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A NEW UNISEXUAL SALAMANDER FROM OHIO

By Fred Kraus\*

ABSTRACT.—Kraus, Fred. 1984. A new unisexual salamander from Ohio. *Occ. Pap. Mus. Zool. Univ. Michigan*, 709: 1-24, figs. 1-11. A new unisexual salamander, *Ambystoma nothagenes*, is described and illustrated from Kelleys Island in Lake Erie, Ohio. Phenotypic characteristics of this species are compared to those of the congeners *A. laterale*, *A. texanum*, *A. tigrinum*, *A. jeffersonianum*, and *A. laterale-texanum* by means of principal components analyses and non-parametric univariate statistics. The new species is allotriploid and incorporates one genome from each of the bisexual species *A. laterale*, *A. texanum*, and *A. tigrinum*. *A. nothagenes* probably originated from a hybridization event involving a diploid *A. laterale-texanum* and an *A. tigrinum*, perhaps within the last 130 years.

Key words: Salamander, *Ambystoma*, unisexual, hybrid origin, polyploidy, Ohio.

INTRODUCTION

The genus *Ambystoma* comprises about 30 species ranging from Hudson Bay to the Central Plateau of Mexico. Unisexual taxa of hybrid origin occur in the Great Lakes region of the United States and Canada and were first described in detail by Uzzell (1964) who assigned the names *A. platineum* and *A. tremblayi* to two taxa formed by hybridization events between *A. laterale* and *A. jeffersonianum*. More recently, Downs (1978) has documented the existence of a third unisexual *Ambystoma* on the Bass Islands of northwestern Ohio which was formed by hybridization between *A. laterale* and *A. texanum*. These have been studied subsequently by Kraus (in press) on the Ohio and

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Michigan mainland and have been given the informal hyphenated name *A. laterale-texanum* in a manner similar to that used for the unisexual taxa of the fish genus *Poeciliopsis* (Schultz, 1969).

Of these three unisexual forms, *A. tremblayi* and *A. platineum* are triploid (Uzzell, 1964; Uzzell & Goldblatt, 1967), whereas *A. laterale-texanum* includes both diploids and triploids with the former predominating in roughly a 3:1 ratio (Kraus, in press). All of these forms are presumed to reproduce by some unisexual means. Uzzell (1964) and MacGregor & Uzzell (1964) have claimed that gynogenesis is operative in *A. tremblayi* and *A. platineum* but Bogart (1982) and Sessions (1982) presented data that are incongruent with the exclusive use of this mechanism, and I (Kraus, in press) have pointed out the insufficient documentation of this mechanism in these species. Morris and Brandon (1984) presented a more convincing case for a gynogenetic reproductive mechanism in unisexual hybrids between *A. texanum* and *A. platineum*. Kraus (in press) tentatively concludes that *A. laterale-texanum* reproduces by parthenogenesis; however, this too requires better documentation.

In the course of studying *A. laterale-texanum* in Ohio, I encountered a form of *Ambystoma* on Kelleys Island in Lake Erie, already alluded to by Kraus & Schuett (1982), that could not readily be assigned to any presently known taxon of the genus. In coloration, head shape, and build it appears to be intermediate between the species *A. texanum* and *A. tigrinum*. This, in combination with the fact that *Ambystoma* of hybrid origin are common in this geographic region, led me to investigate whether the Kelleys Is. form comprises another taxon of hybrid origin, presumably involving *A. texanum* and *A. tigrinum* as the parental species.

## MATERIALS AND METHODS

Nineteen specimens of this new form (hereafter referred to as "Kelleys Is. *Ambystoma*") were collected from 1980 to 1983 on Kelleys Is., Erie Co., Ohio. In an effort to elucidate its relationships, these were compared to preserved specimens of *Ambystoma laterale*, *jeffersonianum*, *texanum*, *laterale-texanum*, and *t. tigrinum* in The University of Michigan, Museum of Zoology (UMMZ). (Field series abbreviations are: FK—Fred Kraus, personal collection; RAN—Ronald A. Nussbaum, personal collection.) *A. opacum* was excluded from analysis because it is an autumnal breeder and, therefore, could not have been involved in a hybridization event leading to the formation of the new form from Kelleys Is. *A. maculatum* was excluded because hybrids between it and *texanum* and *tigrinum*, two species suspected to be involved in the formation of the new form, are inviable (Brandon, 1977). Additionally, both *opacum* and *maculatum* are either

rare or absent on the adjoining Ohio mainland and bear no particular resemblance to the new form.

The following meristic characters were studied on all specimens: standard length (snout to posterior angle of vent, to nearest 0.5 mm); tail length (when complete, to nearest 0.5 mm); internarial distance (to nearest 0.1 mm); snout length (to nearest 0.1 mm); canthal length (to nearest 0.1 mm); number of costal grooves between the limbs; number of costal folds between adpressed limbs; whether the plicae of the tongue radiated from the back of the tongue, from a median furrow, or had a distribution intermediate between these two extremes; number of maxillary-premaxillary teeth; number of rows of maxillary-premaxillary teeth; number of prevomerine teeth; number of patches of prevomerine teeth; and percentage of prevomerine teeth in lateral patches. The first two measurements were taken with a millimeter rule, the last three with an ocular micrometer fitted to a dissecting microscope.

In addition to this, five specimens of the new form were analyzed for 31 characters of the hyoid and associated throat musculature and 24 characters of cranial osteology and compared to the same species listed above. Of these characters, four of the hyoid/throat musculature characters and seven of the cranial osteology characters were found to differ between the bisexual species under consideration and were used to analyze the relationships of the Kelleys Is. *Ambystoma*. These characters were: whether the rectus cervicis superficialis inserts on the first ceratobranchial, the first basibranchial, or both; the extent to which the genioglossus overlaps the anterior end of the geniohyoideus; whether or not the posterior extension of the first basibranchial deflects dorsally at a right angle; whether the otoglossal is pinched in and extends far anterior to the first basibranchial (Fig. 1); whether the premaxilla has or lacks a ventral lamina; whether the premaxilla is long and thin; whether the ventral edge of the pterygoid is flat (Fig. 2); whether the pterygoid has a laterally curved flange (Fig. 2); whether there is a knob on the dorsolateral surface of the squamosal; whether the length of the squamosal is greater than three times its width; and whether most of the prefrontal lies lateral to the braincase. The first two characters are illustrated and discussed by Krogh & Tanner (1972), and the fifth and sixth by Tihen (1958). The remainder are self-explanatory or illustrated herein (Figs. 1, 2).

Cell squashes were prepared for nine individuals using the method of Kezer & Sessions (1979) and stained with Giemsa. These were then analyzed for mitotic chromosomes. Average erythrocyte size per individual was determined by suspending the cells in amphibian Ringer's solution, measuring the long and short axes of 20 cells with an ocular micrometer, calculating their size by using the equation for the area of an ellipse, and averaging the results. Sex of all specimens was determined by dissection. Notes on the color of living animals were taken for most specimens of the Kelleys Is. *Ambystoma*.

Principal components analyses (PCAs) were applied to the first set of morphological characters in an attempt to determine if the Kelleys Is. *Ambystoma* could be distinguished from the other species of Ohio *Ambystoma* in a multivariate plot format. Separate PCAs were applied which included the hyoid and skeletal characters in addition to the characters used in the first PCA, in an effort to determine whether plots with better separation of clouds could be obtained. I employed the shearing method of Humphries, et al. (1981) for morphometric data. Meristic data were analyzed separately by application of a PCA to the correlation matrix. Sheared PC 2 was then plotted against PC 1 of the meristic data (Humphries, et al., 1981).

Univariate analyses of characters employed pairwise Mann-Whitney U tests and maximum likelihood tests, taking into account Bonferroni's inequality (Hays, 1981).

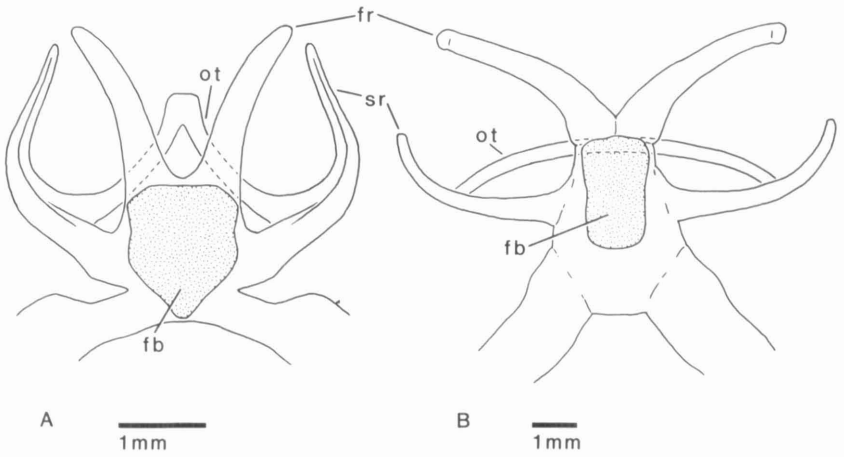


FIG. 1. Hyoids of A) *A. texanum* (UMMZ 174025), and B) *A. t. tigrinum* (UMMZ 174011). fb = first basibranchial, fr = first radial, ot = otoglossal, sr = second radial.

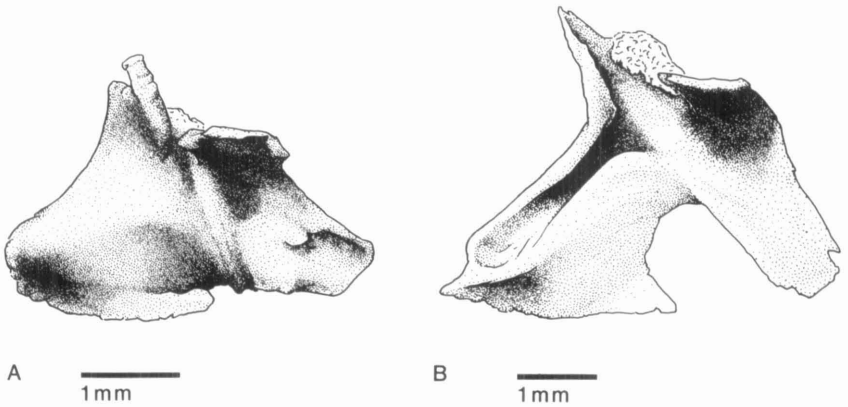


FIG. 2. Pterygoids of A) *A. texanum* (UMMZ 174018), and B) *A. t. tigrinum* (UMMZ 174012).

## RESULTS

In the PCA of all taxa, incorporating only the external morphological characters, the Kelleys Is. *Ambystoma* plotted intermediate to *A. laterale* and *A. tigrinum* (Fig. 3, Table 1). The distribution of *A. jeffersonianum* in the plot was entirely contained within that of the new form. *A. laterale-texanum* fell in between the clouds of *laterale* and *texanum* and did not overlap the cloud of the new form. Principal component 1 accounted for 60 percent of the variation while sheared PC 2 accounted for an additional 10 percent.

In the PCA plot incorporating the hyoid and skeletal characters in addition to the external morphological characters (Fig. 4, Table 2), the Kelleys Is. *Ambystoma* was distinct from all other species and plotted intermediate between *texanum* and *tigrinum*. In this plot PC 1 accounted for 69 percent of the variation while sheared PC 2 accounted for an additional 10 percent. When the sample size of *A. laterale-texanum* was increased by including individuals from other than Kelleys Is., they still formed a cloud of points distinct from all of the remaining taxa, i.e., the plot in Fig. 4 remained unchanged.

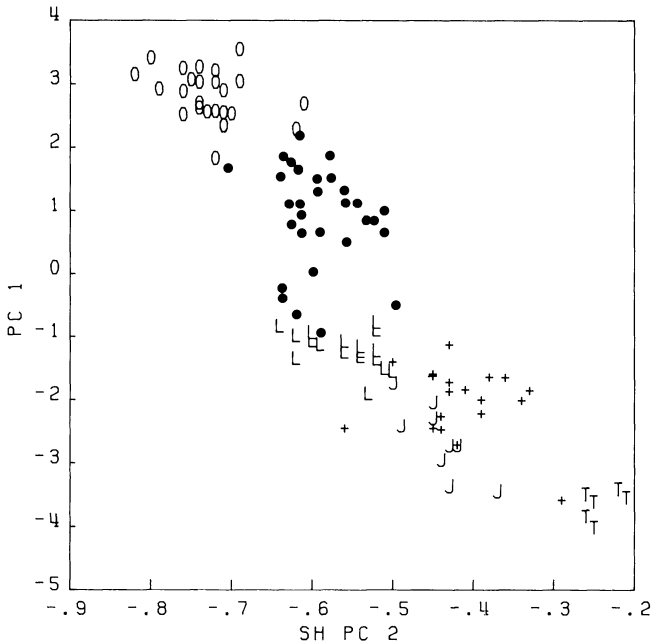


FIG. 3. PC 1 of meristic characters vs. sheared PC 2 of morphometric characters. Only external morphological characters are included. J = *A. jeffersonianum*, L = *A. laterale*, T = *A. tigrinum*, O = *A. texanum*, + = Kelleys Is. *Ambystoma*, ● = *A. laterale-texanum*.

TABLE 1

LOADINGS FOR PRINCIPAL COMPONENTS ANALYSIS OF THE SIX *Ambystoma* TAXA,  
INCORPORATING ONLY EXTERNAL MORPHOLOGICAL CHARACTERS.

Character	PC 1	Sheared PC 2
1) Standard length		-.19
2) Tail length		-.49
3) Internarial distance		.24
4) Snout length		.40
5) Canthal length		.71
6) No. costal folds between adpressed limbs	.37	
7) No. costal grooves	.39	
8) Tongue	.38	
9) No. maxillary-premaxillary tooth rows	.36	
10) No. maxillary- premaxillary teeth	-.24	
11) No. prevomerine tooth patches	-.40	
12) % prevomerine teeth in lateral patches	-.33	
13) No. prevomerine teeth	-.33	

When the Kelleys Is. *Ambystoma* were compared to two taxa at a time, by means of a PCA of only the external characters, they plotted intermediate to the following pairs of taxa: *laterale* and *tigrinum*, *texanum* and *tigrinum*, and *laterale-texanum* and *tigrinum* (Fig. 5, Table 3). When the same analyses also included the osteological and hyoid characters in the PCA, the Kelleys Is. *Ambystoma* plotted intermediate to the following pairs of taxa: *texanum* and *tigrinum*, and *laterale-texanum* and *tigrinum*. In the remaining comparisons to all other pairs of taxa (e.g., with *jeffersonianum* and *texanum*, etc.), the Kelleys Is. *Ambystoma* were not intermediate to the taxa compared. The plots in which the Kelleys Is. *Ambystoma* were intermediate were all similar to that for the combination of the Kelleys Is. *Ambystoma* with *A. tigrinum* and *A. laterale-texanum* (Fig. 5).

Univariate comparisons of meristic characters between the Kelleys Is. *Ambystoma* and *laterale*, *jeffersonianum*, *tigrinum*, and *laterale-texanum* indicated the Kelleys Is. form to be significantly different from each of these species in one to seven characters (Table 4). The new form differed significantly from *texanum* in all eight characters at  $p < 0.001$ .

Morphometric characters could not be tested in the same manner as the meristics as these characters are correlated with standard length in most but not all of these species. However, tail length, internarial distance, snout length, and canthal length may all be plotted against standard length (Figs. 6-9) to attempt to distinguish the Kelleys Is. *Ambystoma* from the other species. Relative tail length serves to distinguish none of the taxa but the remaining three plots separate the Kelleys Is. *Ambystoma* and *A. jeffersonianum* from the other species with a fairly high degree of accuracy. However, these two taxa overlap each other to a great extent.

The Kelleys Is. *Ambystoma* has a black or dark gray ground color largely covered by a pattern of blotches intermediate between the round, well-demarcated blotches of *tigrinum* and the lichenate flecks of *texanum* (Fig. 10). The coloration of these blotches is light bluish gray on the sides, becoming darker gray and olive on the dorsum. However, the Kelleys Is. *Ambystoma* does not attain the same degree of darkness on the dorsum as does the sympatric *A. laterale-texanum*. The undersides of the Kelleys Is. *Ambystoma* are gray with numerous

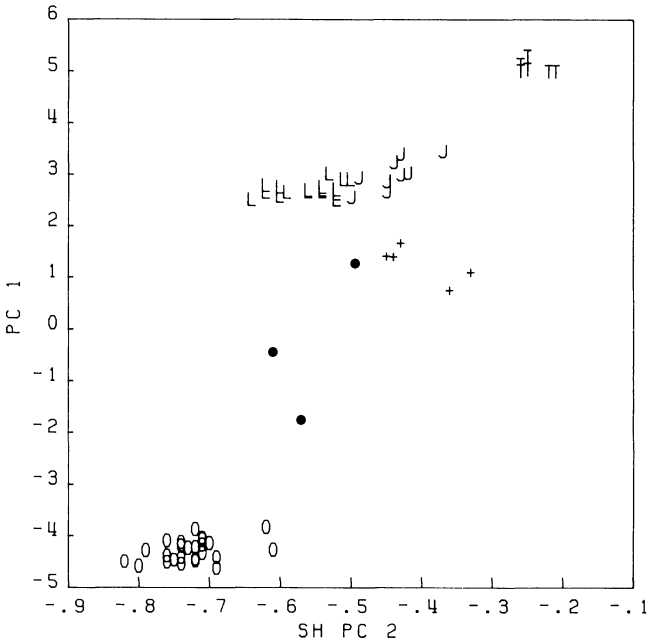


FIG. 4. PC 1 of meristic characters vs. sheared PC 2 of morphometric characters. External, hyoid, and osteological characters are included. Symbols as for Fig. 3.

TABLE 2  
LOADINGS FOR PRINCIPAL COMPONENTS ANALYSIS OF THE SIX *Ambystoma* TAXA,  
INCORPORATING EXTERNAL, HYOID, AND SKELETAL CHARACTERS.

Character	PC 1	Sheared PC 2
1) Standard length		-.19
2) Tail length		-.49
3) Internarial distance		.24
4) Snout length		.40
5) Canthal length		.71
6) No. costal folds between adpressed limbs	-.24	
7) No. costal grooves	-.24	
8) Tongue	-.26	
9) No. maxillary-premaxillary tooth rows	-.24	
10) No. maxillary- premaxillary teeth	.08	
11) No. prevomerine tooth patches	.25	
12) % prevomerine teeth in lateral patches	.22	
13) No. prevomerine teeth	.15	
14) Insertion of rectus cervicis superficialis	.23	
15) Overlap of genioglossus on geniohyoideus	.25	
16) Deflection of 1st basibranchial	.12	
17) Extension of otoglossal	-.27	
18) Lamina of premaxilla	-.27	
19) Shape of premaxilla	-.12	
20) Ventral edge of pterygoid	-.26	
21) Flange of pterygoid	.26	
22) Knob on squamosal	-.25	
23) Width of squamosal	.27	
24) Extent of prefrontal lateral to braincase	.25	



flecks of light bluish gray, and often have a yellowish tinge. The throat and chin are almost always completely suffused with light gray flecks. This serves again to distinguish them from *laterale-texanum*, which typically has a dark chin and throat speckled with a few flecks of light gray. *A. texanum* from northwestern Ohio have a somewhat similar color pattern but are darker and have more lichenate blotches.

All of the Kelleys Is. *Ambystoma* are female. This apparent unisexuality is similar to that reported by Downs (1978) and Kraus (in press) for *A. laterale-texanum* and by Uzzell (1964) for *A. tremblayi* and *A. platineum*. This contrasts with the known ca. 50:50 sex ratios of *laterale* and *jeffersonianum* (Uzzell, 1964) and *texanum* (Kraus, in press). The available *tigrinum* from Kelleys Is. consist of four males and two females.

All Kelleys Is. *Ambystoma* chromosomally analyzed (nine) were triploid. Chromosomal data other than level of ploidy were not noted.

Erythrocyte sizes are available for some individuals lacking chromosome data, and range from 995 to 1650 $\mu^2$  (Fig. 11). Of ten specimens,

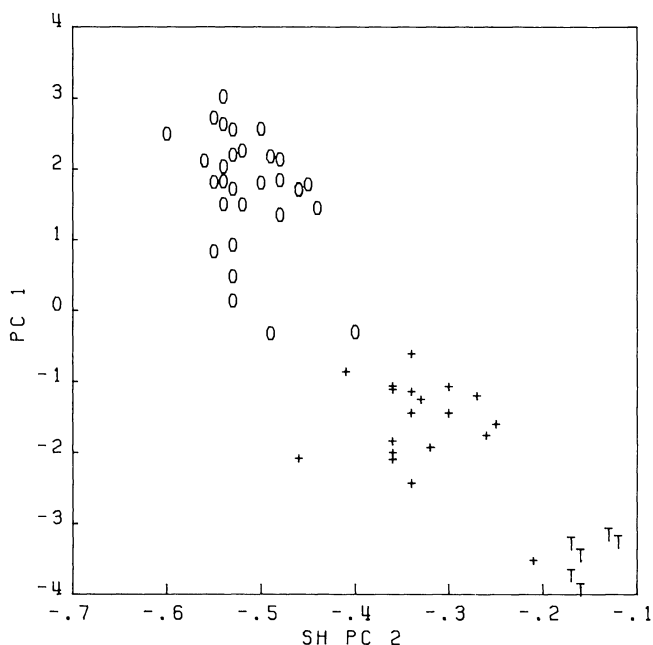


FIG. 5. PC 1 of meristic characters vs. sheared PC 2 of morphometric characters. Only external morphological characters are included. 0 = *A. laterale-texanum*, + = Kelleys Is. *Ambystoma*, T = *A. tigrinum*.

TABLE 3

LOADINGS FOR PRINCIPAL COMPONENTS ANALYSIS OF THE KELLEYS ISLAND  
*Ambystoma*, *A. tigrinum*, AND *A. laterale-texanum*, INCORPORATING  
 ONLY EXTERNAL MORPHOLOGICAL CHARACTERS

Character	PC 1	Sheared PC 2
1) Standard length		-.26
2) Tail length		-.43
3) Internarial distance		.27
4) Snout length		.40
5) Canthal length		.72
6) No. costal folds between adpressed limbs	.36	
7) No. costal grooves	.36	
8) Tongue	.29	
9) No. maxillary-premaxillary tooth rows	.21	
10) No. maxillary- premaxillary teeth	-.40	
11) No. prevomerine tooth patches	-.41	
12) % prevomerine teeth in lateral patches	-.30	
13) No. prevomerine teeth	-.44	

six are known triploids, based on chromosome examination. The range of erythrocyte sizes of the Kelleys Is. *Ambystoma* contrasts strongly with those of known diploid species: *A. laterale*, 600 to 820 $\mu^2$  (Uzzell, 1964); *A. jeffersonianum*, 600 to 860 $\mu^2$  (Uzzell, 1964); *A. texanum*, 600 to 750 $\mu^2$  (Kraus, in press); and *A. tigrinum*, 650 to 800 $\mu^2$  (this study, based on the Kelleys Is. material). Thirteen of the 19 specimens of the Kelleys Is. *Ambystoma* appear, therefore, to be triploid on the basis of chromosomes or erythrocyte sizes (the individual with an erythrocyte size of 1650 $\mu^2$  may be a tetraploid hybrid between the Kelleys Is. *Ambystoma* and one of the bisexual species which occurs on the island).

## DISCUSSION

I interpret the fact that the Kelleys Is. *Ambystoma* plots distinctly from all other *Ambystoma* in the multivariate analyses (Fig. 4) as evidence that it is not assignable to any other presently described taxon.

TABLE 4

CHARACTER DISTRIBUTION COMPARISONS OF THE KELLEYS IS. *Ambystoma*  
TO RELATED SPECIES. KELLEYS IS. *Ambystoma* vs.:

Character	<i>laterale</i>	<i>jeffersonianum</i>
1) No. costal folds between adpressed limbs	same	same
2) No. costal grooves	same	same
3) Tongue	same	same
4) No. maxillary-premaxillary tooth rows	same	same
5) No. maxillary-premaxillary teeth	diff ( $p < .001$ )	diff ( $p < .006$ )
6) No. prevomerine tooth patches	diff ( $p < .001$ )	same
7) % prevomerine teeth in lateral patches	diff ( $p < .001$ )	same
8) No. prevomerine teeth	diff ( $p < .001$ )	same
	<i>tigrinum</i>	<i>laterale-texanum</i>
1) No. costal folds between adpressed limbs	diff ( $p < .001$ )	diff ( $p < .001$ )
2) No. costal grooves	diff ( $p < .002$ )	diff ( $p < .003$ )
3) Tongue	same	diff ( $p < .002$ )
4) No. maxillary-premaxillary tooth rows	same	same
5) No. maxillary-premaxillary teeth	same	diff ( $p < .001$ )
6) No. prevomerine tooth patches	diff ( $p < .001$ )	diff ( $p < .001$ )
7) % prevomerine teeth in lateral patches	same	diff ( $p < .001$ )
8) No. prevomerine teeth	diff ( $p < .010$ )	diff ( $p < .001$ )

Its cohesiveness in the PCA plots (Figs. 3, 4, 5) suggests that it is a single entity and not a composite of different taxa. However, the coloration and morphology of the Kelleys Is. *Ambystoma* suggest influences of different species. This impression is strengthened by the intermediate morphology when a wide array of characters are simultaneously analyzed (Fig. 4, 5). These observations are best explained by postulating a hybrid origin of the Kelleys Is. *Ambystoma*. This hypothesis is consistent with the presence of unisexuality and triploidy, characters highly correlated with a hybrid origin in vertebrates (Cuellar & Kluge, 1972; Schultz, 1977; Wright, 1978). The

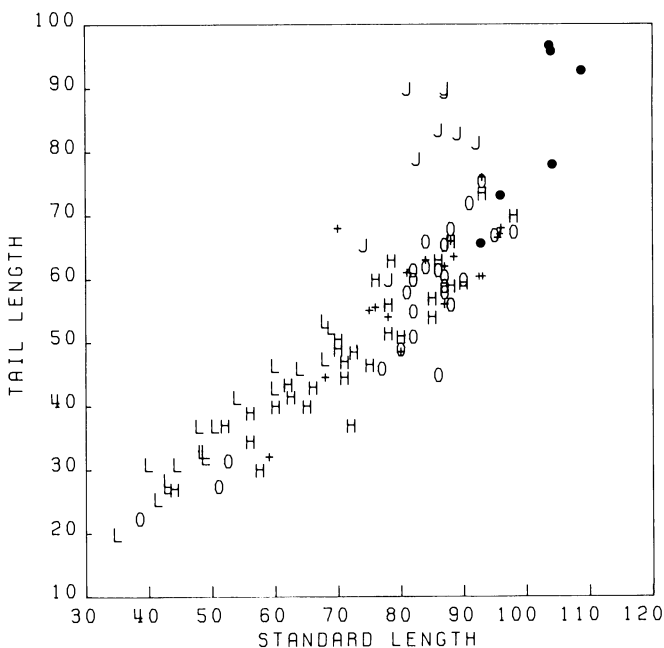


FIG. 6. Tail length against standard length for the six *Ambystoma* taxa under consideration. H = *A. laterale-texanum*, J = *A. jeffersonianum*, L = *A. laterale*, O = *A. texanum*, + = Kelley's Is. *Ambystoma*, ● = *A. t. tigrinum*.

hypothesis of hybrid origin predicts that the parental species involved in the formation of the Kelley's Is. *Ambystoma* would be those species between which the Kelley's Is. *Ambystoma* plots intermediately in a multivariate analysis. The Kelley's Is. *Ambystoma* plots intermediate in the principal components analyses to the following pairs of taxa: *tigrinum*, *texanum*; *tigrinum*, *laterale*; and *tigrinum*, *laterale-texanum*. I interpret this and the new form's triploidy to indicate that the Kelley's Is. *Ambystoma* arose by hybridization involving three bisexual species, *A. tigrinum*, *A. texanum*, and *A. laterale*. That *tigrinum* and *texanum* were involved is also supported by the color pattern of the Kelley's Is. *Ambystoma*, which shows influences of both of these species. That *texanum* was involved is best shown by the tendency of the Kelley's Is. *Ambystoma* to have otoglossals that are pinched in and extend anterior to the first basibranchial (see Fig. 1a). This characteristic is an autapomorphy of the subgenus *Linguaelapsis* (pers. obs.), of which *texanum* is the only member in Ohio. The remaining species of North American *Ambystoma* have otoglossals which form a slightly curved rod extending between the second radials (Fig. 1b).

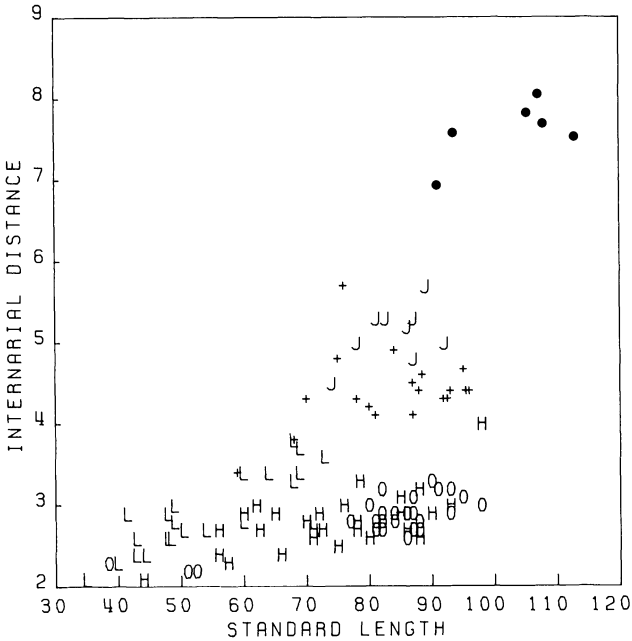
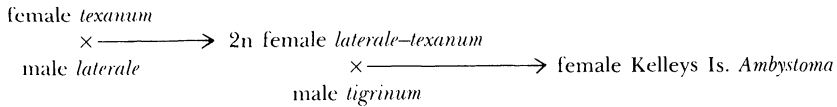


FIG. 7. Internarial distance against standard length for the six *Ambystoma* taxa under consideration. Symbols as in Fig. 6.

Considering the abundance of *A. laterale-texanum* on Kelleys Is., and incorporating the history of *laterale-texanum* as postulated by Kraus (in press), I believe the most reasonable hypothesis for the history of the Kelleys Is. *Ambystoma* is as follows:



This hybridization sequence is consistent with the apparent lack on Kelleys Is. of any diploid unisexuals with a genome from *tigrinum*. However, diploid unisexual salamanders with genomes of *laterale* and *texanum* are common on the island. Inasmuch as the Kelleys Is. *Ambystoma* must be younger than *laterale-texanum* according to the above hypothesis, they too are presumably no older than ca. 130 years. Should the hypothesis that *A. laterale-texanum* is only ca. 130 years old (Kraus, in press) prove incorrect, the apparent restriction of the Kelleys Is. *Ambystoma* to this single island still suggests its origin after the isolation of the island from the mainland ca. 3500 years ago (Forsyth, 1973).

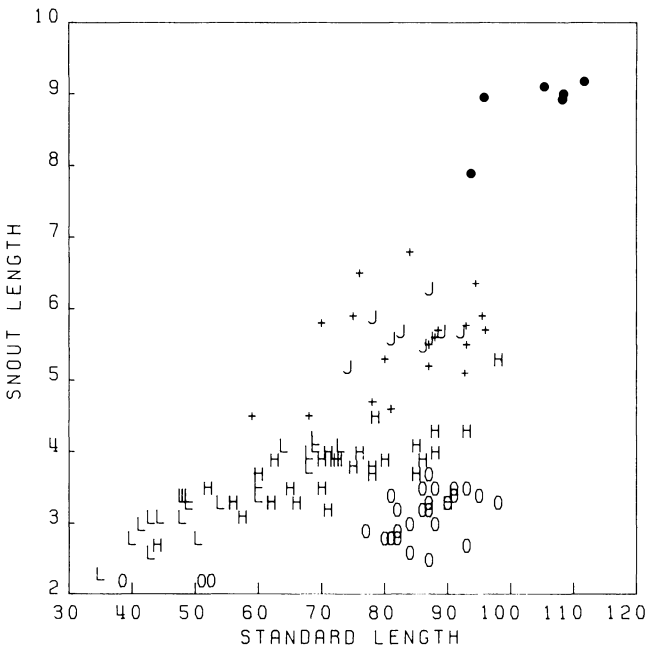


FIG. 8. Snout length against standard length for the six *Ambystoma* taxa under consideration. Symbols as in Fig. 6.

The reproductive mechanism of the Kelleys Is. *Ambystoma* has not yet been studied, but presumably must consist of either parthenogenesis, gynogenesis, or hybridogenesis. J. Kezer (pers. comm.) has observed that in diplotene, the chromosomes in the ova of the Kelleys Is. *Ambystoma* are doubled, suggesting pre-meiotic endoduplication. This tends to negate the possibility of a hybridogenetic mechanism operating in this form.

Because the Kelleys Is. *Ambystoma* is readily distinguishable from all other members of the genus, is morphologically cohesive, is of uniform ploidy, and apparently occupies a very restricted range, I think it likely that it represents an evolutionarily independent lineage and is, therefore, deserving of taxonomic status. In recognition of its apparent allotriploid nature, I propose it be named:

*Ambystoma nothagenes* sp. nov.  
Kelleys Island Salamander  
Fig. 10

HOLOTYPE: UMMZ 176237 (FK 819), an adult female from Kelleys Is., Erie Co., Ohio, collected on 1 October 1982 by Fred Kraus.

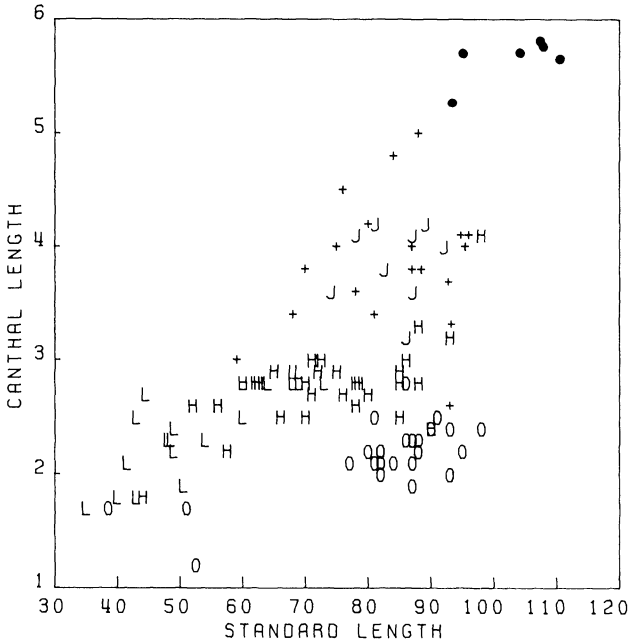


FIG. 9. Canthal length against standard length for the six *Ambystoma* taxa under consideration. Symbols as in Fig. 6.

PARATYPES: UMMZ 172945, 172953–54 (RAN 23066, 23074–75, respectively) and UMMZ 176226, 176228–36, 176238–41 (FK-372, 713–14, 737, 744, 754–56, 817, 818, 820, 827, 831, 832, respectively). All are from the same locality as the holotype.

DIAGNOSIS.—A moderately large unisexual, triploid *Ambystoma* with 1) a long, broad snout, 2) long limbs, 3) one row of maxillary-premaxillary teeth, 4) tongue with plicae radiating from the back, 5) large number of prevomerine teeth, range 46 to 69, mean 57, 6) large number of maxillary-premaxillary teeth, range 106 to 155, mean 131, 7) prevomerine teeth include two lateral patches of “palatine” teeth and have a tendency to lack the diastema which separates the “palatine” from the prevomerine teeth, 8) 13-14 costal grooves, 9) rectus cervicis superficialis inserts only on the first ceratobranchial, 10) genioglossus entirely overlaps the anterior end of the geniohyoideus, 11) posterior extension of the first basibranchial does not deflect at a right angle dorsally, 12) otoglossal pinched in and extends far anteri-

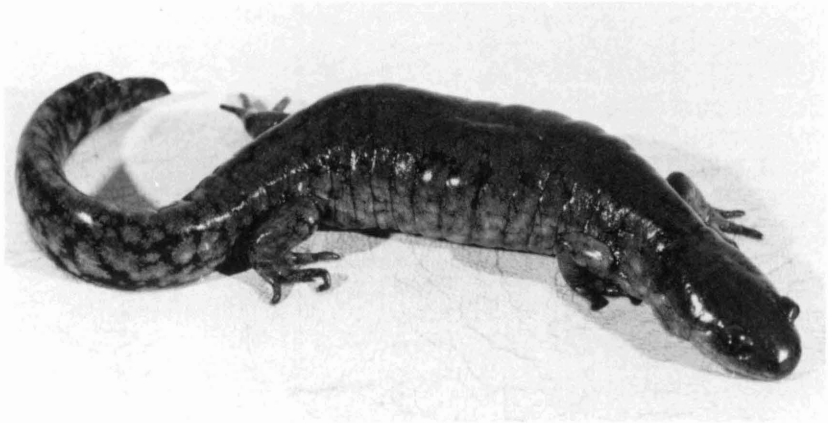


FIG. 10. Adult paratype of *Ambystoma nothagenes* (UMMZ 176234) in life.

or to first basibranchial, 13) premaxilla without a ventral lamina, 14) knob on dorsolateral surface of the squamosal, and 15) color pattern as described above (Fig. 10).

In reference to the numbered characters above, *A. nothagenes* may be distinguished from *A. jeffersonianum* by characters (10), which in *jeffersonianum* only partially overlaps the anterior end of the geniohyoideus; (12); (14); and (15). It differs from *A. laterale* in characters (5), which in *laterale* ranges from 24-41 (mean 32); (6), which in *laterale* ranges from 62-124 (mean 94); (10), which in *laterale* is as described above for *jeffersonianum*; (12); (14); and (15). It differs from *A. tigrinum* in characters (10), which in *tigrinum* does not overlap the anterior end of the geniohyoideus; (11); (12); (14); and (15). It differs from *A. texanum* in characters (1); (2); (3); (4); (5), which in *texanum* ranges from 21-48 (mean 32); (6), which in *texanum* ranges from 72-135 (mean 105); (7); (8), which in *texanum* ranges from 14-16 (median 15); (9), which in *texanum* inserts only on the first basibranchial; (12), which in *texanum* extends even farther anteriorly; (13); and details of (15) noted previously. It differs from *A. laterale-texanum* in characters (1); (2); (4); (5), which in *laterale-texanum* ranges from 10-48 (mean 29); (6), which in *laterale-texanum* ranges from 78-149 (mean 97); (7); (9), which in *laterale-texanum* inserts on both the first ceratobranchial and first basibranchial; and details of (15) noted previously. It differs from *A. tremblayi* and *A. platineum* in the same characters noted above for *A. jeffersonianum*.

*Ambystoma nothagenes* is readily distinguishable from all other species



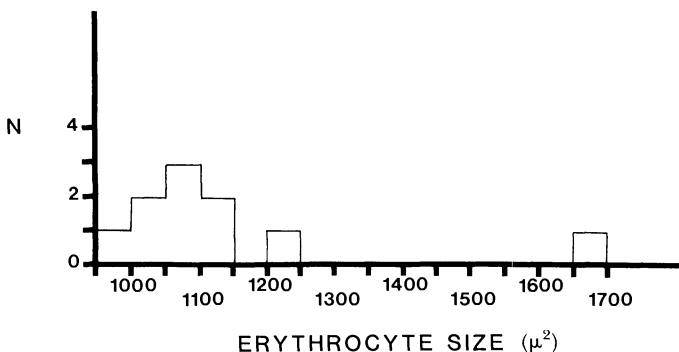


FIG. 11. Distribution of erythrocyte sizes for the Kelleys Is. *Ambystoma*, in  $50\mu^2$  intervals.

of *Ambystoma* on the basis of color pattern and differs from all other *Ambystoma* except *platineum*, *tremblayi*, and *laterale-texanum* in being unisexual and triploid.

**DESCRIPTION OF HOLOTYPE.**—Adult female with midventral incision; heart, spleen, part of liver, and muscle from left hind leg removed. Snout-vent length 85 mm, standard length 93 mm, tail length 61 mm, snout length 5.7 mm, canthal length 3.7 mm, internarial distance 4.3 mm. Adpressed limbs separated by three costal folds, 14 costal grooves on each side. Lower jaw protrudes farther anteriorly than the upper. Plicae of tongue radiate forward from the back, no median furrow present. Maxillary-premaxillary teeth 129, in one row. Preopercular teeth 61, in one continuous row from naris to naris. Coloration in life with black ground color almost entirely suffused dorsally with dark greenish gray rounded lichenate markings and laterally with light blue blotches largely fused together. Venter gray with light bluish flecks. Coloration in preservative dark gray dorsally with a suffusion of light gray blotches on sides. Ground color of venter gray, but not so dark as dorsally, with numerous light gray flecks and blotches. Chin almost entirely suffused with light gray flecks.

**VARIATION.**—The variation in some of the external morphological characters is summarized in Table 5 and detailed in Table 6. The tongues in *A. nothagenes* have a range in variation with lingual plicae that diverge from the posterior of the tongue to some that diverge from a slightly developed median furrow. When the limbs are adpressed to the side of the animal, they vary from being separated by  $3\frac{1}{2}$  costal folds to overlapping by an equal amount. The median condition is to have the limbs separated by  $1\frac{1}{2}$  costal folds; the mode,

TABLE 5  
CHARACTER VARIATION IN *Ambystoma nothagenes*.

Character	range	mean	std. dev.
1) Tail length/SL	5.4–9.7*	7.0*	8.6 <sup>†</sup>
2) Internarial distance/SL	4.6–7.5 <sup>†</sup>	5.4 <sup>†</sup>	7.6 <sup>‡</sup>
3) Snout length/SL	5.5–8.6 <sup>†</sup>	6.7 <sup>†</sup>	9.4 <sup>‡</sup>
4) Canthal length/SL	3.5–5.9 <sup>†</sup>	4.8 <sup>†</sup>	6.8 <sup>‡</sup>
5) No. costal grooves	13–14	14 <sup>§</sup>	—
6) No. maxillary-premaxillary tooth rows	1	—	—
7) No. maxillary-premaxillary teeth	106–155	131	12.67
8) % prevomerine teeth in lateral patches	0–37	16	13.68
9) No. prevomerine teeth	46–69	57	6.39

\*  $\times 10^{-1}$

<sup>†</sup>  $\times 10^{-2}$

<sup>‡</sup>  $\times 10^{-3}$

<sup>§</sup> median and mode

separated by 2 costal folds. The prevomerine teeth vary from being in three well-defined patches (one median and two lateral) to forming a continuous row from under one naris to under the other. The mode is discreet patches, but the diastemas between them are usually small.

One of the five specimens examined has the genioglossus only partially overlapping the geniohyoideus. In one specimen the anterior section of the intermandibularis posterior is not divided by the linea alba. In two specimens the interhyoideus posterior inserts above the second basibranchial instead of posterior to it. In two specimens the posterior extension of the first basibranchial is ossified instead of cartilaginous. The ventral edge of the pterygoid ranges from flat as in *texanum* to highly concave as in *tigrinum* (see Fig. 2). The anterodorsal edge of the pterygoid may have a laterally curved flange or the flange may be fused with the remainder of the pterygoid (see Fig. 2). The knob on the dorsolateral surface of the squamosal is lacking in one specimen.

HABITAT.—I have found adults of *A. nothagenes* most frequently in woodlands in the limestone talus which forms at the bases of ledges on Kelleys Is. I have also found them around the margin of a small seasonal woodland pond about 1 km away from the nearest woodland ledges. Newly metamorphosed individuals of *Ambystoma* have only been found at the latter locality, which is the only suitable breeding

TABLE 6  
SUMMARY OF CHARACTERS FOR 18 *Ambystoma nothogenes*.

Number <sup>1</sup>	SL <sup>2,3</sup>	Tail <sup>3</sup>	Inter-narial distance <sup>3</sup>	Snout <sup>3</sup>	Canthal <sup>3</sup>	No. costal folds <sup>4</sup>	No. costal grooves <sup>5</sup>	No. maxillary-premaxillary teeth	No. prevomerine teeth	Erythrocyte size <sup>6</sup>	Chromosomes
172945	80.0	48.5	4.2	5.3	4.2	+1	14, 14	127	63	1125	3n
172953	88.0	66.0	4.4	5.6	5.0	+½	14, 14	143	57	1089	3n
172954	81.0	61.0	4.1	4.6	3.4	+½	14, 14	120	46	1077	—
176226	59.0	32.0	3.4	4.5	3.0	-1½	14, 14	106	47	1124	—
176228	88.5	63.5	4.6	5.7	3.8	+½	14, 14	128	55	—	—
176229	68.0	44.5	3.8	4.5	3.4	-½	14, 13	120	54	—	—
176230	96.0	68.0	4.4	5.7	4.1	+2	13, 14	128	59	—	—
176231	93.0	60.5	4.3	5.1	3.3	+2	14, 14	125	48	1248	3n
176232	87.0	56.0	4.1	5.2	3.8	+1½	14, 14	118	57	995	3n
176233	75.0	54.0	4.8	5.9	4.0	+2	14, 14	148	64	1060	3n
176234	87.0	62.0	4.5	5.5	4.0	+2	14, 14	129	57	1000	3n
176235	95.0	68.5	4.7	6.4	4.1	+2½	13, 14	144	57	—	3n
176236	78.0	54.0	4.3	4.7	3.6	+1	13, 13	125	59	—	3n
176237	93.0	61.0	4.3	5.7	3.7	+3	14, 14	129	61	—	3n
176238	84.0	63.0	4.9	6.8	4.8	+3½	14, 14	153	65	—	—
176239	95.5	66.5	4.4	5.9	4.0	+1½	13, 14	137	61	1002	—
176240	70.0	68.0	4.3	5.8	3.8	-2	13, 13	129	49	—	—
176241	76.0	55.5	5.7	6.5	4.5	-3½	14, 13	155	69	—	—

<sup>1</sup> All are UMMZ

<sup>2</sup> Standard length

<sup>3</sup> In millimeters

<sup>4</sup> +, separated by; -, overlap by

<sup>5</sup> Right side, left side

<sup>6</sup> In square microns.

habitat on the island that I have located. The woods at collection sites are young, the trees averaging only about 10 to 15 meters high. The understory is dominated by members of the Geraniaceae and Cruciferae.

*A. nothagenes* is sympatric and syntopic with *A. tigrinum*, *A. texanum*, and *A. laterale-texanum*, the last of which is the predominant salamander on the island.

ETYMOLOGY.—The species name is from the Greek *nothagenes*, meaning “bastard-born.” I propose that “Kelleys Island Salamander” be used as the vernacular name of this species.

NOTES.—The individual (UMMZ 176227) with an erythrocyte size of  $1650\mu^2$  may be a tetraploid hybrid between *A. nothagenes* and *A. tigrinum* or *A. texanum*.

Because of its questionable ploidy I do not consider it an example of the new species and it has been excluded from the description and analysis of variation of *A. nothagenes* presented above.

TAXONOMIC INTERPRETATION.—Alternative systems of nomenclature exist for unisexual vertebrate taxa. The first is to identify the taxon with a formal binomial, as is regularly done for bisexual species. This system has typically been applied to parthenogenetic and certain gynogenetic forms. The alternative, used for hybridogenetic and other gynogenetic taxa, is to refer to the unisexual taxon with an informal name which incorporates the names of the parental species separated by a hyphen. One of the parental names is sometimes prefixed with a “2”, if necessary, to indicate if that species has contributed two genomes, instead of one, to the formation of the unisexual taxon. The second method typically has been used when there is evidence that the taxon in question is not genetically well-isolated from its parental species (see Schultz, 1977 for review of *Poeciliopsis*) or consists of distinct clones or ploidy classes (Kraus, in press). Recent evidence indicates that *A. tremblayi* and *A. platineum* have unexpectedly high levels of genomic variation (Bogart, 1982; Sessions, 1982) and probably should not be recognized with formal binomials. Concern that a similar condition might occur in *A. nothagenes* is reasonable and is addressed here.

There are three situations under which designation of a unisexual taxon with a formal binomial name should be avoided. If the unisexual taxon is: (1) a composite of genomically distinct individuals, (2) not genetically well-isolated from its parental species (such as occurs in hybridogenesis or “leaky” gynogenesis), or (3) infertile and unable to reproduce itself (i.e., if it forms an isolation or equilibrium taxon, *sensu* Kraus, in press). All of these situations can be viewed as cases in

which the requirement that the taxon in question be an evolutionarily independent lineage is violated. Because studies on *A. nothagenes* are just beginning, conclusive evidence regarding any of these three points is presently lacking. However, several preliminary lines of evidence suggest that none of these three situations obtains, and each point shall be considered in the correspondingly numbered section below. 1) All individuals of this taxon are triploid and morphologically uniform, suggesting that each individual possesses one and only one genome from each of *A. laterale*, *A. texanum*, and *A. tigrinum*. The taxon is not a composite of diploid and triploid individuals as shown by Downs (1978) and Kraus (in press) for *A. laterale-texanum*, nor does it appear to contain individuals with varying genomic contributions from the three parental species as apparently occurs in *A. tremblayi* and *A. platineum* (Sessions, 1982; Bogart, 1982, pers. comm.). 2) *A. nothagenes* is triploid and apparently undergoes a premeiotic endoduplication (J. Kezer, pers. comm.). Both traits are absent from known hybridogens and are cytologically incompatible with the known mechanism of hybridogenesis. Thus, hybridogenesis appears an unlikely reproductive mechanism in *A. nothagenes*. The only proposed mechanism of leaky gynogenesis (Sessions, 1982) requires that two of the genomes in a triploid gynogen be from the same species so that they may synapse with each other to the exclusion of the third genome during meiosis. The excluded genome is then consigned to the polar body during first meiosis, resulting in a diploid ovum with both genomes derived from the same parental species. One might possibly extend this argument to hypothesize that the two genomes which synapse need not be from the same species, but only from closely related species. However, in *A. nothagenes*, not only aren't two of the genomes from the same species, as required by Sessions' model, but none of the three parental species is closely related to either of the other two (Tihen, 1958; Kraus, pers. obs.). This makes it highly unlikely that even the questionable extension of Sessions' model could be operative in *A. nothagenes* so as to form a leaky gynogenetic system. Additionally, if a leaky gynogenetic system operated in *A. nothagenes*, equal numbers of resultant diploid *A. laterale-texanum*, *A. laterale-tigrinum*, and *A. texanum-tigrinum* would be expected from the meiotic reduction. However, no specimens of the last two forms have been found on Kelleys Is. (the first is common and was formed by the hybridization of the two parental bisexual species (Kraus, in press)), again suggesting that a leaky gynogenetic system does not operate in *A. nothagenes*. 3) All mature individuals of *A. nothagenes* have well-yolked eggs in their oviducts, suggesting that they are fertile and

self-reproducing; however, the fate of these eggs in the wild is unknown. *A. nothagenes* also comprises a sufficiently large percentage of the total number of *Ambystoma* on Kelleys Is., (ca. 30) to make its dependence on continual reformation for survival questionable.

There is yet an additional concern expressed by some that if formal binomials are used to recognize unisexual taxa, each clone that we are capable of recognizing must be given its own formal name. I disagree with this conclusion. Different clones in unisexual taxa may arise by processes of mutation, recombination, or multiple origins (see Parker and Selander, 1976 for discussion). However, clones which arise by mutational or recombinational processes are not independent of their ancestral clone, but are merely derived from the latter by relatively minor changes transmitted from mother to daughter. Instead of forming a series of evolutionarily independent lineages, these clones form a sequence of ancestral-descendant individuals separated from each other by minor phenotypic differences. The derived clones are no more independent of the ancestral clone than are offspring from parents in bisexual species. Because binomials are used to identify evolutionarily independent lineages, clones derived from mutational and recombinational processes are not deserving of formal species recognition separate from the ancestral clone. Only clones derived from multiple origins (independent hybridization events) may be construed as evidence that the unisexual taxon in question may best be treated in a manner other than by formal recognition of the composite of clones with a binomial. There is presently no evidence of multiple origins of *A. nothagenes*.

## SPECIMENS EXAMINED

All specimens listed below are housed in UMMZ.

*Ambystoma* sp. OHIO: Erie Co., Kelleys Is. (176227).

*Ambystoma jeffersonianum*. NEW YORK: Tompkins Co., near Ithaca (170197-201, 173710-12). OHIO: Clermont Co., near Goshen (173708-09); Delaware Co., 1.6 mi. E Norton (150663-71, 170202-15); Marlboro Twp., .5 mi. S Mayfield Corners (122584 (2), 123057-58, 173714); Hocking Co. (151019), near Logan (122785 (3)); Lorain Co., Amherst Twp., 1.5 mi. WNW North Amherst (122839, 122844, 122846-47); Warren Co., Fort Ancient St. Pk. (173713).

*Ambystoma laterale*. ILLINOIS: Cook Co., Palos Park Forest Preserve (173704-06). MAINE: Penobscot Co., .5 mi. W Orono (173703). MASSACHUSETTS: Essex Co., Boxwood (173707). MICHIGAN: Arenac Co., Charity Islands (42147 (2), 42149 (2)); Chippewa Co., Vermilion (45677 (3), 45692-93); Jackson Co., Leonie Twp., Sec. 33 (150804-05); Livingston Co., E. S. George Reserve (151034, 173978-80); Luce Co. (68387), 8.5 mi. W

Newberry (77832); Mackinac Co., Daggett Fire Tower Woods (101349); Bois Blanc Island (122866, 123267); Montmorency Co., Sec. 27 T29N R2E (98925 (7)); Washtenaw Co., Whitmore Lake (150679, 150689-92, 151033), Mud Lake (150693), Lodi Twp. (150802), Northfield Twp., Sec. 20 (150678, 150680-88, 150803, 173981-83), Scio Twp., Zeeb Woods (150800-01, 151035).

*Ambystoma laterale-texanum*. OHIO: Erie Co., Kelleys Is. (173975-96, 176386-99, 176401-02, 176404-07, 176530-33).

*Ambystoma nothagenes*. OHIO: Erie Co., Kelleys Is. (172945, 172953-54, 176226, 176228-41).

*Ambystoma texanum*. ARKANSAS: Monroe Co., 3 mi. W Brinkley (174015-17). IOWA: Lucas Co., 1 mi. W Lucas (174018-21). MICHIGAN: Washtenaw Co., Scio Twp., Zeeb Woods (150809-14, 151015, 151044, 170224, 170228-29). OHIO: Delaware Co., .5 mi. S Mayfield Corners (151043), .5 mi. W Ashley (170231-32, 170261-62); Hancock Co., Eagle Twp., Higbie Woods (122388 (7), 122389 (5), 123575, 124036 (3), 150815-16); Marion Twp. (96004, 123570 (2)), Van Buren Twp., Wilson's Woods (124035 (9)); Union Co., Union Twp., 1.5 mi. S Bridgeport (174022-25).

*Ambystoma t. tigrinum*. MICHIGAN: Livingston Co., Hamburg Twp., Sec. 28 (174009-12); Washtenaw Co., near Ann Arbor (170938); 5 mi. N Whitmore Lake (150823-28); Goss Pond (134965, 134967-68). OHIO: Erie Co., Kelleys Is. (172955-60).

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