.95	1	0.52	0.70
Orthoptera	13	8.23	2,004.88	17.88	13	6.70	10.93
Pseudoscorpiones	1	0.63	2.96	0.03	1	0.52	0.39
Scorpiones	2	1.27	74.34	0.66	2	1.03	0.99
Thysanoptera	1	0.63	0.00	0.00	1	0.52	0.38
Unidentified	17	10.76	519.00	4.63	18	9.28	8.22

May and 13 June 2007. Time spent searching averaged 1.31 ± 0.53 h per searcher. Upon capture all individuals were placed in cloth bags and transported to a field laboratory where they were photographed and sex was determined. Snout-vent length (SVL), tibia-fibula length (TFL), jaw length (JL) and jaw width (JW), and mass were measured. Jaw length was the straight-line measurement from the quadratojugal joint to the mandibular symphysis; JW was straight-line measurement between the quadratojugal joints.

Frogs were fixed in 10% buffered formalin, and entire gastrointestinal tracts were excised and stored in 70% ethanol prior to dissection. All specimens and stomachs are housed at the Texas Cooperative Wildlife Collection, Texas A&M University, College Station, Texas. Prey items were identified to taxonomic order (family level where possible) following Triplehorn and Johnson (2005). Body length (head to thorax) and width (at widest centrally located section of body) of each prey item were measured with digital calipers to the nearest 0.01 mm. The volume of each prey item was estimated using the formula for a prolate spheroid (Vitt et al., 1993). Percent by volume (V), frequency of occurrence (F), and percent composition (N) were summed and divided by three to compute an importance quotient (I) for each prey category (Biavati et al., 2004). This quotient provides a more informed assessment of prey consumption than any of the three components alone. We also compared dietary breadth (Simpson, 1949) of this population to that of previously studied populations mentioned above. Dietary breadth was calculated according to the formula:

$$\beta = \frac{1}{\sum p_i^2},$$

where i is the prey category and p is the proportion of prey in category i .

We tested the null hypothesis of no difference in diet between males and females in our study population to evaluate potential for dietary niche partitioning between sexes. We compared both the symmetrical dietary overlap of males and females (Pianka, 1974) and the asymmetrical overlap of

MacArthur and Levins (1967) comparing males to females and females to males. Furthermore, we compared the occurrence, length, volume, and maximum volume of prey consumed between males and females with ANOVA (Zar, 1999). When necessary, data were log-transformed to homogenize group variances and meet assumptions of normality.

We used linear regression to test for relationships between frog size (SVL, mass, JW, and JL) and prey size and for size-adjusted JW and JL and prey size. To remove the effects of body size, data were natural log-transformed; then $\ln JW$ and $\ln JL$ were regressed against $\ln SVL$. The residuals were used as size-adjusted JW and JL. All statistical tests, transformations, and simulations were conducted in Microsoft Excel 2003 using the add-in PopTools version 3.0, build 3 (<http://www.cse.csiro.au/poptools/>). All means are reported ± 1 SD, and we determined a relationship to be significant if P -values were <0.05 .

RESULTS

Less than 10% (10) of the 105 *H. cinerea* stomachs were empty, and a total of 194 prey items were identified. Prey items represented 13 unique categories (Table 1). The mean number of prey items per individual was 1.85 ± 1.43 (range: 0–8). Mean prey length was 6.67 ± 2.86 mm (range: 1.9–24.05), and average prey volume was 56.47 ± 86.17 mm³ (range: 0.16–610.00). The dietary breadth of *H. cinerea* in this semi-isolated wetland were 5.11, compared to 9.13 from the Okefenokee, Georgia, population, 4.08 and 5.94 from the Gainesville, Florida, samples, and 5.38 from the Everglades, Florida (Table 2). The most frequently occurring prey items were beetles, ants, cockroaches, and spiders, which comprised 70.3% of the total number of prey items. Cockroaches, beetles, and grasshoppers and crickets accounted for 80.3% of the total volume of prey. The most important (I) items were beetles, cockroaches, grasshoppers and crickets, ants, and spiders (Table 1), which accounted for 82.6% of this value. Beetles from the family Coccinellidae (ladybugs) were the most prevalent for the order Coleoptera (59.0%), and most cockroaches were from the family Blattellidae (68.0%).

TABLE 2. Comparison of dietary breadth and occurrence of prey items in *Hyla cinerea* from locations in North America.

	Location					
	Status:	BBNP, TX Current Study Nonnative	Everglades, FL (Meshaka, 2001) Native	Gainesville, FL (Freed, 1982) Native	Gainesville, FL (Kilby, 1945) Native	Okefenokee Swamp, GA (Haber, 1926) Native
Niche breadth		5.11	5.39	5.94	4.08	9.13
No. frogs sampled		105	130	163	497	100
Prey categories						
Acari		—	—	—	0.6	—
Araneae		12.0	7.7	14.1	36.9	24.0
Blattodea		13.3	1.5	4.9	—	11.0
Chilopoda		—	—	—	0.2	1.0
Coleoptera		31.0	32.3	18.4	13.0	24.0
Collembola		—	—	—	0.2	—
Dermaptera		—	—	—	—	1.0
Diplopoda		—	—	—	0.6	—
Diptera		0.6	13.1	—	66.3	6.0
Hemiptera		4.4	15.4	14.7	6.4	16.0
Hymenoptera (Formicidae)		13.9	—	7.4	10.3	12.0
Hymenoptera (other)		1.3	10.0	1.8	2.0	3.0
Insect larvae		1.3	—	32.5	8.1	—
Isopoda		—	2.3	—	—	—
Isoptera		—	0.8	—	—	—
Lepidoptera		—	5.4	5.5	2.6	21.0
Mantodea		—	—	—	—	1.0
Mollusca		—	0.8	4.9	0.2	—
Neuroptera		0.6	—	—	0.6	1.0
Notostraca		—	—	—	—	1.0
Odonata		—	0.8	—	4.6	7.0
Opiliones		—	—	—	0.4	2.0
Orthoptera		8.2	3.8	3.7	4.8	12.0
Psocoptera		—	—	—	1.0	—
Pseudoscorpiones		0.6	—	—	0.2	1.0
Scorpiones		1.3	—	—	—	—
Thysanoptera		1.0	—	—	—	2.0
Thysanura		—	0.8	—	—	—
Trichoptera		—	—	—	—	1.0
Categories		13	14	10	19	19

There were no significant relationships between frog size (SVL, mass) and prey size (SVL: $P = 0.21$, $r^2 < 0.01$; mass: $P = 0.36$, $r^2 < 0.01$). There was no pattern of increasing prey size with increasing JW or JL (JW: $P = 0.66$, $r^2 < 0.01$; JL: $P < 0.01$, $r^2 = 0.07$). The regression between JL and prey volume was statistically significant, but the trend was for slightly decreasing prey volume with JL, and less than 10% of variance was explained by the relationship (JW $P = 0.64$, $r^2 < 0.01$; JL $P < 0.01$, $r^2 = 0.10$). Similarly, when SVL was taken into account, the regression between size-adjusted lnJL and ln prey volume was statistically significant, but the trend was weak and opposite the predicted direction ($P < 0.01$, $r^2 = 0.09$). The regression between size adjusted lnJW and ln prey volume was not significant ($P = 0.73$, $r^2 < 0.01$). Thus, it appears that frogs of all sizes were consuming a wide variety of prey volumes.

Females ate significantly longer prey and larger prey items by volume than did males (prey length: $F_{1,202} = 501.55$, $P < 0.001$; female: 7.96 ± 5.17 , male:

6.42 ± 4.18 ; prey volume: $F_{1,202} = 26.76$, $P < 0.001$; female: 89.75 ± 105.27 , male: 50.04 ± 80.77). The maximum sized prey of females was also larger than that of males ($F_{1,93} = 5.47$, $P = 0.02$; female: 160.94 ± 137.32 , male: 83.11 ± 102.78).

Symmetrical niche overlap between males and females was 0.88 by occurrence and 0.95 by volume. Asymmetrical overlap comparing males to females was 1.06 and 1.07 (occurrence, volume), and comparing females to males was 0.74 and 0.85 (occurrence, volume). These results indicate males consumed all prey categories found in females' stomachs, and females consumed most of the categories consumed by males. There was no significant difference in the number of prey items consumed between males and females ($F_{1,103} = 0.83$, $P = 0.36$; male: 1.81 ± 1.37 , female: 2.07 ± 1.75). These data suggest similar foraging and prey composition by males and females, with females consuming slightly larger prey in accordance with their larger body size.

DISCUSSION

Fourteen prey categories consisting of 11 taxonomic orders in two classes of Arthropoda were identified in the gut contents of *H. cinerea* in this new location. This is also the first report of the order Scorpiones in the diet of *H. cinerea*, which demonstrates this invader's ability to exploit novel resources outside of its native range. Beetles, cockroaches, grasshoppers and crickets, ants, and spiders were the most important prey items. Beetles and cockroaches combined accounted for more than 50% of the volumetric consumption and nearly 50% of the numerical consumption. This introduced population appears to have a relatively similar dietary breadth to native populations that have been studied, with notable differences in diet composition (Table 2). All populations demonstrate the consumption of spiders, beetles, ants, and grasshoppers and crickets (Table 2). The narrowest dietary breadth for *H. cinerea* was found by Kilby (1945) in Gainesville, Florida, where the most prevalent gut contents from 497 *H. cinerea* were Dipterans (flies) and "Araneidae" (Aranea: spiders), which together were found in more than 90% of the stomachs analyzed. In contrast, another Gainesville, Florida, sample studied by Freed (1982) of 163 *H. cinerea* had a dietary breadth more similar to the BBNP population but showed more even consumption of beetles, spiders, and both moth and beetle larvae. In a sample of 130 individuals from southern Everglades of Florida Meshaka (2001) reported lower occurrence of spiders and a greater prevalence of flies, true bugs, and beetles. The broadest diet for *H. cinerea* was found by Haber (1926) in the Okefenokee Swamp of Georgia, where frogs consumed nearly similar proportions of beetles, spiders, ants, true bugs, and grasshoppers and crickets. These data appear to agree with the findings of Brown (1974) who reported *H. cinerea* feeding primarily on "Cicadellidae (leafhoppers), Acrididae (grasshoppers), Lepidoptera larvae, Chrysomelidae and Arachnida (spiders)."

This dietary analysis conducted on *H. cinerea* from BBNP does not indicate predation is likely to be impacting (sensu Parker et al., 1999) native invertebrate prey populations or the endemic, syntopic, *G. gaigei*. However, exotic fish (*Lepomis cyanellus*) and nonnative American Bullfrogs (*Lithobates catesbeiana*) are also present in these ponds in BBNP. It is possible that this invasion of *H. cinerea* may facilitate persistence of other invasive species, by reducing predation on them or serving as prey for new invasives (Simberloff and Von Holle, 1999). Our study is the first dietary analysis of a nonnative population of *H. cinerea* and a first step in understanding how this species is interacting with native invertebrates in this Chihuahuan Desert wetland. Future research on direct and indirect interactions among the larval and adult *H. cinerea* with both native and nonnative inhabitants of this area will help complete the picture of what impacts are caused by this nonnative species in this habitat.

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LITERATURE CITED

- BIAVATI, G. M., H. C. WIEDERHECKER, AND G. R. COLLI. 2004. Diet of *Epipedobates flavopictus* (Anura: Dendrobatidae) in a Neotropical savanna. *Journal of Herpetology* 38:510–518.
- BROWN, R. L. 1974. Diets and habitat preferences of selected anurans in southeast Arkansas. *American Midland Naturalist* 91:468–473.
- BYERS, J. E., S. REICHARD, J. M. RANDALL, I. M. PARKER, C. S. SMITH, W. M. LONSDALE, I. A. E. ATKINSON, T. R. SEASTEDT, M. WILLIAMSON, E. CHORNESEY, AND D. HAYES. 2002. Directing research to reduce the impacts of nonindigenous species. *Conservation Biology* 16:630–640.
- ENSERINK, M. 1999. Biological invaders sweep in. *Science* 286:1091–1091.
- FREED, A. N. 1982. A treefrog's menu: selection for an evening's meal. *Oecologia* 53:20–26.
- FRITTS, T. H., AND G. H. RODDA. 1998. The role of introduced species in the degradation of island ecosystems: a case history of Guam. *Annual Review of Ecology and Systematics* 29:113–140.
- HABER, V. R. 1926. The food of the Carolina Treefrog, *Hyla cinerea* Schneider. *Journal of Comparative Psychology* 6:189–219.
- HUBBS, C., AND H. J. BRODRICK. 1963. Current abundance of *Gambusia gaigei*, an endangered fish species. *Southwestern Naturalist* 8:46–48.
- HUBBS, C., R. J. EDWARDS, AND G. P. GARRETT. 2002. Threatened fishes of the world: *Gambusia gaigei* Hubbs, 1929 (Poeciliidae). *Environmental Biology of Fishes* 65:82–82.
- KILBY, J. D. 1945. A biological analysis on the food and feeding habits of two frogs, *Hyla cinerea cinerea* and *Rana pipiens sphenoccephala*. *Quarterly Journal of the Florida Academy of Sciences* 8:71–104.
- LEAVITT, D. J., T. C. MULLET, C. M. RITZI, AND J. R. SKILES. 2007. Geographic distribution. *Hyla cinerea* (Green Treefrog). *Herpetological Review* 38:97.
- LOVEL, G. L. 1997. Biodiversity: global change through invasion. *Nature* 388:627–628.
- MACARTHUR, R., AND R. LEVINS. 1967. Limiting similarity convergence and divergence of coexisting species. *American Naturalist* 101:377–385.
- MESHAKA, W. E., JR. 2001. The Cuban Treefrog in Florida: Life History of a Successful Colonizing Species. University Press of Florida, Gainesville.
- MINCKLEY, W. L., AND J. E. DEACON. 1968. Southwestern fishes and enigma of endangered species. *Science* 159:1424–1432.
- PAINE, R. T. 1980. Food webs: linkage, interaction strength and community infrastructure. *Journal of Animal Ecology* 49:667–685.
- PARKER, I. M., D. SIMBERLOFF, W. M. LONSDALE, K. GOODELL, M. WONHAM, P. M. KAREIVA, M. H. WILLIAMSON, B. VON HOLLE, P. B. MOYLE, J. E. BYERS, AND L. GOLDWASSER. 1999. Impact: toward a framework for understanding the ecological effects of invaders. *Biological Invasions* 1:3–19.

- PHILLIPS, B. L., G. P. BROWN, AND R. SHINE. 2003. Assessing the potential impact of Cane Toads on Australian snakes. *Conservation Biology* 17:1738–1747.
- PIANKA, E. R. 1974. Niche overlap and diffuse competition. *Proceedings of the National Academy of Sciences of the United States of America* 71:2141–2145.
- PLATT, S. G., K. R. RUSSELL, W. E. SNYDER, AND L. W. FONTENOT. 1999. Distribution and conservation status of selected amphibians and reptiles in the Piedmont of South Carolina. *Journal of the Elisha Mitchell Scientific Society* 115:8–19.
- REDMER, M., L. E. BROWN, AND R. A. BRANDON. 1999. Natural history of the Bird-Voiced Treefrog (*Hyla avivoca*) and Green Treefrog (*Hyla cinerea*) in Southern Illinois. *Illinois Natural History Survey Bulletin* 36:37–66.
- SAENZ, D. 1996. Dietary overview of *Hemidactylus turcicus* with possible implications of food partitioning. *Journal of Herpetology* 30:461–466.
- SIMBERLOFF, D., AND B. VON HOLLE. 1999. Positive interactions of nonindigenous species: invasional meltdown? *Biological Invasions* 1:21–32.
- SIMPSON, E. H. 1949. Measurement of diversity. *Nature* 163:688.
- TINER, R. W. 2003. Geographically isolated wetlands of the United States. *Wetlands* 23:494–516.
- TRIPLEHORN, C. A., AND N. F. JOHNSON. 2005. Borror and Delong's Introduction to the Study of Insects. Thomson Learning, Inc., Belmont, CA.
- VITOUSEK, P. M., C. M. D'ANTONIO, L. L. LOOPE, M. REJMANEK, AND R. WESTBROOKS. 1997. Introduced species: a significant component of human-caused global change. *New Zealand Journal of Ecology* 21:1–16.
- VITT, L. J., P. A. ZANI, J. P. CALDWELL, AND R. D. DURTSCHKE. 1993. Ecology of the Whiptail Lizard *Cnemidophorus deppii* on a tropical beach. *Canadian Journal of Zoology* 71:2391–2400.
- WILES, G. J., J. BART, R. E. BECK, AND C. F. AGUON. 2003. Impacts of the Brown Tree Snake: patterns of decline and species persistence in Guam's avifauna. *Conservation Biology* 17:1350–1360.
- ZAR, J. H. 1999. *Biostatistical Analysis*. Prentice Hall, Upper Saddle River, NJ.
- ZERBA, K. E., AND J. P. COLLINS. 1992. Spatial heterogeneity and individual variation in diet of an aquatic top predator. *Ecology* 73:268–279.

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