

Currently, *Hyla nana* and *H. sanborni* are placed in the *H. microcephala* group (Klappenbach and Langone, 1992). Langone and Basso (1987) showed that the tadpole of *H. nana*, described by Bokermann (1963) and Cei (1980), is actually *H. sanborni*. The tadpole of *H. rubicundula* is similar to *H. nana* described by Lavilla (1990) and to *H. sanborni*, described by Bokermann (1963). The three species have (1) reduced oral disc without papillae, denticles, and dermal ridges; (2) similar position and type of spiracle; and (3) tail with flagelliform tip. Furthermore, *H. rubicundula* resembles *H. nana* in having a violin-shaped body and a modified oral disc. The oral disc is modified into a protrusible tube in both species, but it is longer in *H. nana*. *Hyla rubicundula* and *H. sanborni* are similar in color pattern and in having elliptical nostrils (Bokermann, 1963). The tadpole of *H. rubicundula* differs from those of *H. sanborni* and *H. nana* by having (1) a larger body, about 34% of total length (27% in *H. nana*, 25% in *H. sanborni*); (2) larger nostrils, about 29% of eye diameter (18% in *H. nana*); (3) conspicuously pigmented tail; and (4) homogenous ventral coloration (with sparkle dark spots in *H. sanborni*; see Bokermann, 1963; Lavilla, 1990). The morphological similarity among tadpoles of *H. rubicundula*, *H. nana*, and *H. sanborni* may suggest a close relationship as proposed by Lutz (1973).

*Acknowledgments.*—We are grateful to C. A. G. Cruz, W. E. Duellman, R. Fernandes, C. J. E. Lamas, G. Mejdalani, and H. Wogel for critically reviewing the manuscript; the Laboratório de Diptera from Museu Nacional/UFRJ for lending the micrometer ocular; C. J. E. Lamas for assistance in fieldwork; P. R. Nascimento for the help with illustrations; and CNPq, FAPERJ, and FUJB for the financial support.

#### LITERATURE CITED

- ALTIG, R. 1970. A key to the tadpoles of the continental United States and Canada. *Herpetologica* 26:180–207.
- ALTIG, R., AND R. W. McDIARMID. 1999. Body plan. Development and morphology. In R. W. McDiarmid and R. Altig (eds.), *Tadpoles: The Biology of Anuran Larvae*, pp. 24–51. Univ. of Chicago Press, Chicago.
- BOKERMANN, W. C. A. 1963. Girinos de anfíbios brasileiros I (Amphibia: Salientia). *An. Acad. Bras. Ciênc.* 35:465–474.
- CEI, J. M. 1980. Amphibians of Argentina. *Monitore Zoologico Italiano (N.S.)*, Monografia 2:1–609.
- DUELLMAN, W. E., AND M. J. FOUQUETTE JR. 1968. Middle American frogs of the *Hyla microcephala* group. *Univ. Kans. Publ. Mus. Nat. Hist.* 17: 517–557.
- DUELLMAN, W. E., AND L. TRUEB. 1983. Frogs of the *Hyla columbiana* group: taxonomy and phylogenetic relationships. In A. G. J. Rhodin and K. Miyata (eds.), *Advances in Herpetology and Evolutionary Biology*, pp. 33–51. Museum of Comparative Zoology, Harvard Univ., Cambridge, MA.
- FROST, D. R. 1985. *Amphibian Species of the World. A Taxonomic and Geographical Reference*. Allen Press, Inc., Lawrence, KS.
- GOSNER, K. L. 1960. A simplified table for staging anuran embryos and larvae with notes on identification. *Herpetologica* 16:183–190.
- KLAPPENBACH, M. A., AND J. A. LANGONE. 1992. Lista sistemática y sinonímica de los anfibios del Uruguay con comentarios y notas sobre su distribución. *An. Mus. Nac. Hist. Nat. Montevideo* 8:163–222.
- LANGONE, J., AND N. G. BASSO. 1987. Distribución geográfica y sinonimia de *Hyla nana* Boulenger, 1889 y de *Hyla sanborni* Schmidt, 1944 (Anura: Hylidae) y observaciones sobre formas afines. *Com. Zool. Mus. Hist. Nat. Montevideo* 11:1–17.
- LAVILLA, E. O. 1990. The tadpole of *Hyla nana* (Anura: Hylidae). *J. Herpetol.* 24:207–209.
- LUTZ, B. 1973. Brazilian species of *Hyla*. Univ. of Texas Press, Austin.
- NAPOLI, M. F., AND U. CARAMASCHI. 1998. Duas novas espécies de *Hyla* Laurenti, 1768 do Brasil Central afins de *H. tritaeniata* Bokermann, 1965 (Amphibia, Anura, Hylidae). *Bol. Mus. Nac. Nova Série, Zool. RJ* 391:1–12.
- . 1999a. Geographic variation on *Hyla rubicundula* and *Hyla anataliasiasi*, with the description of a new species (Anura, Hylidae). *Alytes* 16:165–189.
- . 1999b. The taxonomic status of *Hyla elongata* Lutz, 1925. *J. Herpetol.* 33: 484–487.
- PUGLIESE, A., A. C. R. ALVES, AND S. P. CARVALHO E SILVA. 2000. The tadpoles of *Hyla oliveirai* and *Hyla decipiens* with notes on the *Hyla microcephala* group (Anura, Hylidae). *Alytes* 18: 34–41.
- WILD, E. R. 1992. The tadpoles of *Hyla fasciata* and *H. allenorum*, with a key to the tadpoles of the *Hyla parviceps* group (Anura: Hylidae). *Herpetologica*, 48: 439–447.

Accepted: 8 March 2001.

*Journal of Herpetology*, Vol. 35, No. 4, pp. 688–693, 2001  
Copyright 2001 Society for the Study of Amphibians and Reptiles

### Analysis of Sympatric Populations of *Lampropeltis triangulum sypbila* and *Lampropeltis triangulum elapsoides*, in Western Kentucky and Adjacent Tennessee with Relation to the Taxonomic Status of the Scarlet Kingsnake

MICHAEL P. ARMSTRONG,<sup>1</sup> DAVID FRYMIRE,<sup>2</sup> AND EDMUND J. ZIMMERER<sup>3,4</sup>

<sup>1</sup>U.S. Fish and Wildlife Service, 711 Stadium Drive, Suite 252 Arlington, Texas 76011, USA

<sup>2</sup>318 Dogwood Court, Versailles, Kentucky 40383, USA

<sup>3</sup>Department of Biological Sciences, Murray State University, P.O. Box 9, Murray, Kentucky 42071, USA

The *Lampropeltis triangulum* species complex ranges from portions of northern South America through Central America, most of the continental United States and into southeastern Canada. It is a variable group,

<sup>4</sup> Corresponding Author.

both in size and color pattern (for a review of the taxonomy and distribution, see Williams, 1978, 1988). In the eastern U.S. one putative subspecies, the scarlet kingsnake, *Lampropeltis triangulum elapsoides*, differs more in certain characters than perhaps any other member of this group. It is sympatric, or intergrades, with the eastern milk snake, *Lampropeltis triangulum triangulum* (portions of Virginia, North Carolina, South Carolina, eastern Kentucky, northern Georgia, and northern Alabama), the red milk snake, *Lampropeltis triangulum sypila* (western Kentucky, northwest Tennessee, and northern Mississippi), and the Louisiana milk snake, *Lampropeltis triangulum amaura*, in Louisiana (Williams, 1978, 1988; Collins and Hirschfeld, 1964). *Lampropeltis triangulum elapsoides* differs significantly from all three of these subspecies in size, color/band/ring patterns, head shape, and scutellation. Along the East Coast, it presumably intergrades with *L. t. triangulum* to produce an intermediate form once called the coastal plains milk snake, *Lampropeltis triangulum temporalis*. However, the latter is no longer recognized as a valid subspecies (Williams, 1988). In other areas, including the middle and upper Tennessee River Valley of Kentucky and Tennessee, the Cumberland Plateau of Kentucky, the Great Smoky Mountains, and western North Carolina, *L. t. elapsoides* occurs sympatrically with *L. t. triangulum* with little or no intergradation. According to Williams (1988), *L. t. elapsoides* intergrades with *L. t. sypila* in a broad band from northwestern Tennessee, through western Kentucky, and into southern Indiana. He also cites tentative intergrades of *L. t. sypila* × *L. t. elapsoides* in Mississippi.

The sympatric occurrence of *L. t. elapsoides* over much of its peripheral range with *L. t. triangulum*, and the presence of zones of intergradation in other areas, along with presumed intergradation with *L. t. sypila* and *L. t. amaura* in the central and western portions of its range pose questions regarding the taxonomic status of *L. t. elapsoides*. Such questions can best be resolved when confusion is eliminated regarding areas of population overlap where little is known about the nature of the interaction (sympatry or intergradation). This confusion results from the variable nature of intergrades, and from the fact that judgments are often based solely on head and belly patterns, traits that are highly variable in *L. t. triangulum* and *L. t. sypila* and perhaps more variable in *L. t. elapsoides* than commonly assumed. Furthermore, the determination as to whether the overlap represents intergradation or sympatry is often based on only a few specimens. In the following report, we present an in-depth analysis of a previously reported (Williams, 1988) intergrade zone in western Kentucky with evidence that this area does in fact represent two populations in sympatry.

Data for 27 pattern and meristic characters were obtained from reference samples of two subspecies of *Lampropeltis triangulum*: *L. t. sypila* ( $N = 16$ ) from Missouri and Kansas, and *L. t. elapsoides* ( $N = 23$ ) from South Carolina, Georgia, Florida and Mississippi. These data were compared to those obtained from specimens of *L. triangulum* from far western Kentucky and adjacent Tennessee ( $N = 63$ ). Specimens examined in this study either were collected in the field or came from institutional collections. Collection sites

and additional information for all specimens examined for this study are provided in Figure 1 and Appendix 1.

The set of characters used in this analysis includes those used by Williams (1978, 1988) and Conant and Collins (1998). These include sex, head pattern, total length, tail length, and the number of ventral scales, subcaudals, anterior dorsal scale rows (A), midbody dorsal scale rows (B), dorsal scale rows anterior to the vent (C), total blotch/rings, supralabials, infralabials, preoculars, postoculars, loreals, and temporal scales. Two other characters, blotch drop and number of rings behind the vent were added to the set. Blotch drop was measured as the length, in number of scale widths, that a blotch extends into the ventrals of a specimen. It was measured as a means to quantify as one character whether a specimen had blotches, rings, or an intermediate condition. A single blotch or ring was defined as red bordered by black separated from the next by white yellow or gray. Tail bands were sometimes missing the red pigment. Dorsal scale row counts were taken at three points along the body: one-half head length behind the nape (A), at midbody (B), and one head length in front of the vent (C). The number of rings found posterior to the vent was measured from the first ring behind the vent to the tip of the tail. Sex of each specimen was determined by visual inspection of everted hemipenes, or by relative length and basal taper of the tail.

General descriptive statistics were produced using the Statistical Analysis System (SAS) statistical package (SAS Institute, Inc., Cary, NC, 1990). The univariate procedure was used to compute simple statistics such as mean, standard deviation, variance, range, coefficient of variation, standard error, and minimum and maximum values for 18 of the 27 characters for each group. A matrix of character correlation was constructed using Pearson's product-moment correlation coefficients and used to assess redundancy of information and character associations.

Because sample size for some groups was relatively small, sexually dimorphic traits were eliminated from further analysis to avoid the reduction in statistical strength by separating groups by sex. Characters lacking variation within or between groups were also eliminated from further analysis at this time.

Principal component analysis was used to examine relationships among the remaining quantitative variables. This analysis was performed by calculating the loadings on each component from the remaining variables. Multivariate analysis of variance (MANOVA) was then applied to the new dataset of eigenvalues to determine which of the principal components were statistically significant to the groups. Where statistical significance was observed, Tukey tests were performed to identify whether Tukey groupings correspond to the four recognized groups of *L. triangulum*. Relationships between the four groups of *L. triangulum* were further examined by canonical discriminant-function analysis.

Summary statistics for 18 characters are given in Table 1. Of these, 15 characters showed significant differences between the reference *L. t. sypila* and *L. t. elapsoides* ( $t$ -test, 0.001 significance level). Only supralabials, preoculars, and postoculars showed no significant difference between the two reference groups and

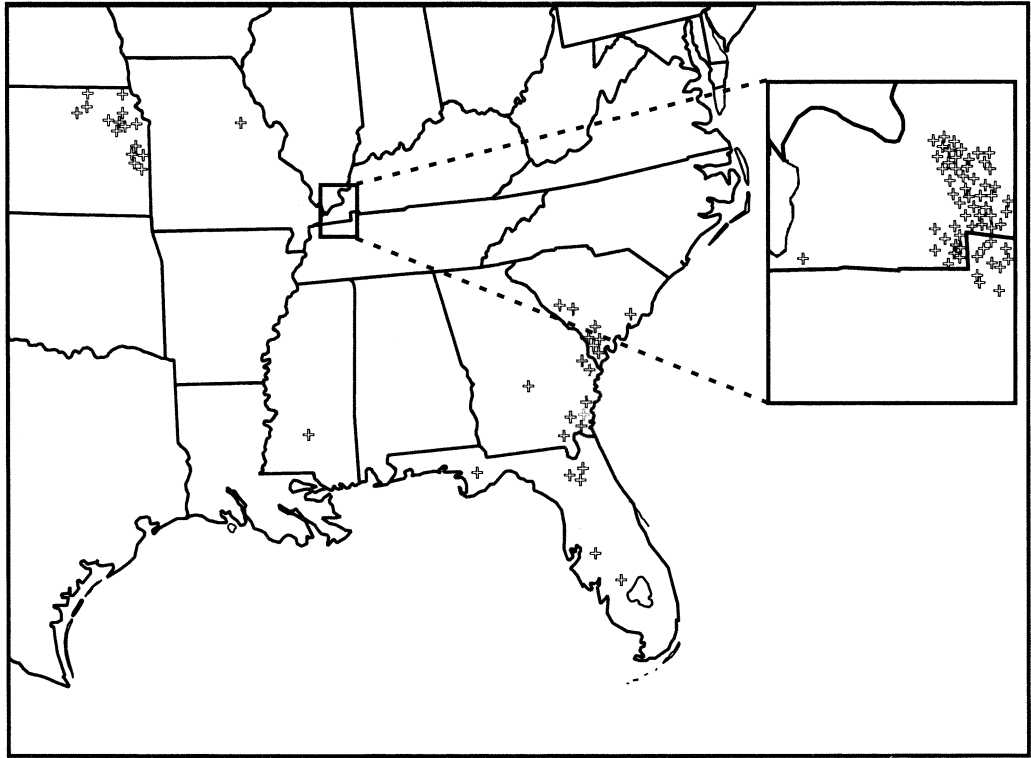


FIG. 1. Map of the eastern United States showing collection localities (marked with crosses) for *Lampropeltis* specimens analyzed in this study.

TABLE 1. Summary statistics for 18 characters from Kentucky *Lampropeltis triangulum* group 1 (*Lampropeltis triangulum sypila*), group 2 (*Lampropeltis triangulum elapsoides*), and reference specimens.

Character	Reference <i>L. t. elapsoides</i>			Kentucky <i>L. t. elapsoides</i>			Reference <i>L. t. sypila</i>			Kentucky <i>L. t. sypila</i>		
	N	$\bar{x}$	SD	N	$\bar{x}$	SD	N	$\bar{x}$	SD	N	$\bar{x}$	SD
Total length	23	364.8	89.6	17	372.3	107.1	16	539.9	168.1	47	605.5	206.5
Tail length	23	53.6	15.5	17	50.4	16.5	16	72.9	19.0	47	77.0	27.3
Ventral scales	23	172.6	4.6	17	176.9	2.4	16	205.9	5.8	47	206.0	45.1
Subcaudal scales	23	39.6	4.4	17	37.4	5.2	16	47.0	5.2	47	45.1	6.6
Dorsal scale rows A	23	17.1	0.4	17	17.4	0.8	16	22.6	0.8	47	21.2	0.6
Dorsal scale rows B	23	19.0	0	17	18.8	1.0	16	21.0	0	47	20.2	0.4
Dorsal scale rows C	23	17.0	0	17	17.0	0	16	18.6	0.8	47	17.9	0.9
Rings/blotch ant. to cloaca	23	20.3	2.1	17	18.3	2.6	16	28.4	4.2	47	32.5	4.9
Blotch drop A	23	-4.78	0.73	17	-3.2	2.52	16	-2.03	2.81	47	0.96	1.62
Blotch drop B	23	-5.00	0	17	-4.66	0.89	16	-0.28	2.59	47	0.83	1.10
Blotch drop C	23	-5.00	0	17	-5.00	0	16	0.31	1.86	47	0.23	1.28
Rings post. to cloaca	23	3.87	0.81	17	4.08	0.9	16	5.06	1.24	47	5.57	1.55
Supralabials	23	7.00	0	17	7.00	0	16	7.00	0	47	7.00	0
Infralabials	23	8.13	0.31	17	8.25	0.45	16	8.81	0.36	47	8.87	0.14
Preoculars	23	1.00	0	17	1.00	0	16	1.00	0	47	1.00	0
Postoculars	23	2.00	0	17	2.00	0	16	2.00	0	47	1.98	0.15
Loreals	23	0.56	0.48	17	0.50	0.52	16	1.00	0	47	0.98	0.15
Temporals	23	1.04	0.14	17	1.29	0.40	16	1.84	0.35	47	1.79	0.40

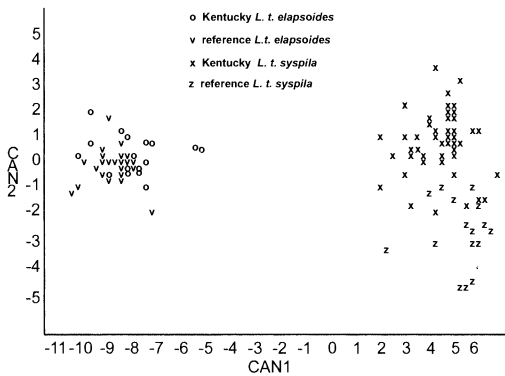


FIG. 2. Plot of canonical coefficients for western Kentucky/Tennessee *Lamppropeltis triangulum* specimens (o and x) as unknown groups against reference *Lamppropeltis triangulum elapsoides* (v) and *Lamppropeltis triangulum sypila* (z).

were, therefore, eliminated from further analysis. Total length, tail length, and the number of subcaudals were also eliminated from further analysis because they were sexually dimorphic and may introduce an ontogenetic bias. Head patterns for the Kentucky and Tennessee specimens were so variable as to be of little use in delineating groups.

The first three principal components explained 72.4% of the total variation represented in the 13 characters; thus there was little loss of information in the reduction of 13 dimensions to three. Factor 1 was most strongly influenced by 10 characters: ventral number, dorsal scale row A and B, blotch/ring number, blotch drop A, B, and C, infralabial number, loreal number, and temporal number. Factor 2 expressed an additional 8.9% of the total variability in the character set, whereas factor 3 accounted for another 7.2% of the total variability. Multivariate analysis of variance when applied to the new dataset with just three dimensions found only Factor 1 to be significant ( $r = 0.94$ ).

Our analysis illustrates how specimens of *L. triangulum* from western Kentucky and adjacent Tennessee fall into two groups. One group resembling typical *L. t. sypila* exhibits numbers that do not differ significantly from the reference *L. t. sypila* (Tukey's range test, 0.667 vs. 0.619). The second group, those that at first glance appear as either *L. t. elapsoides* or possible intergrades, reveals numbers that are similar to the reference *L. t. elapsoides* (Tukey's range test,  $-1.249$  vs.  $-1.359$ ). This becomes even more apparent when using canonical discriminant analysis to compare the four groups (reference *L. t. sypila*, reference *L. t. elapsoides*, group 1 KY (*sypila*), and group 2 KY (*elapsoides*)). Figure 2 clearly shows that the Kentucky *L. t. elapsoides* (group 2) overlaps with the reference *L. t. elapsoides* from Florida, Georgia, South Carolina, and Mississippi. The Kentucky and Tennessee *L. t. sypila* (group 1) overlaps the reference *L. t. sypila* from Kansas and Missouri.

Superficially, members of the *L. triangulum* species group in west Kentucky and adjacent Tennessee fall into at least three groups. The majority of individuals observed appear to be *L. t. sypila*, although variability

in blotch count, head pattern, and total length may indicate the influence of *L. t. triangulum* (for data on *L. t. triangulum*, see Williams, 1988). The second group appears to be *L. t. elapsoides*. A third group is similar to *L. t. elapsoides* from the dorsal view, but the ventral area is highly variable, ranging from no pattern (white), to broken rings, to checkering. It is likely members of this group that Williams (1978, 1988) observed and stated as intergrades between *L. t. sypila* and *L. t. elapsoides* and that we initially considered the same. It is these two latter groups that in our analysis comprise the group 2 KY (*elapsoides*).

Our putative intergrades, and possibly those mentioned by Williams (1988) were placed into that group based primarily on subjective judgments about rings/blotch and head patterns. In particular, those specimens that would have otherwise been deemed *L. t. elapsoides*, but for the rings that did not meet in the middorsal area, the presence of some ventral checkering, or no ventral pigment at all, were considered possible intergrades. Although these characters were quantified and entered in as part of our analysis (blotch drop), they were only one part of the analysis. All other characters strongly suggest that the "intergrade group" is identifiable as *L. t. elapsoides*. Although overall size was not used in our analysis to eliminate any ontogenetic bias, when the size of those that we were confident were mature was compared, these same two groups emerged, those within the reported size range (from Conant and Collins, 1998) of *L. t. elapsoides* (Conant and Collins range 360–510 mm, our data range 345–504 mm,  $N = 15$ ,  $\bar{x} = 372.5$  mm), and those significantly larger and within the size range typical of *L. t. sypila* (Conant and Collins range 530–710 mm, our data range 571–915 mm,  $N = 32$ ,  $\bar{x} = 712.8$  mm). The forgoing evidence suggests that there are just two groups of *L. triangulum* in western Kentucky and adjacent Tennessee, *L. t. sypila*, and *L. t. elapsoides*, which are sympatric with little or no gene flow. Although we cannot claim that there is no gene flow between these two populations, if there is, it must be minimal because the two groups appear to have maintained their integrity. If this were a true intergrade zone, we would have expected to see points of the canonical plot intermediate between the two reference groups.

We do not discount the possibility of some (current or past) gene flow between the two groups. The best evidence for this is the greater variability of ventral patterns of the Kentucky *L. t. elapsoides* when compared with the reference populations. However, we note that there is some variability in the ventral pattern of the reference groups as well. One specimen each from southern South Carolina and Mississippi has incomplete ventral rings, resulting in partially white bellies. Much of the ventral pattern variability in the western Kentucky population of *L. t. elapsoides* may therefore be explained as part of the natural variability of *L. t. elapsoides* with possible exaggeration resulting from being a peripheral and perhaps disjunct population. The observation that most of the *L. t. elapsoides* came from Lyon and northern Trigg counties of The Land Between The Lakes National Recreation Area, with only one from southern Trigg County, and none from adjacent areas of Tennessee, indicate that this population is disjunct, and may be several

hundred kilometers from either central Kentucky or Tennessee populations.

In regard to ventral pattern, Collins and Hirschfeld (1964) noted a similar phenomenon in regard to *L. t. elapsoides* from areas of sympatry with *L. t. triangulum* in central Kentucky. They noted that the specimens of *L. t. elapsoides* from these areas are typical except for their ventral markings. They further stated that these marking resembled those of *L. t. temporalis* × *L. t. elapsoides* (now *L. t. triangulum* × *L. t. elapsoides*, Williams, 1988). As with the western Kentucky population, it is unclear whether these ventral patterns are a result of limited gene flow.

The variability we see in head and body patterns weakens the value of these characters in determining subspecies status. Williams (1988) stated that his single most important character in determining subspecies differences in the *L. t. triangulum* complex is head pattern. This may be valid for western, Mexican or Central American subspecies, but only adds to the confusion in many areas east of the Mississippi River. In our area, ventral patterning is also highly variable and of limited use in delineating groups.

The sympatry observed in western Kentucky, as well as that observed by Collins and Hirschfeld, brings into question the validity of other putative zones of intergradation, particularly those of *L. t. elapsoides* × *L. t. amaura* or *L. t. amaura* × *L. t. sypila* in the western portion of the *L. t. elapsoides* range in Louisiana, and zones of intergradation in the central and western portions of Virginia and North Carolina between *L. t. triangulum* and *L. t. elapsoides*. Williams (1978, 1988) proposed that *L. t. triangulum* and *L. t. elapsoides* may be sympatric over the southwestern portions of the Piedmont in southwestern North Carolina, and the coastal plain of southern Virginia. He also stated that *L. t. elapsoides* from southeastern Virginia and northeastern North Carolina reflect a trend in several characters toward intergradation. These include a higher than overall mean ventral scale number for *L. t. elapsoides*, and red rings terminated by black pigment on the edge of the ventrals, leaving a midventral light area in most specimens from this locale. Although we do not see a significant increase in ventral scale counts in the western Kentucky *L. t. elapsoides*, the breakup of the ventral ring pattern seems similar to what we have observed.

A more recent and comprehensive dataset for Virginia is presented in the Reptiles of Virginia (Mitchell, 1994). Mitchell stated that *L. triangulum* in Virginia occurs in three color phases: the blotched phase, apparently typical of *L. t. triangulum*; a ringed phase, and intermediates. He further stated that the ringed and intermediate phases represent varying degrees of intergradation with *L. t. triangulum*. The only exceptions he claimed in Virginia are those found in extreme southeastern Virginia based solely on the existence of completely ringed specimens. However, based on that character, some individuals from as far south as southern South Carolina would not qualify as pure *L. t. elapsoides*. Examination of a photograph of a putative intergrade phase in Mitchell's book and summary data presented on ventral, subcaudal, and blotch/ring counts suggests to us that the situation in Virginia, may be similar to what we see in Kentucky. More data, particularly from the Southern Piedmont region of Virginia is needed.

In summary, we conclude that in western Kentucky,

*L. t. elapsoides* and *L. t. sypila* exist in sympatry, with minimal, if any, gene flow between these populations. Although variability in head and body patterns may indicate a degree of genetic exchange, both populations have maintained their integrity with regard to most other characters including size, ring/blotch counts, and scutellation. In areas where both are found, the habitat consists of hilly terrain of primarily mixed hardwoods. The pine flatwood habitat most often associated with *L. t. elapsoides* of the southeastern coastal plain does not exist naturally in this area. Although stands of introduced loblolly pine (*Pinus taeda*) have been planted, these are not where we find *L. t. elapsoides*. *Lampropeltis triangulum elapsoides* from our area, therefore, uses a different habitat than more southern populations of *L. t. elapsoides* and may differ in other niche aspects as well.

*Acknowledgments.*—We wish to thank J. Simmons, J. Collins, and F. Scott for their interest, support and generous access to the Museum of Natural History of the University of Kansas and the Austin Peay State University collection. We also wish to thank D. Auth and K. Krysko from the Florida State Museum at the University of Florida and M. Mills from the Savannah River Ecology Laboratory for the loan of preserved material. We finally wish to thank B. Pendley, C. Crockett, M. Hatfield, D. Reed, and the many other Murray State students who have spent many miles and hours on the back roads of western Kentucky and northwestern Tennessee. Portions of this project were supported by a grant from the Committee on Institutional Studies and Research at Murray State University.

#### LITERATURE CITED

- COLLINS, J. T., AND C. J. HIRSCHFELD. 1964. *Lampropeltis doliata doliata* (Linnaeus) in Kentucky. *Herpetologica*. 19:292–293.
- CONANT, R., AND J. T. COLLINS. 1998. A field guide to reptiles and amphibians of eastern and central North America. 3rd ed. Houghton Mifflin Co., Boston, MA.
- MITCHELL, J. C. 1994. The Reptiles of Virginia. Smithsonian Institution Press, Washington, DC.
- WILLIAMS, K. L. 1978. Systematics and Natural History of the American Milk Snake, *Lampropeltis triangulum*. Milwaukee Publ. Mus., Milwaukee, WI.
- . 1988. Systematics and Natural History of the American Milk Snake, *Lampropeltis triangulum*. Milwaukee Publ. Mus., Milwaukee, WI.

Accepted: 17 February 2001.

#### APPENDIX 1

*Lampropeltis* specimens analyzed in this study listed by museum number followed by the county and state from which they were collected. APSU = Austin Peay State University Museum, KU = University of Kansas Museum of Natural History, MUSU = Murray State University, SREL = Savannah River Ecology Lab., UF = University of Florida, Florida State Museum.

Reference *L. t. elapsoides*: MUSU 1549, Charlton, GA; MUSU 1550, Alachua, FL; SREL 627, Chatham, GA; SREL 669, Chatham, GA; SREL 1925, Copiah, Mississippi; SREL 2557, Aiken, SC; SREL 2560, Aiken, SC; SREL 3118, Jasper, SC; UF 4639, Camden, GA; UF 8867, Glades, FL; UF 62897, Alachua, FL; UF 67838, Jasper,

SC; UF 67839, Berkeley, SC; UF 68129, Liberty, FL; UF 74459, Hillsborough, FL; UF 111412, Glynn, GA; UF 111413, Glynn, GA; UF 111414, Glynn, GA; UF 111415, Crawford, GA; UF 111417, Jasper, SC; UF 111418, Jasper, SC; UF 111420, Jasper, SC; UF 111424, Duval, FL.

Reference *L. t. sypspila*: KU 84671, Douglas, KS, KU 82205, Linn, KS, KU 154033, Atchison, KS, KU 144804, Douglas, KS, KU 91606, Johnson, KS, KU 82206, Linn, KS, KU 148465, Douglas, KS, KU 155262, Pottawatomie, KS, KU 155260, Riley, KS, KU 145886, Douglas, KS, KU 84672, Douglas, KS, KU 82204, Linn, KS, KU 82207, Linn, KS, KU 157966, Marshall, KS, MUSU 1502, Warren, MO, MUSU 1503, Linn, KS.

Kentucky/Tennessee Specimens: APSU 00285, Stewart, TN; APSU 03508, Montgomery, TN; APSU 04512, Davidson, TN; APSU 02128, Montgomery, TN; APSU 05164, Montgomery, TN; APSU 00443, Trigg, KY; APSU 00547, Stewart, TN; APSU 00559, Stewart, TN; APSU 00555, Stewart, TN; APSU 05073, Stewart, TN; APSU 04746, Stewart, TN; APSU 05381, Lyon, KY; APSU 00540, Lyon, KY; KU 154194, Trigg, KY; KU 214461, Fulton, KY; MUSU 1357, Calloway, KY; MUSU 1404, Trigg, KY; MUSU 1457, Trigg, KY; MUSU 1458, Christian, KY; MUSU 1459, Lyon, KY; MUSU 1460, Trigg, KY; MUSU 1462, Lyon, KY; MUSU 1463, Lyon, KY; MUSU 1464, Trigg, KY; MUSU 1465, Calloway, KY; MUSU 1466, Trigg, KY; MUSU 1467, Lyon, KY; MUSU 1468, Lyon, KY; MUSU 1469, Lyon, KY; MUSU 1470, Lyon, KY; MUSU 1471, Trigg, KY; MUSU 1472, Trigg, KY; MUSU 1473, Trigg, KY; MUSU 1474, Trigg, KY; MUSU 1475, Trigg, KY; MUSU 1476, Lyon, KY; MUSU 1477, Trigg, KY; MUSU 1478, Calloway, KY; MUSU 1479, Calloway, KY; MUSU 1480, Lyon, KY; MUSU 1481, Calloway, KY; MUSU 1482, Calloway, KY; MUSU 1483, Calloway, KY; MUSU 1484, Calloway, KY; MUSU 1485, Calloway, KY; MUSU 1486, Calloway, KY; MUSU 1487, Montgomery, TN; MUSU 1488, Trigg, KY; MUSU 1489, Trigg, KY; MUSU 1490, Trigg, KY; MUSU 1491, Trigg, KY; MUSU 1492, Lyon, KY; MUSU 1493, Lyon, KY; MUSU 1494, Trigg, KY; MUSU 1495, Trigg, KY; MUSU 1496, Trigg, KY; MUSU 1497, Trigg, KY; MUSU 1498, Trigg, KY; MUSU 1523, Lyon, KY; MUSU 1524, Trigg, KY; MUSU 1525, Trigg, KY; MUSU 1526, Trigg, KY; MUSU 1527, Trigg, KY.

species into areas where they are not normally found can result in biotic disturbances to native ecosystems with concomitant adverse effects on native biota (Brockie, 1988; Crossland, 2000). For example, in the United States, various species of introduced fish and anurans have had detrimental effects on native amphibian populations (Lachner et al., 1970; Berven, 1990). The impact of introduced species on native fauna may, in some cases, be subtle and difficult to discern (Moyle, 1976; Howarth, 1991). In other instances, the effects may be profound, resulting in the altering of species composition and dynamics of native communities (Elton, 1958; Vitousek, 1990).

The giant (cane) toad, *Bufo marinus* is native to central and tropical South America (Zug and Zug, 1979) and was first introduced into Florida in 1936 in Palm Beach County as a control agent against insect pests of sugar cane (Lobdell, 1936; Krakauer, 1968). Since then, *B. marinus* has spread throughout southern Florida and has extended its range into south-central Florida (Butterfield et al., 1997). Adult *B. marinus* are generalist predators and will capture and ingest almost anything they can swallow including arthropods, other frogs, lizards, birds, and small mammals (Alexander, 1964; Zug and Zug, 1979). As a result, this toad can potentially contribute to the decline of local native amphibian populations (Covacevich and Archer, 1975; Crossland, 2000). In the 1930s, the giant toad was also introduced into other regions including the Philippines and Australia as a potential control agent against insect pests has become a widespread pest in these regions (Alcala, 1957; Cogger, 2000).

The eggs of some anuran species provide an important food source for a variety of aquatic organisms including gastropods, insects, fish, and tadpoles of other amphibians (Wassersug, 1971; Grubb, 1972; Zaret, 1980; Petranka et al., 1995; Crossland, 1998). However, some anuran eggs possess toxins that can adversely effect predators and may even cause death (Licht, 1967, 1968; Duellman and Trueb, 1986; Peterson and Blaustein, 1992; Crossland and Alford, 1998). Numerous species of *Bufo* are unpalatable or toxic, or both, as eggs, larvae, or adults (Licht, 1967, 1968; Wassersug, 1971; Kruse and Stone, 1984; Punzo, 1995).

Recent studies have shown that the eggs (from fertilization through hatching), as well as hatchlings and tadpoles, of *B. marinus* are toxic to oophagous native Australian tadpoles (Crossland, 1997; Crossland and Alford, 1998). The purpose of our study was to evaluate the toxicity of eggs of *B. marinus* to potential native aquatic predators in south-central Florida. We have been monitoring several small retention ponds in Hillsborough and Pinellas Counties for a number of years as part of a long-term study focusing on pond ecology, anuran behavior, and the effects of pollutants on local amphibian populations (Punzo, 1992, 1993, 1995, 1997). These small ponds are used as breeding sites in urban areas by many anuran species (Dodd, 1992; Punzo 1992, 1995), and in some cases, many if not all native amphibian populations have declined precipitously when *B. marinus* invades these ponds (unpubl. data).

We conducted a series of laboratory experiments to assess the toxicity of fertilized eggs of *B. marinus* to native aquatic predators. The fertilized eggs of several common aquatic invertebrate (insects, crayfish, and

*Journal of Herpetology*, Vol. 35, No. 4, pp. 693–697, 2001  
Copyright 2001 Society for the Study of Amphibians and Reptiles

## The Toxicity of Eggs of the Giant Toad, *Bufo marinus* to Aquatic Predators in a Florida Retention Pond

FRED PUNZO<sup>1</sup> AND LAURA LINDSTROM

Department of Biology, University of Tampa, Tampa, Florida 33606, USA

The accidental or deliberate introduction of exotic

<sup>1</sup> Corresponding Author. E-mail: fpunzo@alpha.utampa.edu