Final Report:

Population estimates and age class structure of the salamander *Plethodon albagula* (Plethodontidae) at Fort Hood, Texas

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Executive summary

Populations of geographically disjunct and morphologically unique *Plethodon albagula* salamanders, which may represent an undescribed species, occur at Ft. Hood, Texas. Prior studies focused on their distribution in karst features (caves, sinkholes and springs) and co-occurrence with red imported fire ants, a known threat to the karst ecosystem. This study examines population size and age class structure at two of the 27 known localities at Fort Hood, using timed area searches, morphological measurements, and mark-release-recapture methods.

Sampling at Bear Spring and Estes Cave on nine occasions each between February 2004 and July 2005, yielded a total of 378 salamander encounters in timed area searches. Schnabel and Schumacher-Eschmeyer estimates of population sizes were 331 and 318, respectively, for Bear Springs, and 71 and 80, respectively, for Estes Cave. Four size metrics (snout-vent length, total length, mass and volume) were significantly different in the spring versus cave populations, with salamanders in the cave population being larger on average. Size class distribution at Bear Springs showed distinct peaks, probably associated with breeding season age cohorts, suggesting that hatching may occur in November through January. The salamanders reach sexual maturity after two years and live for three or more years. The population at Estes Cave had a dissimilar pattern, with no obvious hatching time and salamanders were less abundant in the younger cohorts. Minimum size for adult males (with mental glands) was 47.9 mm SVL and for gravid females (individuals with distended abdomens, but without mental glands) was 49.6 mm SVL. Growth was observed in 33 individuals and averaged 17.5 mm per year, with faster growth in smaller individuals.

Search effort and salamander capture rates indicated that during the hot, dry summer months (June through August) the salamanders rarely are encountered, violating the assumptions of equal catchability and closure inherent in the population estimate models. Although *P. albagula* has been reported only from karst features (caves, sinks, and a spring complex) at Fort Hood, we report 54 salamander encounters in surface habitats, up to 644.8 meters from known karst features during this study. This indicates that the species may be less restricted to karst features than previously thought, but inaccessible for sampling during much of the year. Threats to this species include the red imported fire ant (*Solenopsis invicta*) and habitat alteration by humans and livestock. Monitoring salamander populations, controlling fire ants, and limiting accessibility to the sites by vehicular and livestock traffic are recommended.

Introduction

Salamanders of the genus *Plethodon* occur in a variety of mesic woodland habitats and are generally distributed throughout the eastern United States (Petranka 1998). Recently, populations which may represent an undescribed species have been discovered at Fort Hood, Texas (Reddell 2001). These geographical distinct and morphologically unique salamanders, which by taxonomic convention should be referred to as *Plethodon albagula* until some other determination is published, are associated with karst features such as caves and sinkholes, and have been identified from 27 (Charles Pekins, personal communication 14 October 2005) of the roughly 250+ such features at Ft. Hood (Reddell 2001, Taylor and Phillips 2002). All but one of the salamander caves at Fort Hood are in Bell County, the single Coryell County site representing an unconfirmed record. Red imported fire ants (RIFA), *Solenopsis invicta* Buren (Hymenoptera: Formicidae), have been listed as a serious threat to karst communities in Bexar County, Texas (USFWS 2000) and these ants occur across most of Fort Hood (Taylor *et al*. 2003b). RIFA are thought to compete with cavernicoles for food and at least occasionally prey upon animals in the caves (Elliott 1993). Recent work (Taylor *et al*. 2003a) at Fort Hood (Bell and Coryell counties, Texas) has demonstrated that the ants forage far below ground and well into the dark zone of the caves, though they are most frequently found only in the entrance and twilight zones of Ft. Hood caves.

During our previous study of *P. albagula* at Ft. Hood (Taylor and Phillips 2002), we documented especially large populations of this salamander at Estes Cave and at Bear Springs (divided into Bear Springs East and Bear Springs West in our 2002 study). During that study we marked individuals with toe clips, and returned to Estes Cave and Bear Springs in an attempt to estimate population size. Because we did not recover any of the marked individuals, we know the populations are large, but were unable to produce estimates using available mark/recapture models (e.g., Jolly-Seber,

Cover Photo: *Plethodon albagula* at Estes Cave, 26 July 2005. Photo by Jean Krejca.

Schnabel, Schumacher and Eschmeyer – see Phillips *et al*. [2001] for a discussion and analysis of these methods). Finally, snout vent length (SVL) data from Taylor and Phillips (2002) showed promise as a method for obtaining more detailed data on life history and age class structure for this salamander.

In this report, we describe more intensive surveys of *P. albagula* at Estes Cave and Bear Springs over a two year period, designed to derive robust estimates of population size at these sites. In addition, we report new data on life history and age class structure for *P. albagula* at these sites.

Methods

Plethodon albagula were surveyed at Estes Cave and Bear Springs (Fort Hood, Texas) using time and area constrained visual encounter survey (VES; Heyer *et al.* 1994), as in Taylor and Phillips (2002). This is brute force searching with time recorded to the nearest minute and area recorded with a text description and sketched on a map of the feature. This basic quantitative method allows for estimating presence-absence, relative abundance and absolute abundance. These sites were examined on an approximately bi-monthly basis for two years for a total of nine sample periods (Estes Cave: 17 February 2004, 20 April 2004, 29 June 2004, 24 August 2004, 18 November 2004, 26 January 2005, 30 March 2005, 3 May 2005, 26 July 2005; Bear Springs: 13 February 2004, 21 April 2004, 29 June 2004, 24 August 2004, 17 November 2004, 25 January 2005, 29 March 2005, 2 May 2004 and 26 July 2005). Historical weather data (temperature, precipitation) were obtained from the Fort Hood/Killeen airport weather station KHLR, about 19 km WSW from the study sites (approximate, UTM: [NAD83] zone 14 R 622350mE 3445090mN). These data were obtained over the internet (<http://www.wunderground.com/> [accessed September 20, 2005]). A recently installed weather station at Owl Creek (Fort Hood) is much closer to the study area, but went into operation too late to cover the full duration of our study.

Bear Springs is composed of two primary spring heads and the associated spring runs (Figure 1). The west spring branch is approximately three times as long as the east branch. The springs and spring branches were broken up into seven stream segments of approximately equal size, that were marked with wire flags for the duration of the study. During each visit to Bear Springs, individual researchers searched each zone (Figure 2) marking the site for each *P. albagula* they found with a wire flag (Figure 3). At the end of each sample period, the location of each wire flag was plotted using a compass and tape survey, the distance to the nearest open water was measured, and the flag was removed.

Estes Cave is a simple, narrow, vertical pit (Figure 4), which can safely be entered using single rope techniques. Given these limitations, it was not feasible to have more than one researcher in the cave at a time, so searches were conducted by a single individual. The bottom of the cave was searched first, then the ledges part way up along the sides of the pit (Figure 4).

Plethodon albagula encountered were weighed to the nearest 0.1 g using Pesola spring scales (Figure 5). Snout-vent length (SVL) and total length (ToTL) were recorded to the nearest 0.1 mm using digital calipers (Figure 6) – the salamander was confined to a moist ziplock bag for this procedure so that we could ensure that the animal was as straight as possible. The volume of each salamander was determined by displacement, using a graduated cylinder and spring water (Figure 7). Individuals over 25 mm SVL were marked by clipping one toe under the marking system of Medica *et al.* (1971; see also Fig. 5B of Ferner 1979). We chose to clip a different toe for each of the two sites (outside left front for Estes Cave, outside right front for Bear Springs). Each individual was checked for the presence of a mental gland (indicative of a mature male) and mature eggs (indicative of a gravid female).

We also used colored injectable latex microbeads – first used in the study of Salmon in the Pacific Northwest – to give each captured salamander a unique set of marks (Figures 8, 9). Each mark was made using a new, sterile syringe to avoid transferring disease. All animals were chilled on ice in ziplock bags prior to marking. Marks were located on the lateral surface at the insertion of each limb, and the color marking scheme was then read like a book (left front, right front, left rear, right rear) so that a salamander with a green mark associated with the left front leg and with both hind legs, but no mark on the right front leg, would be identified as “GnGG”, whereas a salamander with yellow marks associated with both front legs and red marks associated with both hind legs would be identified as “YYRR”. This system seemed to be working well until the company producing the latex dye went out of business. We switched to another company, but unfortunately the colors of the new dye, a visible implant elastomer, comprised of a two-part silicone based material made by Northwest Marine Technology (Washington, USA), were different from those of the first, causing difficulty in determining the actual colors on recaptured, previously tattooed salamanders. We attempted to alleviate these problems by adding a fifth tattoo mark in the middle of the back to indicate the new dye had been used. This mark was coded as a “+” and thus if the salamanders in the example above were tattooed with the new dye, they would be identified as “GnGG+” and “YYRR+” respectively (Figure 10). Salamanders were released unharmed at the point of capture.

We used two closed models of population estimation: the Schnabel (1938) method and a regression method (Schumacher and Eschmeyer 1943). We chose closed models because we could not read all of the dye marks with certainty and therefore our sample of uniquely marked individuals was small. We addressed the validation of equal catchability and population closure using linear regression as outlined by Krebs (1999). This was accomplished by examining the relationship between the recapture rate at time **i** and the cumulative number of individuals available for recapture at that time. If the regression is significant and the **y**-intercept is not significantly different from zero, the assumptions of equal catchability and closure are met.

Results

During nine site visits to Bear Springs and Estes Cave, we recorded 378 salamander encounters within the timed area search zones. Some individuals were captured more than once, so that number does not represent the numbers of salamanders present. Of these, 261 salamander occurrences were recorded in the timed area searches at Bear Springs, and 117 salamander occurrences were recorded in the timed area searches at Estes Cave. The number of salamander occurrences in timed area searches varied greatly among sample periods and by cave, with numbers at Bears Springs ranging from 1 to 78 salamanders ($\bar{x}$ = 29) and numbers at Estes Cave ranging from 3 to 24 salamanders ($\bar{x}$ = 13, Figure 11). Our impression in the field was that the salamanders were less available for sampling during the hot, dry periods of the year, and this impression is supported in part by weather data (Figure 12), especially in terms of temperature. At Bear Springs, the timed area search was divided into seven search zones (Figure 1), and salamander occurrences varied both seasonally and by search zone. The spatial differences in overall abundance of salamander occurrences are apparent in Figure 1, and differences among sample periods suggest that during hot/dry periods, salamanders are more available for sampling in zones closest to the springheads (zones 5-6 and 9-10; Figures 13, 14). At Bear Springs, total search time ranged from 2 hrs 5 min to 6 hrs 7 min, averaging 4 hrs 25 min. At Estes Cave, the search time for the bottom and ledges (Figure 4), ranged from 21 to 47 min, averaging 33.3 min. The great fluctuation in search times was strongly correlated with the increased handling times associated with capturing the salamanders, thus during periods when salamanders were abundant, minutes of searching spent per salamander encountered was quite low, and when salamanders were not abundant, the minutes of searching spent per salamander was often higher (Figure 15).

Distance from water at Bear Springs did not appear to vary significantly by season or sample zone (Figure 16), although the data were not tested statistically due to small sample sizes for some zones and dates. In timed area searches at Bear Springs, salamanders were found on average at 0.69 (n=285) meters from water, almost always under loose rocks.

At both Bear Springs (Figure 17) and Estes Cave (Figure 18), we spent some time searching for salamanders in potential habitat away from the study site during several of the sample periods. At both sites we found *P. albagula* away from known karst features. At Bear Springs, these searches were conducted on 19 November 2004, and 25 January, 26 January, and 29 March 2005, with 7, 2, 14 and 19 salamanders discovered, respectively. At Estes Cave, these searches were conducted on 18 November 2004, and 25 January, 26 January, and 30 March 2005, with 3, 3, 4 and 2 salamanders discovered, respectively. The habitat searches were nonrandom, focusing especially on what might be suitable habitat (e.g., moist, shaded areas or under logs and rocks). These 54 salamanders were found during periods when salamanders were readily available in the timed area search areas at the two study sites. At Bear Springs, the average distance from the nearest springhead to the location at which salamanders were encountered was 105.4 meters (range 4.0-644.8 meters, n=42), while at Estes Cave the average was 98.5 meters (range 35.3-253.6 meters, n=12) from the cave entrance.

Potentially notable among the salamanders found away from the study sites is one female found 196 meters from Bear Springs on 19 November 2004, about 5-6 meters from a flowing surface stream under a stone (0.3 x 0.45 meters) which was associated with 20 eggs, many of which contained visible embryos (Figure 19). While some experts examining the photographs do not believe that the eggs are those of a plethodontid salamander (Hillis, personal communication 2005), the matter remains unresolved at this time. A single egg was collected into 95% ethanol and is present stored in a -80 ºF freezer in hopes of future analysis revealing the true identity of the eggs. Snout-vent length for salamanders found away from the study sites averaged 45.5 mm (range 20.7-73.2 mm, n=30) and total length averaged 86.8 mm (range 30.5 – 141.1 mm, n=22).

Within the timed search areas, salamander snout-vent length (SVL) ranged from 20.7 to 73.2 mm ($\bar{x}$=40.77 mm, n=278) at Bear Springs and from 20.5 to 80.1 mm ($\bar{x}$=54.2 mm, n=115) at Estes Cave. Total length (TL) of salamanders in timed search areas ranged from 28.0 to 143 mm ($\bar{x}$=74.15 mm, n=270) at Bear Springs and from 34.9 to 159.8 mm ($\bar{x}$=102.2 mm, n=114) at

Estes Cave. Salamander mass (grams) in timed search areas ranged from 0.1 to 6.9 g ($\bar{x}$=1.7 g, n=233) at Bear Springs and from 0.1 to 9.9 g (mean=3.45 g, n=108) at Estes Cave. Volume (mm^3) of salamanders in timed search areas ranged from 0.1 to 7.0 mm^3 ($\bar{x}$=1.66 mm^3, n=232) at Bear Springs and 0.1 to 9.8 mm^3 ($\bar{x}$=3.44 mm^3, n=93) at Estes Cave.

The smallest individual with a mental gland (thus a male) is 47.9 mm SVL, and the smallest individual that was and obviously a gravid female was 49.6 mm SVL.

The size distributions for the four metrics differ among sites (Table 1), with Bear Springs having a smaller averages than Estes Cave for all four metrics (Figures 20, 21). Snout vent length (SVL) at Bear Springs was characterized by a large number of individuals in the 24-32 mm size range, with a second, smaller peak around 44 mm (Figure 19A), while few salamanders at Estes Cave were below about 43 mm SVL; the majority being in the 44-74 mm SVL range (Figure 20A). An almost identical pattern is seen for total length (TL) (Figure 20B), although the pattern is somewhat obscured, perhaps as a result of some individuals having lost the tips of their tails, which were found to be in various states of regrowth. For both metrics, the largest individuals are from Estes Cave. For mass, a similar pattern is observed – at Bear Springs, there is a large peak in the 0.375 – 0.875 range, with a lesser peak around 1.375 – 1.875 g, and perhaps a smaller peak at a slightly heavier size range (Figure 21A), with few individuals over 4.5 g, while Estes Cave samples contain relatively few individuals under 2.5 g mass, with no discrete peaks being readily discernible. All of the individuals heavier than about 6.5 g, with one greater than 9.7 g, are reported from Estes Cave. The volume measurements (Figure 21B) exhibit a similar, but somewhat less clear, pattern with all of the individuals over 7.5 mm^3 being from Estes Cave.

The four size metrics showed a high degree of correlation (Figure 22) with some variation attributable to tail tip loss (affecting total length, mass, and volume) and some error attributable to measurement or data recording errors. The two length measures (total length and snout-vent length) were linearly correlated (Figure 22E) as were mass and volume (Figure 22F), while all other pair-wise correlations fit to a power curve (Figure 22A-D).

We could determine the unique identity of only 13 recaptured individuals from Bear Springs and 20 recaptured individuals from Estes Cave. All of these salamanders were recaptured once with the exception of two Estes Cave salamanders that were captured twice, and thus we can assess growth rates (Figure 23A). Recaptures of these individuals occurred from 34 to 462 days after initial marking, with this interval averaging 186.5 days. The snout-vent length of these individuals increased from 0.2 to 43.2 mm between capture events, averaging 7.83 mm. Average growth rate was 17.5 mm per year, with smaller individuals tending to have higher growth rates at the time of their recapture than larger individuals (Figure 23B). The data in Figure 23B, in combination with the assignment of yearly generations to data in Figure 20A, allows us to get some idea of the age of the largest individuals encountered during this study. Actual calculation of a growth model will be done after a third year of sampling, currently underway. Because cohorts of same-year individuals are obscured in Figures 20 and 21 as a result of combining all sample periods, we replotted the data from January 2005 and February 2004 to try to discern peaks for first and second year animals (Figure 24). From these graphs, we can see that first year animals during these sample periods ranged in size from 19 to <33 mm SVL, with a modal value of about 28 mm SVL. Second year individuals during these sample periods ranged from 33 to <59 mm SVL with a modal value of about 47 mm SVL.

The population estimates and confidence intervals for both Bear Springs and Estes Cave are shown in Table 2 and are based on calculations using both the Schnabel and the Schumacher & Eschmeyer population estimation models (Appendix 1). Both sites had **y**-intercepts not significantly different from zero (Figure 25), but the regressions were both non-significant. Thus, both sites showed a deviation from the assumptions of equal catchability and closure.

At Estes Cave, we saw no evidence of current human activity or livestock use of the area, other than impacts from our own visits and those of Reddell. At Bear Springs, we observed two major impacts to the habitat during the course of our study. First, someone came in with a bulldozer and moved around a great deal of soil, apparently in an attempt to control the flow route of the spring runs. Our observations suggest that this disturbance was followed by an increase in the number of red imported fire ant (*Solenopsis invicta*) mounds in the vicinity of Bear Springs. Further, this action destroyed *P. albagula* habitat, by covering loose, moist talus with relatively impermeable (to salamanders) soil. The second disturbance we observed was the use of the spring run by cattle. Hoof prints of cattle were observed on numerous occasions well up into the areas where we regularly found *P. albagula* in good numbers, and the damage from livestock activity appears to be degrading the habitat. A well used livestock trail crosses the spring run just upstream (ca. 1-2 meters) of the retention pond shown in Figure 1, and this trail has been in use since we first visited this site in April of 2002 (Taylor and Phillips 2002). The area around the north and northeast sides of the retention pond is normally densely covered with a luxuriant growth of horsetails, ferns, etc. During one of our visits, we noted that most of this vegetation had been browsed down to the ground and the area was heavily trampled by cattle – during later visits we observed some regrowth of the vegetation, but it has not fully recovered. One fire ant colony was noted within 1 meter of the west springhead.

We had an opportunity to visit two more remote sites in southeastern Fort Hood, Tweedle Dee Cave and Tweedle Dum Cave, where Reddell has recently discovered additional populations of the salamanders. The salamanders we observed in those caves had much more apparent light spots (Figure 26B) relative to typical salamanders found at Bear Springs and Estes Cave (Figure 26C, cover photo), which nearly or completely lack light spots. The absence of light spots is also typical of the caves where we recorded *P. albagula* in our earlier study (Taylor and Phillips 2002), with one notable exception (Figure 26A).

Discussion

Large numbers of salamanders were reported from the two sites, and numbers in our analyses are fairly robust for such a cryptic organism. The salamanders were found in numbers only during the cooler months of the year, perhaps in association with relatively moist conditions. While salamanders were almost never found directly in the water, they were associated with moist habitats – searches further away from the study sites yielded the most specimens during cool/moist conditions; under stones and logs, or in association with seeps or a surface stream.

The presence of salamanders up to 644 meters away from known karst features suggests that they are broadly distributed in karst terrain under suitable conditions of moisture and temperatures, but perhaps largely unavailable for sampling. We suspect that animals move away from hot, dry surface conditions deeper down into the cracks and pores in the karstified bedrock, where they would not likely be encountered except where such habitats intersect enterable caves. During cool, moist conditions, it may be possible to find these salamanders under stones and logs in suitable habitat where they have previously been overlooked – north facing slopes with springs and seeps seem to hold the greatest promise.

If the eggs found with the female salamander 196 meters from Bear Springs prove to be *P. albagula* eggs (determination awaits molecular analysis), this would provide new data on clutch size and seasonality of reproduction.

The broad distribution of snout vent lengths (SVL) at both sites (Figure 20A) suggests that the overall age structure of the two populations is healthy. Using data for *Plethodon glutinosus* from Florida (Highton 1956), the smallest individuals in Figure 20A (20 to 40 mm SVL) probably hatched in the late fall to January, prior to their capture and are in their first year of growth. Individuals 40 to 60 mm SVL would likely be in their second year of growth and sexually mature. Individuals above 60 mm SVL are at least in their third year of growth, and our data suggest that the oldest individuals are likely five or more years old.

The separation of the size classes is blurred by the fact that this graph contains data for all sampling periods combined. Therefore individuals in their first spring during the February sampling were intermediate in size between first and second year cohorts when they were measured in April. Reexamining these data for only the months of January and February at Bear Springs (Figure 24) reveals clearer peaks for the first two age cohorts, with slightly different size ranges. Collectively, our results (Figures 21, 22, 23, 24) suggest that the largest individuals encountered in our study may be five years old or older, but growth model analyses will not be conducted until the completion of ongoing sampling of a third year of data from the two sites.

The differences between Bear Springs and Estes Cave in size class distribution (Figures 20 and 21) are suggestive of differences in how the salamanders utilize these different habitats. While Bear Springs has a perennial supply of flowing water, flowing water is only apparent at Estes Cave during or shortly after rain events. It may be that the higher moisture levels at Bear Springs offer more suitable habitat for reproduction in this species, although this is clearly speculative at present.

The total number of salamanders in the Bear Springs population probably exceeds 300 based on two different models used in estimating population size. At Estes Cave, the population is composed of around 70 to 80 individuals. But both of these figures are, perhaps, underestimates because the populations are not closed populations, as is demonstrated by the salamanders encountered outside of the timed area searches and the regression of the recapture rate at time **i** and the cumulative number of individuals available for recapture at that time. These two sites were chosen for this study because we had *a priori* knowledge that they seemed to harbor more salamanders than other known *P. albagula* sites at Fort Hood. Given that the species has been previously recorded from 16 sites at Fort Hood (Taylor and Phillips 2002), and more recent work by Reddell (unpublished) has yielded additional sites in more remote portions of southeastern Fort Hood, it is likely that hundreds of salamanders occur on base beyond those recorded from Bear Springs and Estes Cave.

Management considerations for these populations should focus on threats including habitat destruction and exotic species (i.e., the red imported fire ant). The vast majority of salamanders were found underneath stones along the edges of spring runs and seeps. This environment, particularly at Bear Springs where the majority of salamanders occur, is in danger of excessive trampling by cattle, and has also been damaged by bulldozer activity. We observed that stones in the path of cattle were compacted into the mud and dirt and almost never had salamanders associated with them. Conversely, stones in the same survey zone adjacent to the trunks of trees or tucked up ledges away from where cattle travel did harbor salamanders. It is not possible to separate the effects of the bulldozing activity and livestock activity, but again it seems that as a result of these disturbances, there has been an increase in the numbers of red imported fire ant mounds in the Bear Springs area over the course of our study. We recommend that the road from the top of the plateau down to the spring be blocked with large boulders at the top of the plateau, and that a similar blockage be placed in the valley floor below the spring near the closest access still available to four wheel drive vehicles. In addition, we feel that this habitat would be best protected by fencing the area around Bear Springs to exclude livestock, including all of the spring run area demarcated by dashed lines in Figure 1, along with the more sensitive habitat to the north and northeast of the retention pond. A narrow passage along the pre-existing livestock trail could remain accessible, and livestock access to the southwest side of the retention pond could be allowed without direct damage to the salamander habitat. Fencing of habitat should be done by hand, as additional vehicular traffic in the vicinity of Bear Springs will only increase disturbance levels and could result in still higher levels of fire ant infestation of this area. Occasional steam treatments of fire ant mounds should be considered in this area, although not during time periods when salamanders are abundant. In general, activities which disturb the surface of the ground should be limited to hot, dry summer months when the salamanders are out of reach.

Conclusions & Research Recommendations

New information on the population levels, distribution, and life history of Fort Hood *Plethodon albagula* populations, which constitute a potentially new species of *Plethodon*, will help land managers monitor changes over time and minimize impact of military activities around sensitive areas. This report adds to the body of knowledge about this species, including population estimates, size class distributions, growth, seasonality, habitat use, and sensitivity to disturbance.

This study reports, for the first time, the occurrence of salamanders at Fort Hood well away from known karst features, raising questions about what these moisture sensitive animals do when the surface environment is too hot and dry. They may simply burrow deeper into the subsurface (the soil or interstices between talus blocks, or enlarged joints or fractures in the bedrock) while remaining in the same small area, or perhaps they move much greater distances (overland during moist periods or through enlarged joints and fractures in the bedrock) from less permanently habitable environments to perennially cool/moist areas. A study of population genetics would reveal the degree of connectivity among known populations, and thus would facilitate the design of management units prioritizing conservation of maximal genetic diversity. We have also characterized the habitat where the salamanders were found, including the distance to nearest water, and general observations of the high-humidity microhabitats in which they are usually found. However, we still know nothing of the food choice and food resource availability of Fort Hood *P. albagula* populations. Diet and resource availability can affect population health and be an indicator of more subtle impacts to the species' habitat. Finally, the population estimates presented here are for two sites known to have the high numbers of individuals. It would be helpful to monitor sites in other areas for comparison. Population levels at such sites may be more typical of the average Fort Hood *Plethodon* locality, and therefore could be applied to the majority of sites where the species was known.

Table 1. Mann-Whitney-Wilcoxon tests comparing Bear Springs and Estes Cave (Fort Hood, Texas) *Plethodon albagula* populations based on the values for four size metrics measured on animals from the timed area searches (February 2004 – July 2005).

Metric	Site	$\bar{x}$±Std.Err.	n	z	p
Snout-vent length (mm)	Bear Springs	40.76±0.78	278		
	Estes Cave	54.17±1.30	115	8.062	<0.0001
Total length (mm)	Bear Springs	74.15±1.63	270		
	Estes Cave	102.24±2.67	114	8.165	<0.0001
Mass (g)[1]	Bear Springs	1.70±0.10	233		
	Estes Cave	3.45±0.20	108	7.447	<0.0001
Volume (mm^3)[1]	Bear Springs	1.66±0.10	232		
	Estes Cave	3.44±0.24	93	6.806	<0.0001

[1]Variances were unequal, so the Satterthwaite method of calculating the t value was used, reducing the degrees of freedom.

Table 2. Population estimates and 95% confidence interval for *Plethodon albagula* at two sites at Fort Hood, Texas.

Site	Model: Schnabel Estimate	Model: Schnabel 95% Conf. Interval	Model: Schumacher- Eschmeyer Estimate	Model: Schumacher- Eschmeyer 95% Conf. Interval
Bear Springs	331	(250<N<490)	318	(234<N<499)
Estes Cave	71	(55<N<102)	80	(60<N<123)

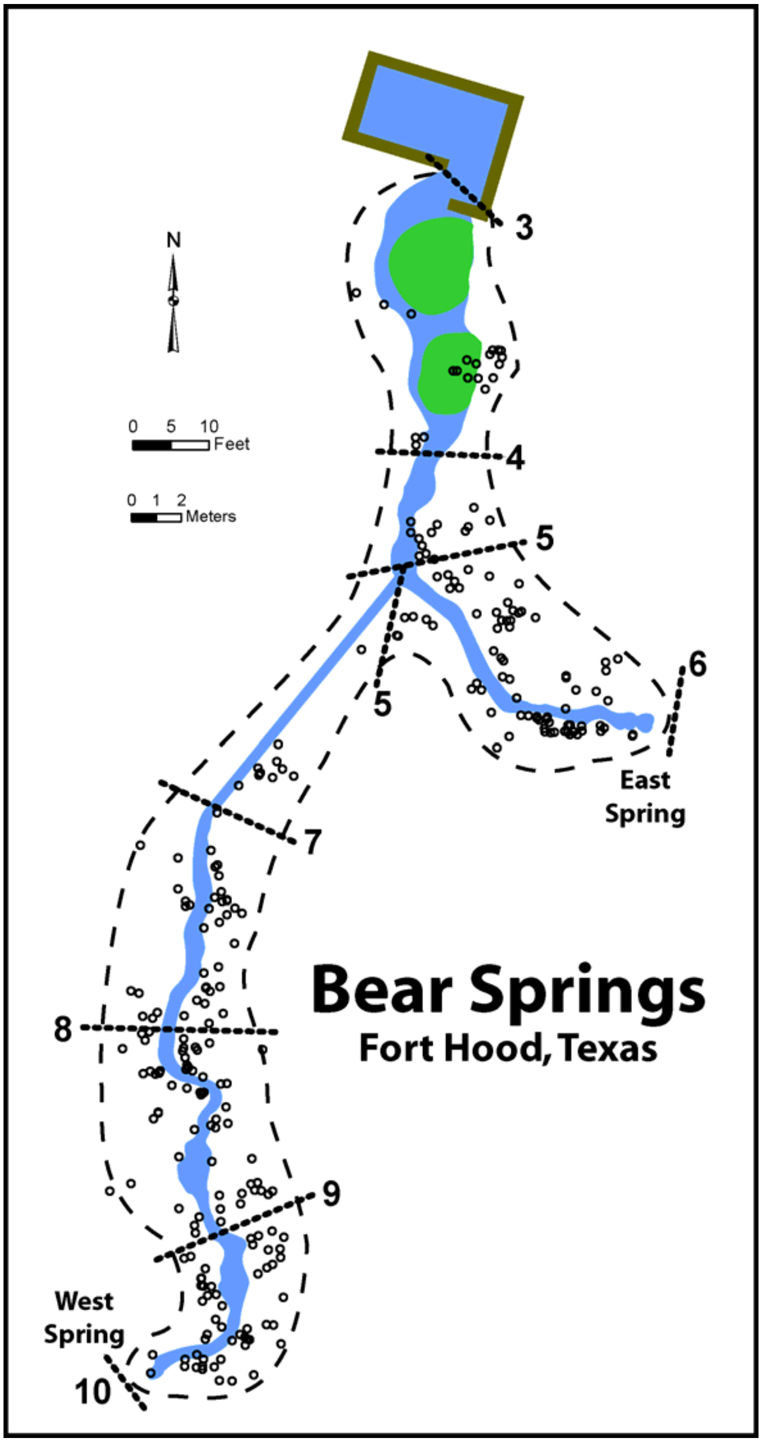


Figure 1. Study site at Bear Springs with two springheads labeled, man-made retention wall around pool to north overflows onto a steep slope leading to stream in ravine. Green areas near pool are watercress. Open circles represent locations where salamanders were found, all sample dates combined. Dashed lines represent approximate boundary of search area. Numbered, dotted lines represent boundaries of search zones.

Figure 2. Searching habitat in zone 3-4 at Bear Springs on 18 February 2004. Note watercress in foreground.

Figure 3. Searching for salamanders at Bear Springs – note the wire flags marking the locations of individual salamanders.

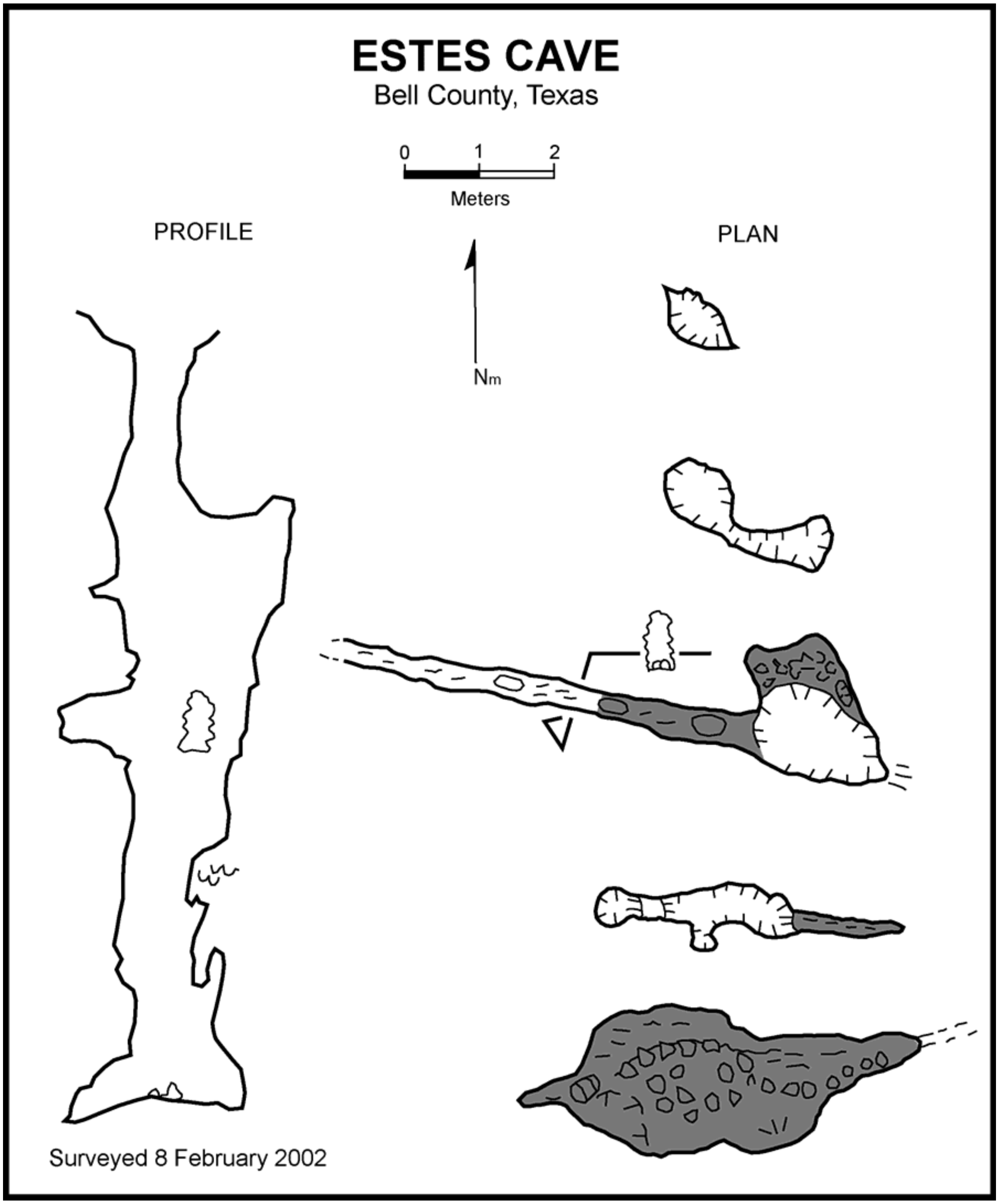


Figure 4. Estes Cave - search areas on bottom and two ledges are shaded in gray. No salamanders were seen on the walls of the cave (excluding ledges), and the wall are not included in the timed search.

Figure 5. Weighing salamander with Pesola spring scales. The weight of the bag is subtracted.

Figure 6. Measuring salamander snout-vent and total length with digital calipers.

Figure 7. Measuring volume of salamander using graduated cylinder and water displacement.

Figure 8. Kit for injecting dye marks in salamanders.

Figure 9. Syringes filled with three colors of latex dye in preparation for marking salamanders..

A **B**

Figure 10. Dye-marked salamanders. A. GnGG+   B. YYRR+ .

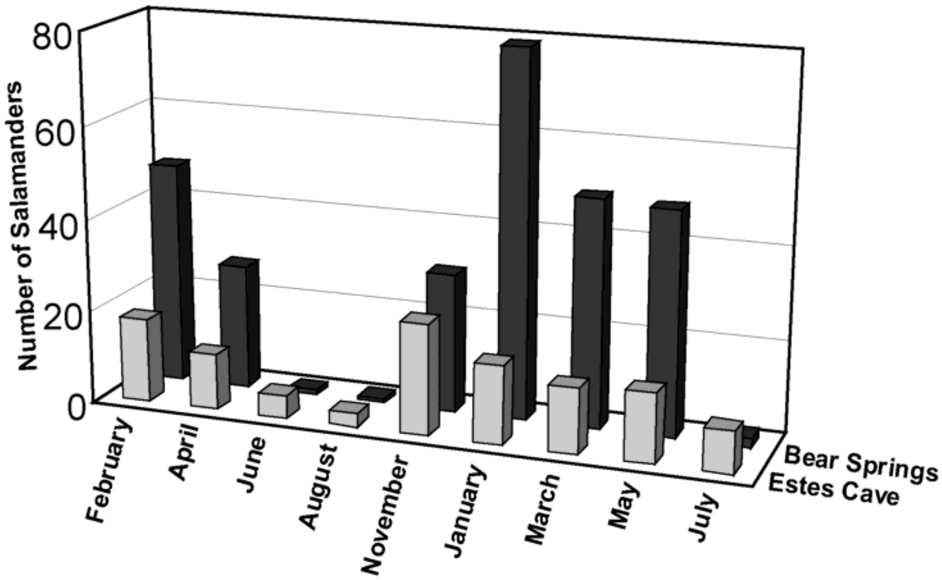


Figure 11. Total number of salamanders observed in the field (i.e., includes individuals which could not be caught) at Fort Hood, Texas during periods (February 2004 through July 2005).

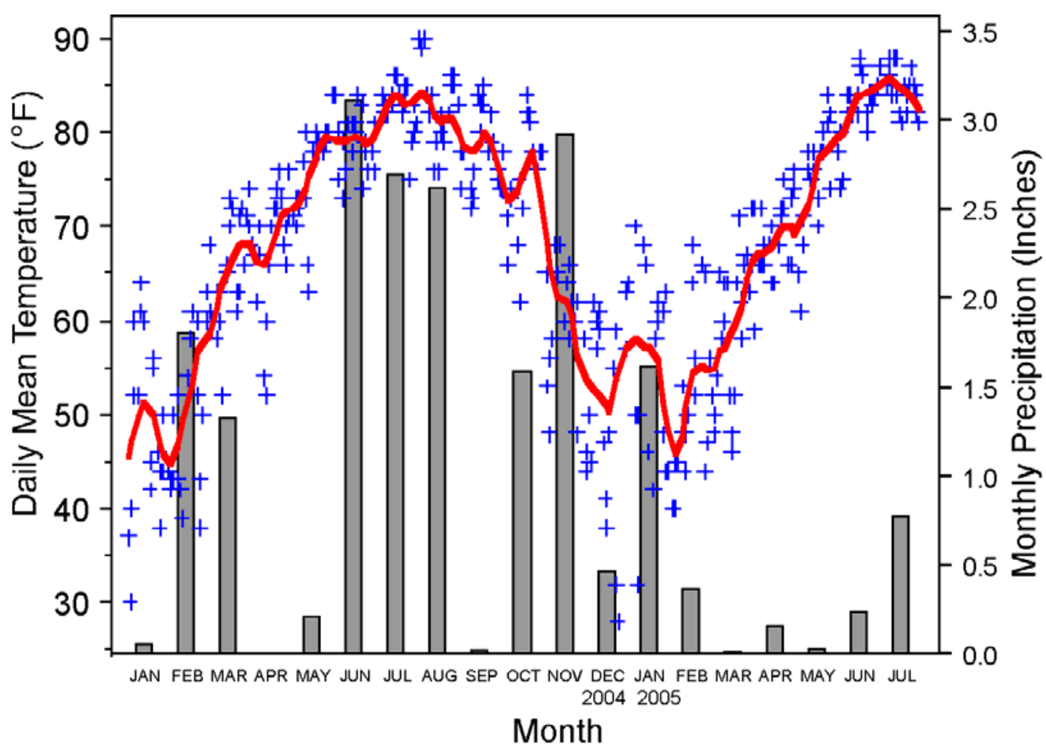


Figure 12. Historical temperature (red line, blue crosses) and precipitation (gray bars) for Killeen, Texas (data from Fort Hood/Killeen airport weather station KHLR). Red line is Loess smoothed best fit line of daily mean temperature.

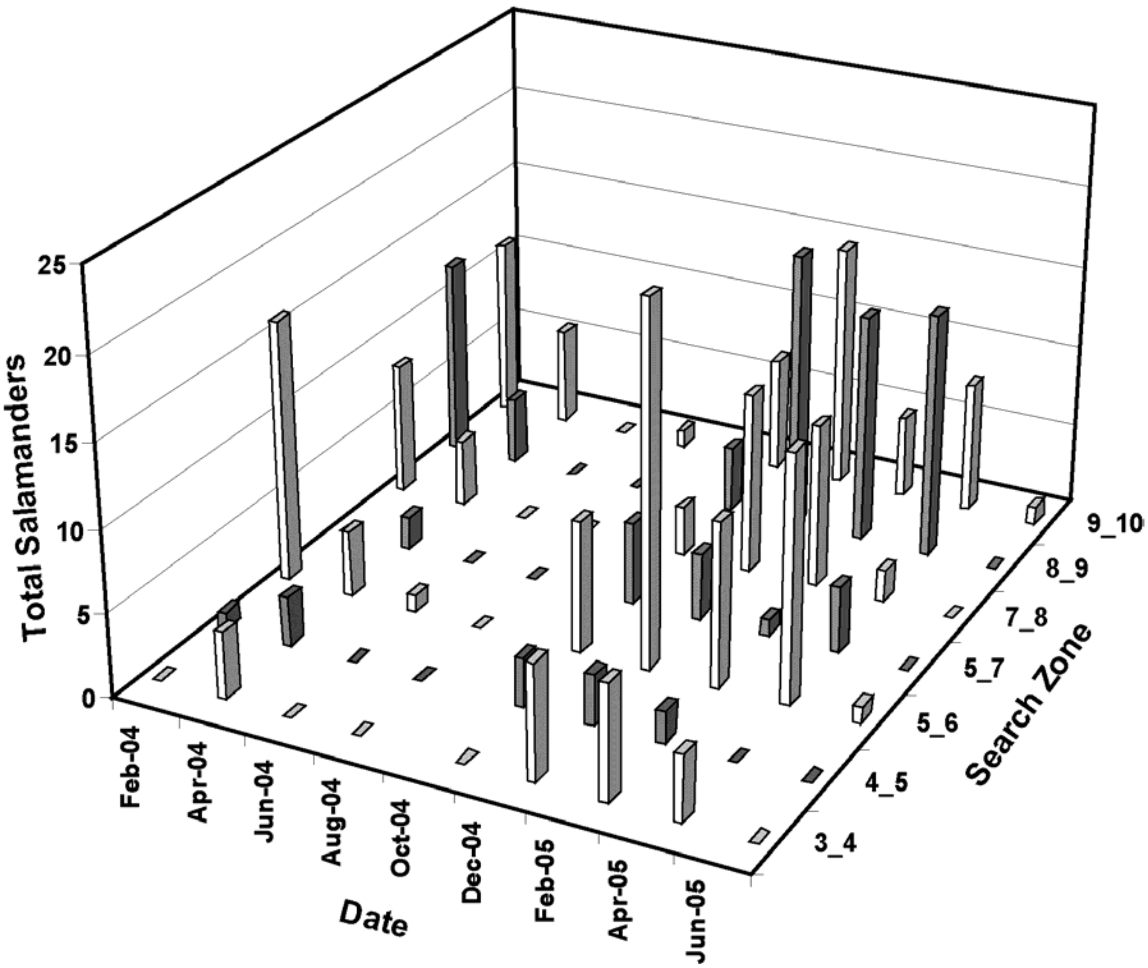


Figure 13. Numbers of salamanders found by sample date and search zone (see Figure 1) at Bear Springs, Fort Hood, Texas. Zones 5-6 and 9-10 are the closest to the springheads.

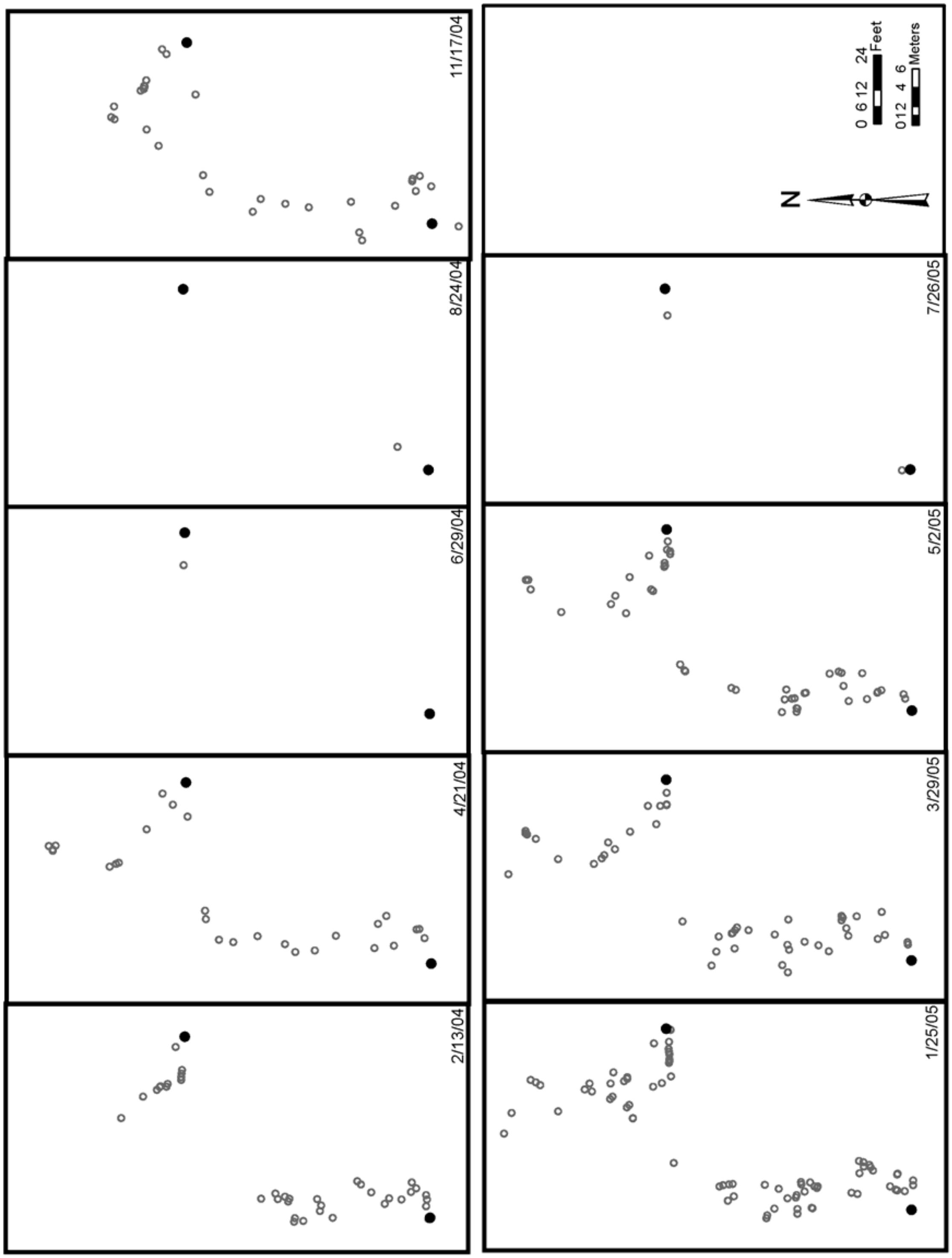


Figure 14. Distribution of salamanders found at Bear Springs by sample date. Refer to Figure 1 for overall distribution. The filled black dots represent the east and west springheads, open circles are salamander encounters. Note that salamanders are more widely distributed in the cool/moist periods of the year.

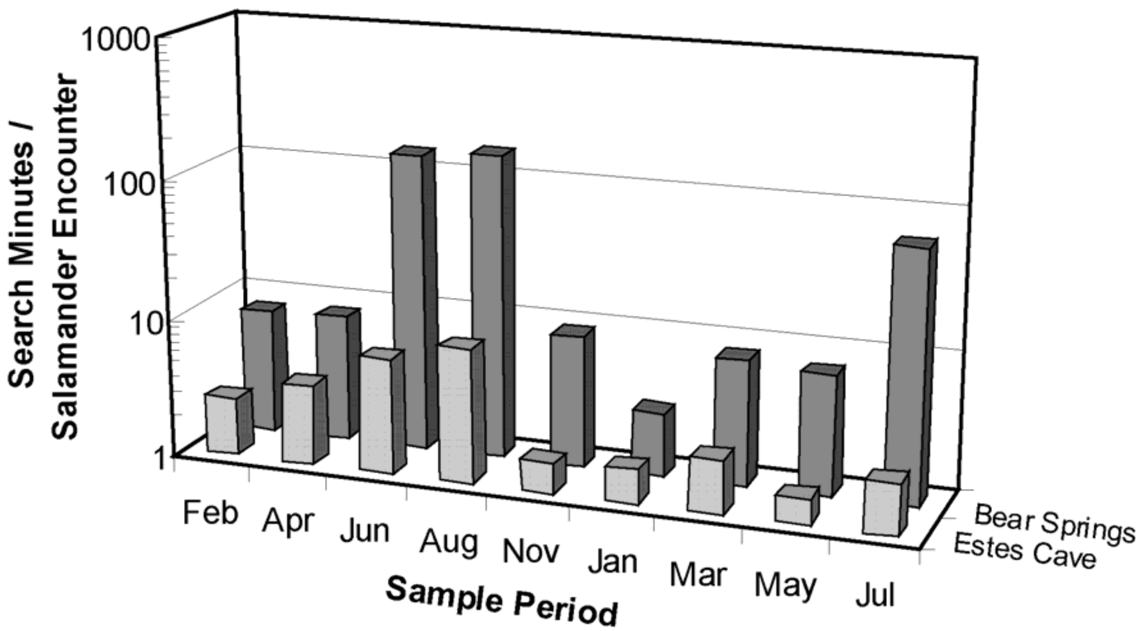


Figure 15. Number of search minutes per salamander encountered (note Log_{10} scale) for all sample periods from February 2004 through July 2005 and Bear Springs and Estes Cave (Bell County, Fort Hood, Texas).

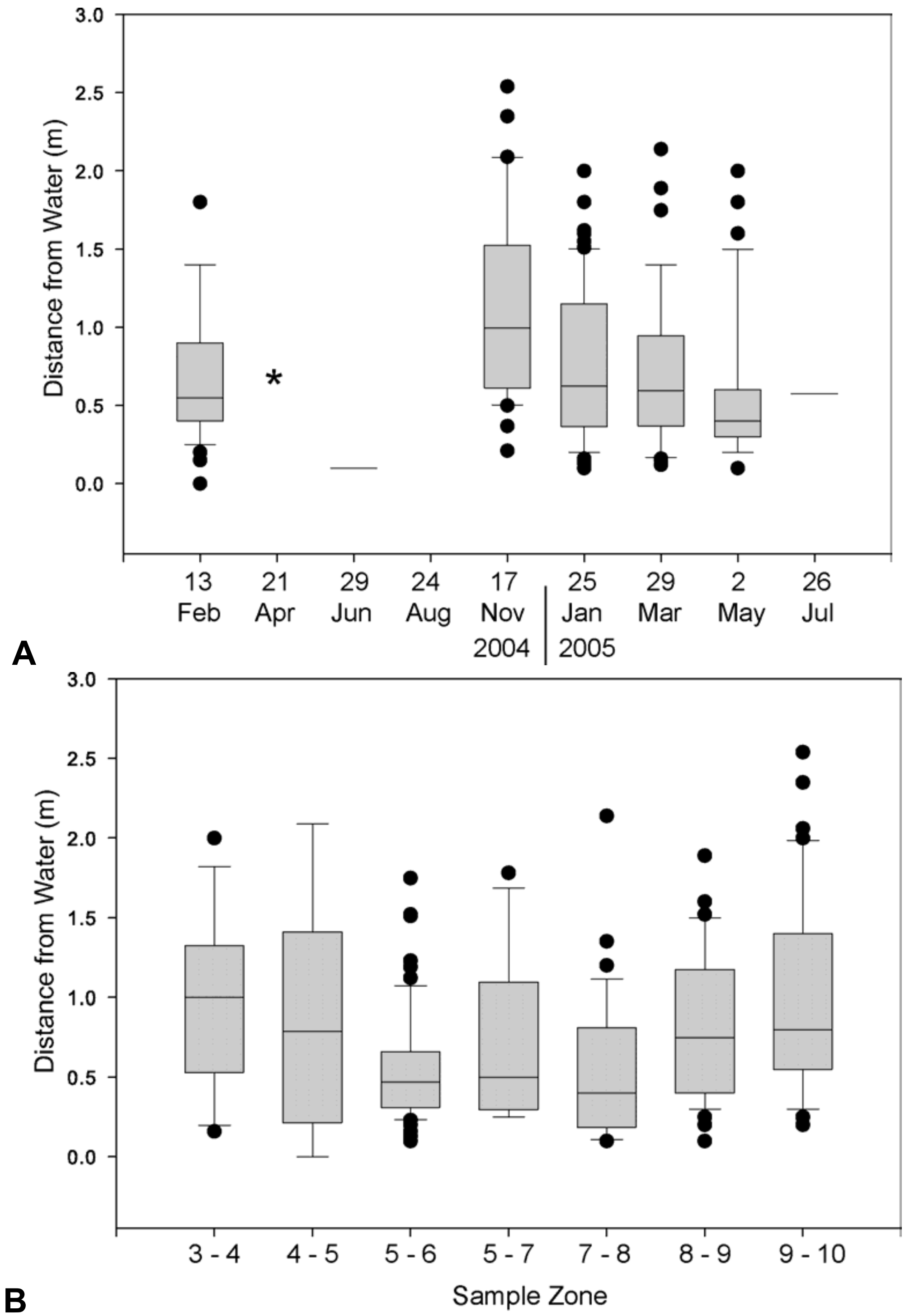


Figure 16. Boxplots showing distance from water at which salamanders were found at Bear Springs, Fort Hood, Texas. A. Distance by sample date (* = distance from water not recorded in April 2004 sampling period). B. Distance from water by sample zone (see Figure 1). Zones 5-6 and 9-10 are the closest to the springheads.

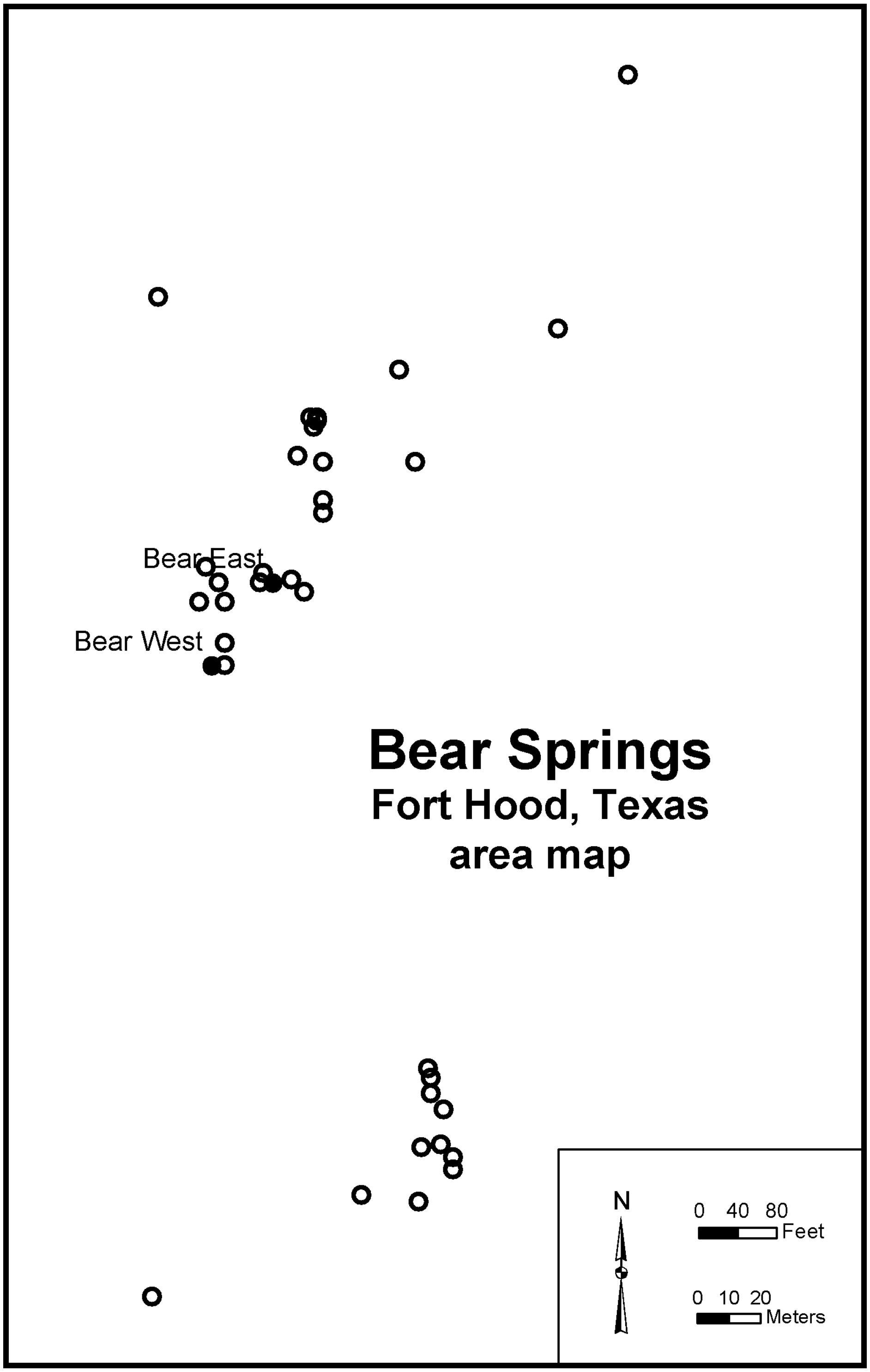


Figure 17. Bear Springs area map, showing locations at which salamanders were found during occasional searches away from the spring-run. Inappropriate (typically dry) habitat was not closely examined. Filled black dots represent the east and west springheads, open circles represent salamander encounters.

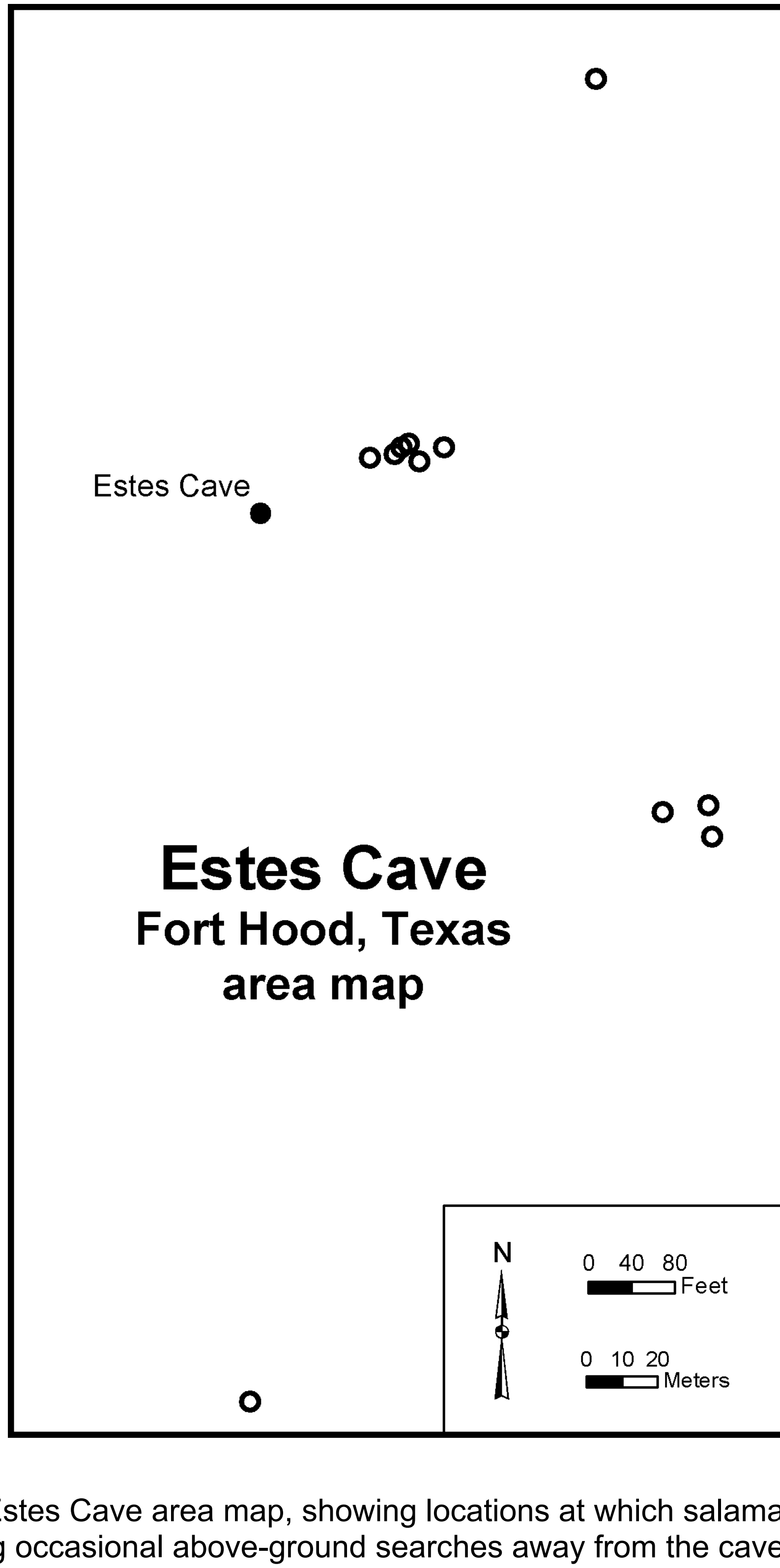


Figure 18. Estes Cave area map, showing locations at which salamanders were found during occasional above-ground searches away from the cave. Filled black dot represents Estes Cave, open circles represent salamander encounters.

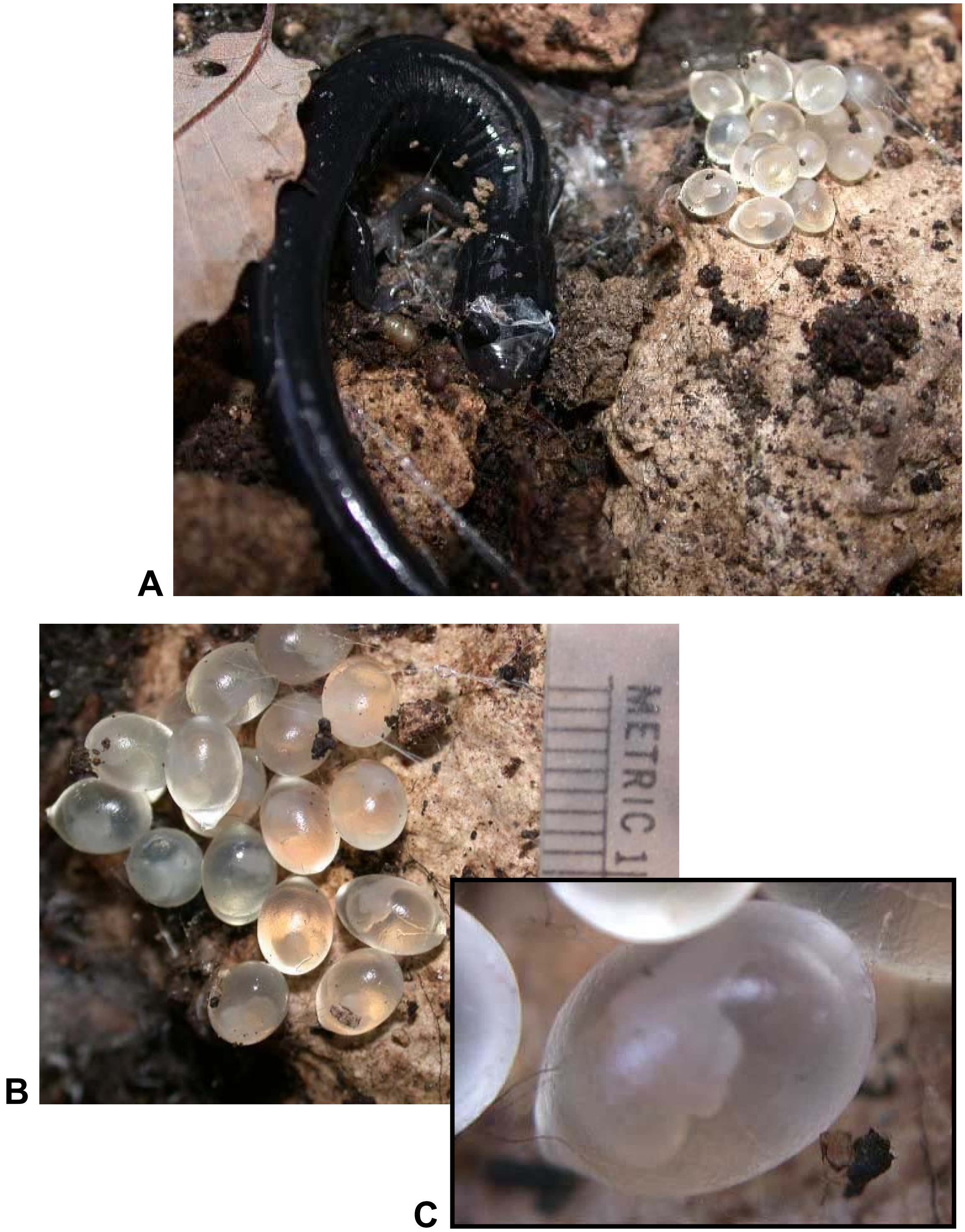
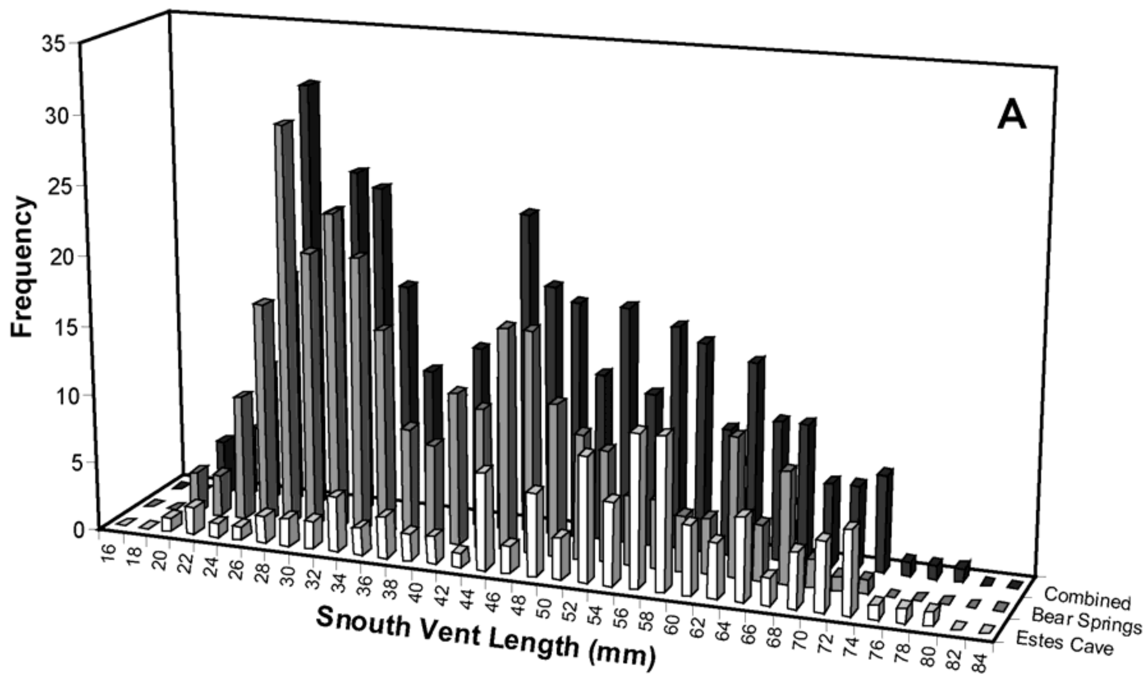


Fig 19. *Plethodon albagula* and eggs at Fort Hood, Texas on 19 November 2004. Whether or not the eggs pictured here are really salamander eggs has been questioned by some authorities. A. Female salamander *in situ* under stone with eggs, as discovered in the field 196 meters away from Bear Springs. B. Close-up of egg cluster, minor divisions on ruler are 1mm. C. Close-up of a single egg with embryo visible. Photos by Steve Taylor.



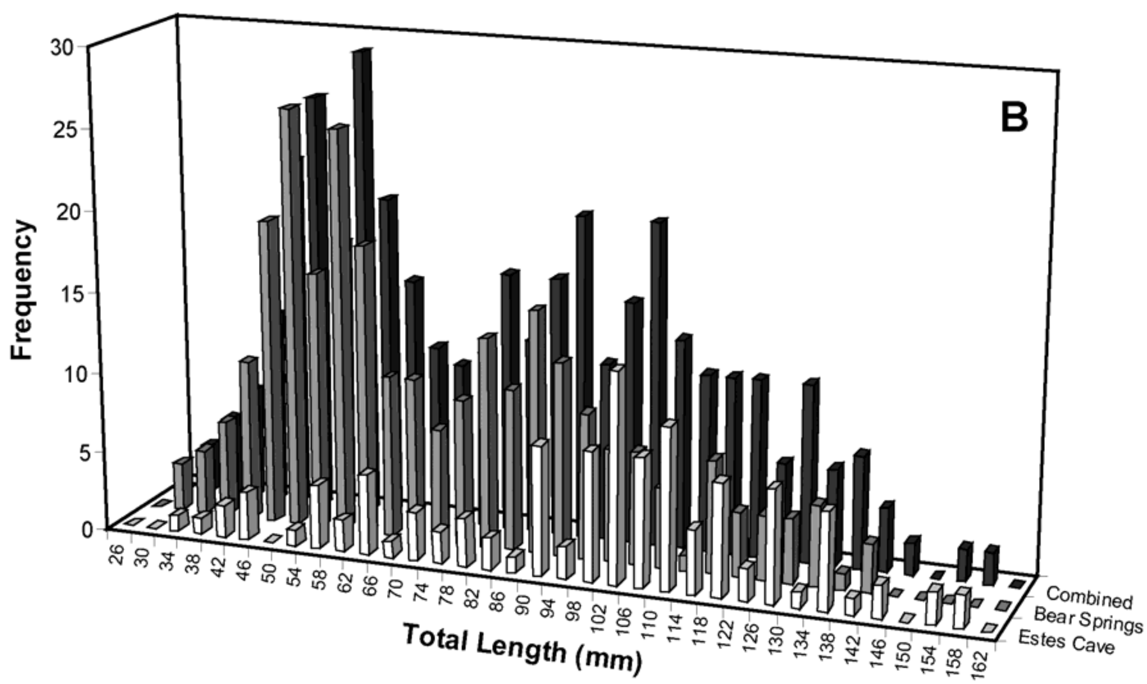


Figure 20. Size class frequencies for snout vent length (A) and total length (B) of salamanders from timed area searches at Bear Springs and Estes Caves (Fort Hood, Texas), based on sampling from February 2004 through July 2005.

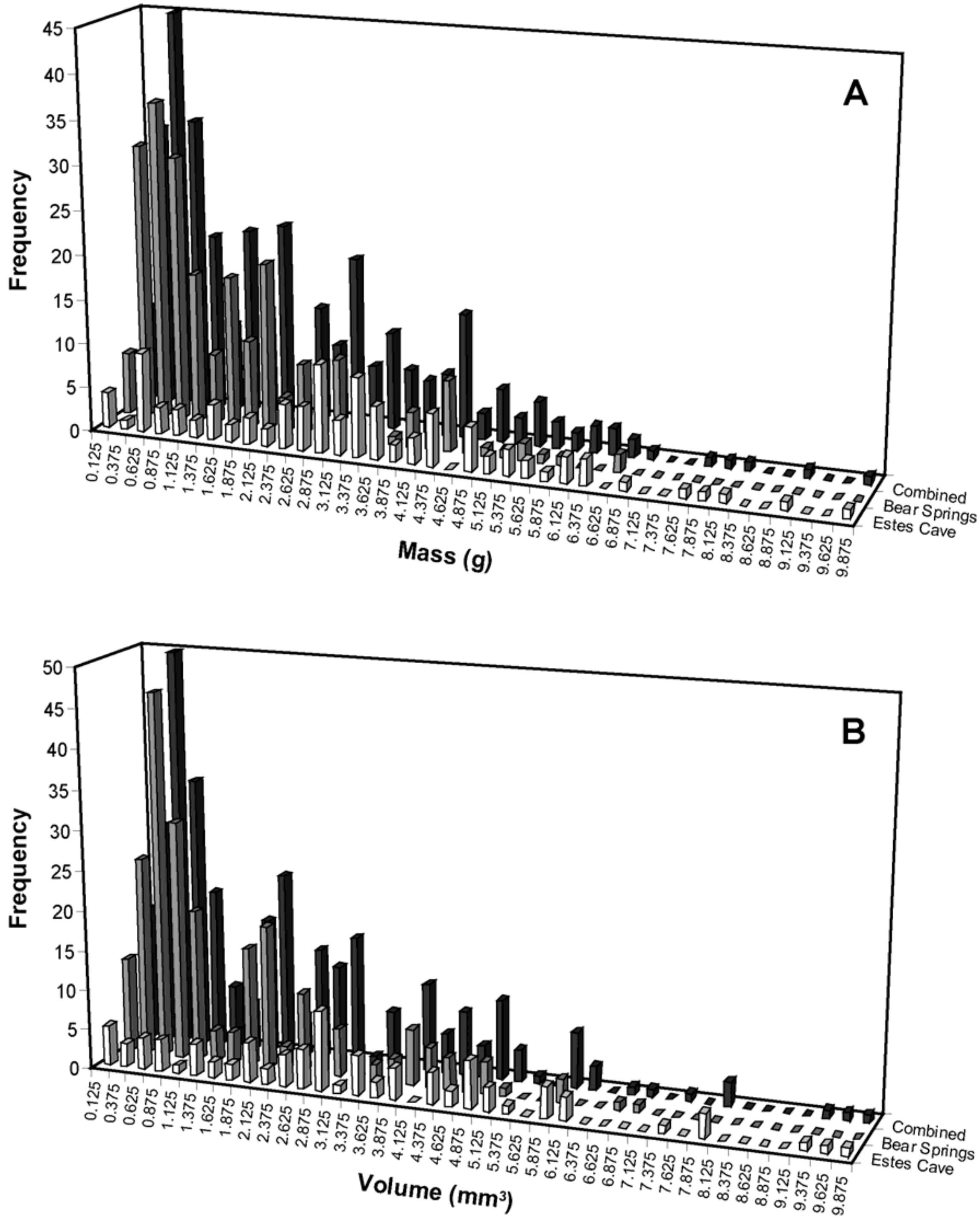


Figure 21. Size class frequencies for mass (A) and volume (B) of salamanders from timed area searches at Bear Springs and Estes Caves (Fort Hood, Texas), based on sampling from February 2004 through July 2005.

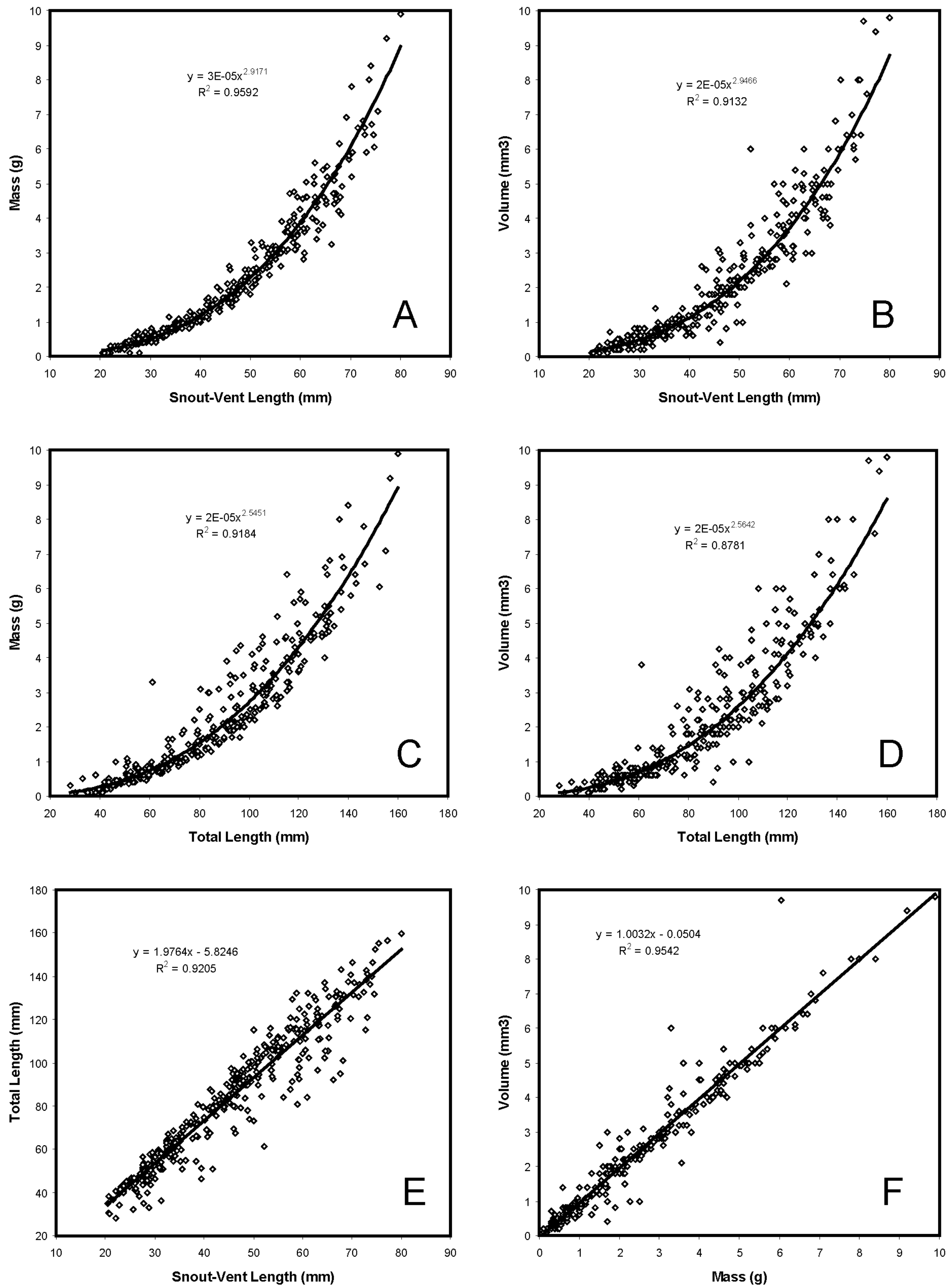


Figure 22. Correlations among size metrics of *Plethodon albagula* from timed-area search samples at Bear Springs and Estes Cave, combined. Best fit lines for A-D are power curves, for E and F are linear regressions.

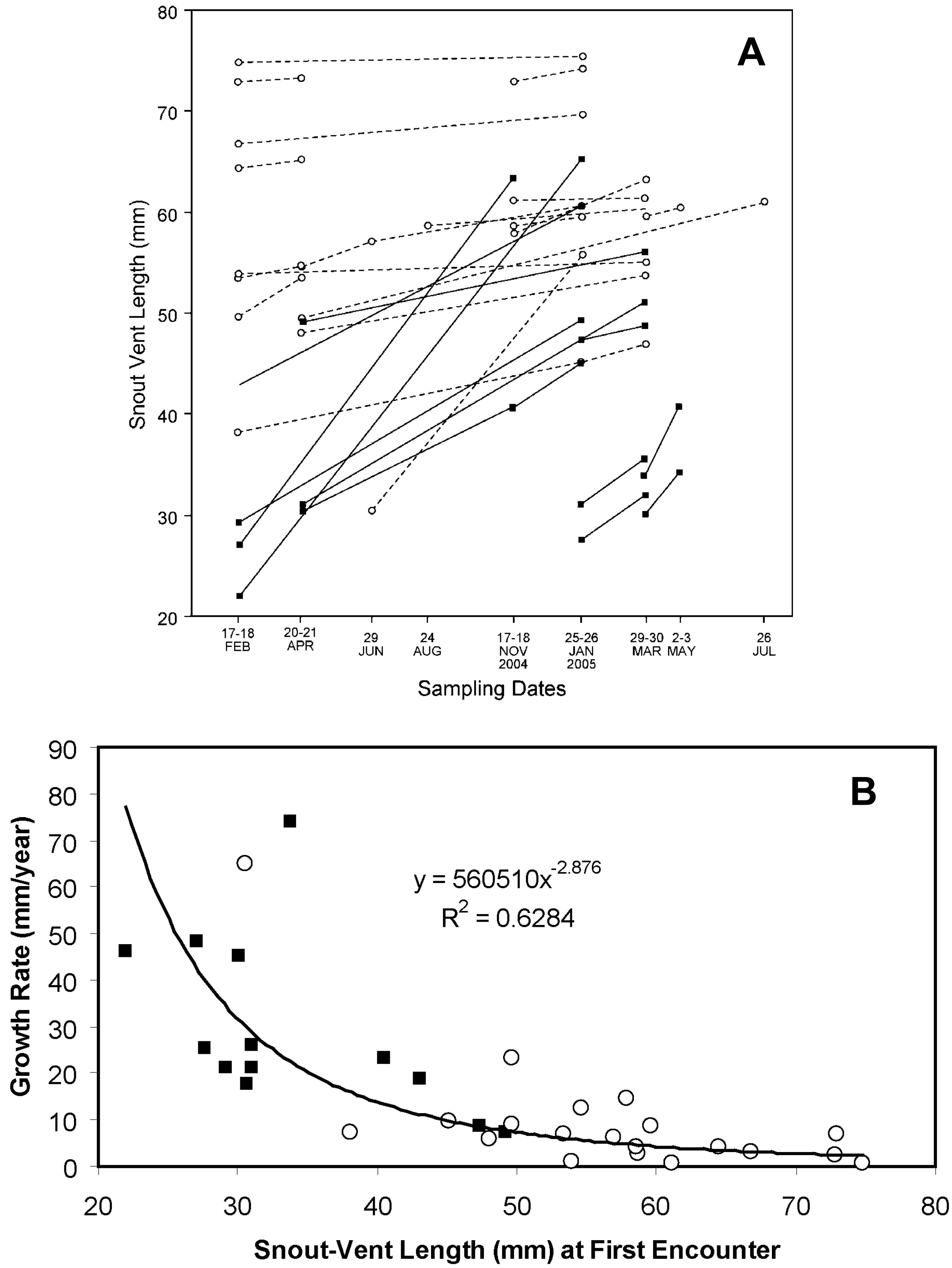


Figure 23. Thirty-three salamanders (Bear Springs=■, n=13; Estes Cave=○, n=20) whose unique identity could confidently be determined upon recapture. A. Initial capture and recapture dates in relation to growth (snout-vent length). B. Size (snout-vent length) at initial capture in relation to growth rate (= slope of lines in part A), best fit line is a power curve.

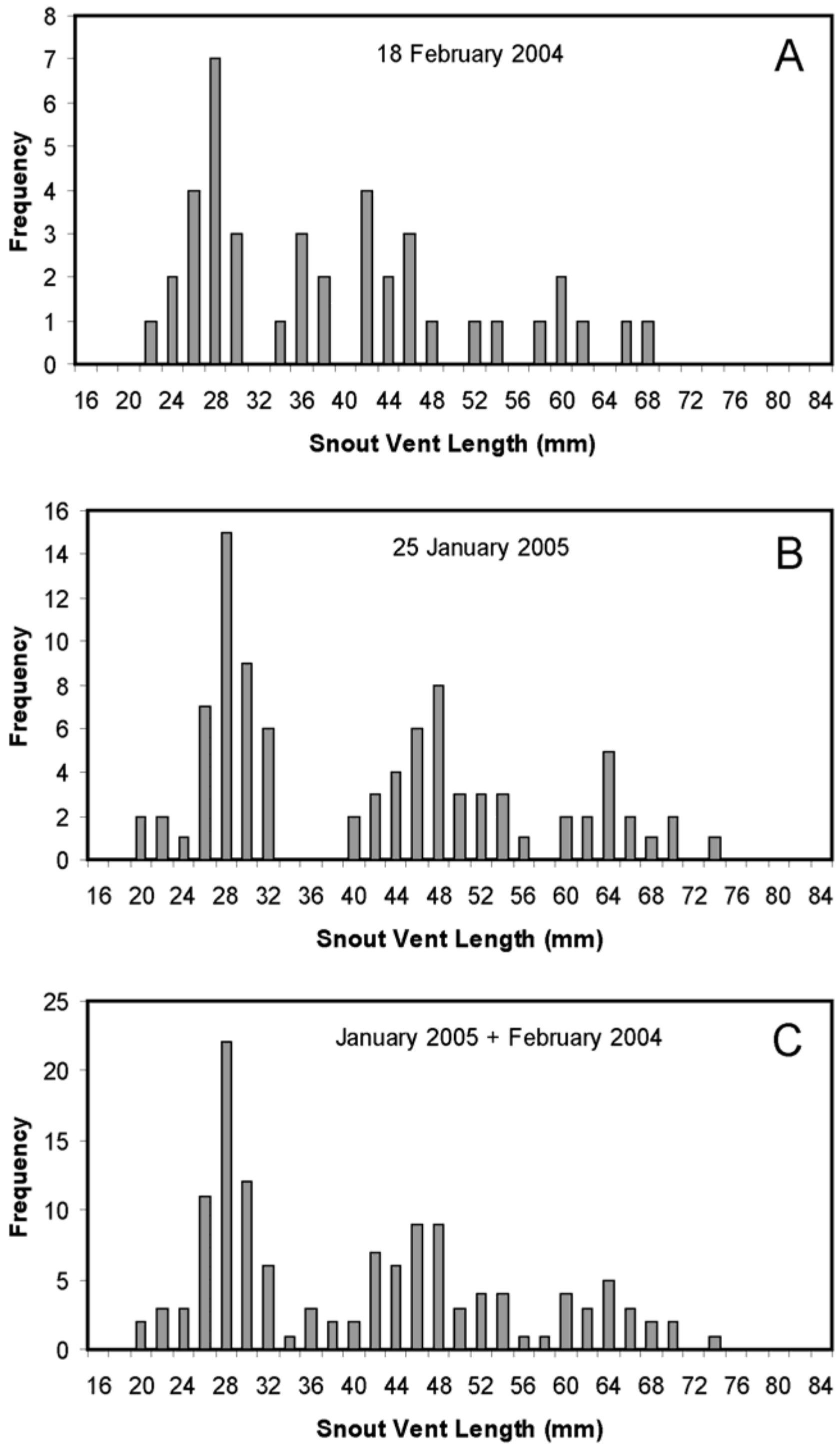


Figure 24. Size class distributions for Bear Springs salamanders collected on 18 February 2004 (A), 25 January 2005 (B), and the data for these two samples combined (C). Note distinct peaks corresponding to first and second year cohorts.

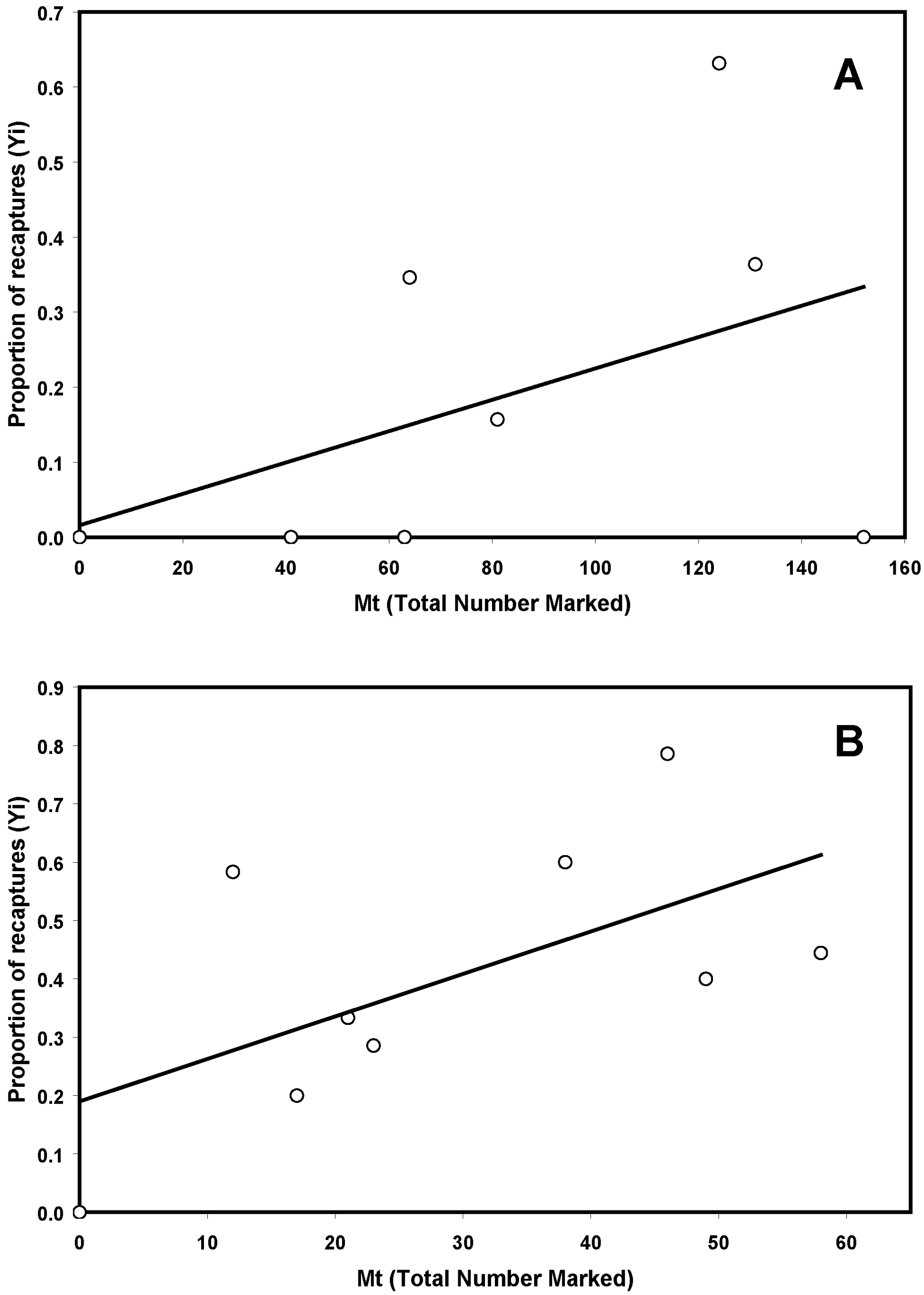


Figure 25. Linear regression of recapture rate at time **i** and the cumulative number of individuals available for recapture at that time. A. Bear Springs, B. Estes Cave.

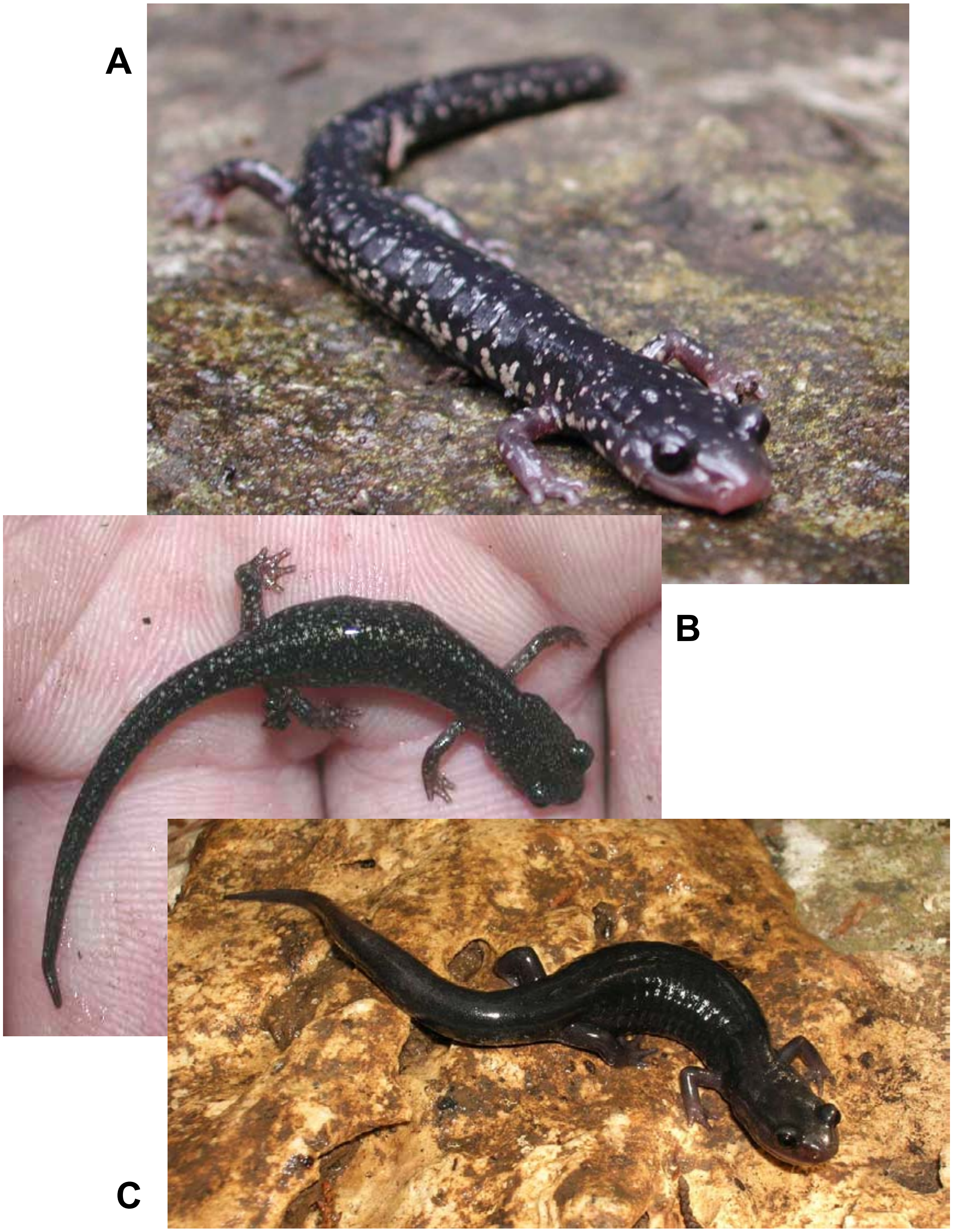


Figure 26. Variations in pigmentation of Fort Hood *Plethodon albagula*. A. Salamander (23 April 2004) with unusually distinct markings – most salamanders in this part of Fort Hood lack such distinct spots. B. A young salamander from Tweedle Dee Cave (28 January 2005) – most salamanders from this area have distinct spots. C. A salamander from Estes Cave (26 July 2005) – all salamanders encountered at this cave and at Bear Springs have few or no spots.

Acknowledgements

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Appendix 1. Calculations for population estimates.

Bear Springs:

CD	Date	Ct	Rt	Ut	Mt	CtMt	$CtMt^2$	Rt^2/Ct	RtMt	yi
1	2/18/2004	41	0	41	0	0	0	0	0	0.000
2	4/21/2004	22	0	22	41	902	36982	0	0	0.000
3	6/29/2004	1	0	1	63	63	3969	0	0	0.000
4	8/24/2004	0	0	0	64	0	0		0	
5	11/17/2004	26	9	17	64	1664	106496	3.115	576	0.346
6	1/25/2005	51	8	43	81	4131	334611	1.255	648	0.157
7	3/29/2005	19	12	7	124	2356	292144	7.579	1488	0.632
8	5/2/2005	33	12	21	131	4323	566313	4.364	1572	0.364
9	7/26/2005	1	0	1	152	152	23104	0	0	0.000
9	**Totals**	194	41			13591	1363619	16.31	4284	

Schnabel Population Estimate

N = å (C_tM_t) / å R_t = **331.488**

When the ratios C_t/N and M_t/N are both < 0.1 then use N = å (C_tM_t) / å R_t + 1

Variance 1/N = åR_t / å$(C_tM_t)^2$ = 2.21963E-07

SE of 1/N = Sq. Rt. of (Variance 1/N) = 0.00047113

95% Confidence Intervals = åC_tM_t / åR_t (When åR_t < 50 use Poisson Dist., Ecol. Methods Appendix 1.2).

When åR_t ³ 50 the CI = 1/N ± ta SE, then invert or take reciprocal of N (i.e. 1/x)

For Schnabel t values are based on (s - 1) degrees of freedom

Degrees Freedom = 8

T Value @ .05 (ta) = 2.074

95% Lower CI = 0.0039938 = **250.4**

95% Upper CI = 0.0020396 = **490.3**

Shumacher-Eschmeyer Estimate

N = å ($CtMt^2$) / å (RtMt) = **318.305**

Variance 1/N = å (R_t^2/C_t) - ((å$R_tM_t)^2$ / å ($C_tM_t^2$)) / s-2 = 2.854 0.407726362

SE of 1/N = Sq. Rt. of Variance / å ($C_tM_t^2$) = 0.000546812

CI = 1/N ± ta SE, then invert or take reciprocal of N (i.e. 1/x)

For Shumacher-Eschmeyer t values are based on (s-2) degrees of freedom

Degrees of Freedom = 7

T Value @ .05 (ta) = 2.080

95% Lower CI = 0.004279 = **233.7**

95% Upper CI = 0.0020043 = **498.9** 265.2 0.833

Test of Closure and Equal Catchability

Date	Mt	yi
2/18/2004	0	0
4/21/2004	41	0
6/29/2004	63	0
11/17/2004	64	0.346
1/25/2005	81	0.157
3/29/2005	124	0.632
5/2/2005	131	0.364
7/26/2005	152	0

SUMMARY OUTPUT

Regression Statistics

Multiple R	0.45
R Square	0.2
Adjusted R Square	0.07
Standard Error	0.23
Observations	8

ANOVA

	df	*SS*	*MS*	*F*	*p*
Regression	1	0.079	0.1	1.509	0.2654
Residual	6	0.316	0.1		
Total	7	0.395			

	Coeff	*SE*	*t*	*p*	
Intercept	0.02	0.161	0.1	0.925	Intercept not sig. different from zero
X Variable 1	0	0.002	1.2	0.265	Regression N.S. Violation in assumptions

Estes Cave:

CD	**Date**	**Ct**	**Rt**	**Ut**	**Mt**	**CtMt**	**CtMt^2**	**Rt^2/Ct**	**RtMt**	**yi**
1	2/17/2004	12	0	12	0	0	0	0	0	0.000
2	4/20/2004	12	7	5	12	144	1728	4.083	84	0.583
3	6/29/2004	5	1	4	17	85	1445	0.2	17	0.200
4	8/24/2004	3	1	2	21	63	1323		21	0.333
5	11/18/2005	21	6	15	23	483	11109	1.714	138	0.286
6	1/26/2005	20	12	8	38	760	28880	7.2	456	0.600
7	3/30/2005	14	11	3	46	644	29624	8.643	506	0.786
8	5/3/2005	15	6	9	49	735	36015	2.4	294	0.400
9	7/26/2005	9	4	5	58	522	30276	1.778	232	0.444
9	**Totals**	111	48			3436	140400	26.02	1748	

Schnabel Population Estimate

N = å (C_tM_t) / å R_t = **71.583**
When the ratios C_t/N and M_t/N are both < 0.1 then use N = å (C_tM_t) / å R_t + 1
Variance 1/N = åR_t / å$(C_tM_t)^2$ = 4.0657E-06
SE of 1/N = Sq. Rt. of (Variance 1/N) = 0.002016357
95% Confidence Intervals = åC_tM_t / åR_t (When åR_t < 50 use Poisson Dist., Ecol. Methods Appendix 1.2).
When åR_t ³ 50 the CI = 1/N ± ta SE, then invert or take reciprocal of N (i.e. 1/x)
For Schnabel t values are based on (s - 1) degrees of freedom
Degrees Freedom = 8
T Value @ .05 (ta) = 2.074
95% Lower CI = 0.0181517 = **55.1**
95% Upper CI = 0.0097878 = **102.2**

Shumacher-Eschmeyer Estimate

N = å ($CtMt^2$) / å (RtMt) = **80.320**
Variance 1/N = å (R_t^2/C_t) - ((å$R_tM_t)^2$ / å ($C_tM_t^2$)) / s-2 = 4.255 0.607914995
SE of 1/N = Sq. Rt. of Variance / å ($C_tM_t^2$) = 0.002080836
CI = 1/N ± ta SE, then invert or take reciprocal of N (i.e. 1/x)
For Shumacher-Eschmeyer t values are based on (s-2) degrees of freedom
Degrees of Freedom = 7
T Value @ .05 (ta) = 2.080
95% Lower CI = 0.0167783 = **59.6**
95% Upper CI = 0.008122 = **123.1** 63.5 0.791

Test of Closure and Equal Catchability

2/17/2004	0	0.00
4/20/2004	12	0.58
6/29/2004	17	0.20
8/24/2004	21	0.33
11/18/2005	23	0.29
1/26/2005	38	0.60
3/30/2005	46	0.79
5/3/2005	49	0.40
7/26/2005	58	0.44

SUMMARY OUTPUT

Regression Statistics	
Multiple R	0.6
R Square	0.4
Adjusted R Square	0.3
Standard Error	0.2
Observations	9

ANOVA

	df	*SS*	*MS*	*F*	*p*
Regression	1	0.158	0.2	3.911	0.088
Residual	7	0.283	0		
Total	8	0.442			

	Coefficients	*se*	*t*	*p*	
Intercept	0.2	0.127	1.5	0.179	Intercept N.S. Diff from zero - fine
X Variable 1	0	0.004	2	0.088	Regression N.S. Violation in assumptions