Herpetological Review

Volume 35, Number 3 — September 2004

and lodging facilities during our many stays on the island. Russell Hall reviewed the manuscript and we appreciate suggestions for improvement.

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Herpetological Review, 2004, 35(3), 235–238.

Putting the Squeeze on Venomous Snakes: Accuracy and Precision of Length Measurements Taken with the "Squeeze Box"

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Snout–vent length (SVL) is the most commonly reported measure of linear body size for snakes (Fitch 1987). SVL is correlated with litter or clutch size in many species (Gregory and Larsen 1996; Seigel and Ford 1987) and is an indicator of sexual maturity (Macartney and Gregory 1988) and sometimes of age class (Waye 1999). Measurement of SVL in the field can be difficult for venomous snakes for several reasons: (1) investigator safety must be maintained while restraining the snake, (2) stress and injury to the snake must be minimized, and (3) measurements need to be completed in a timely fashion. Various methods have been developed that deal with each of these constraints to different degrees including physical handling or pinning and stretching (Fitch 1987), anesthesia (Fitch 1987; Hardy and Greene 1999), nooses (Gregory et al. 1989; King and Duvall 1984), and squeeze boxes (Cross 2000; Quinn and Jones 1974).

Quinn and Jones (1974) introduced the 'squeeze box' to estimate the SVL of venomous snakes, which effectively addresses the three concerns stated above. Tongs are used to place the snake into a clear plastic box and a piece of foam or rubber and a clear plastic lid are placed over the snake to hold it in place. The box is inverted and the SVL of the snake is traced for measurement with a map reader, flexible measuring tape, or other device. The procedure is time efficient and relatively safe, and investigators with little experience in handling venomous snakes (e.g., seasonal assistants) show little difficulty in learning its use quickly. The squeeze box thus appears to be an efficient tool to obtain SVLs of snakes, venomous or otherwise.

Despite the apparent merits and widespread use of the squeeze-box method to measure SVLs of snakes, to our knowledge no rigorous analysis of its accuracy and precision has been conducted. Fifty repeated measurements of a single juvenile False Water Cobra (*Hydrodynastes gigas*) showed little variation (Quinn and Jones 1974), but these measurements were not compared to the SVL of the snake measured using alternative methods such as pinning and stretching. Measurements of 10 Cottonmouths (*Agkistrodon piscivorus*) using a modified squeeze box suitable for field studies were compared to the same animals when anaesthetized, and differed by <1 cm (Cross 2000). Here we use data collected during a study of the Northern Pacific Rattlesnake (*Crotalus oreganus*) to address two questions in the use of squeeze boxes: (1) How accurate are length measurements obtained from squeeze boxes compared to those obtained from stretched lengths? (2) How precise are repeated tracings and measurements using squeeze boxes?

We measured SVLs of *C. oreganus* collected during an inventory project (1999–2001) in the vicinity of Kamloops, British Columbia, Canada. Additional data came from animals encountered during the monitoring of a hibernaculum near Vernon, British Columbia (P. T. Gregory and K. W. Larsen, unpubl. data). To measure its snout-vent length, each snake was pinned and grasped behind the head, then stretched along a meter stick. For consistency, one of us (KWL) collected all measurements of SVL. Hereafter we refer to these data as 'standard measurements' of SVL.

After the standard measurement of SVL was obtained for each snake, the animal was transferred to a squeeze box. Our squeeze box consisted of an open-top clear plastic box measuring 42 x 26 x 13 cm. Inside the box were two 5-cm thick pieces of commercial upholstery foam, on top of which fitted a clear lid equipped with a handle that could be grasped by commercial snake tongs. Three tracings were taken for each snake (described below); between tracings the snake was placed on the ground and then picked up and replaced in the squeeze box. We manipulated the position of the animal in the squeeze box only if it was coiled such that a section of its body overlapped another section, e.g., if part of the snake was in a 'figure-eight' position, we would gently prod it until it assumed another position. After the three tracings were made, we took three measurements of each tracing, using a pocket On Tour Map Measurer™ (Pico Design, Toronto, Ontario).

In total, nine measurements (three tracings, three measurements each) were recorded for each of 21 snakes ranging in SVL from 347–845 mm. We had an additional seven animals for which we had one tracing (with three measurements each). We included these animals in some of our analyses (see below), because they improved the range of sizes of measured snakes. Standard SVL was measured only once for all 28 animals. We determined the sex of each snake by probing for hemipenal pouches or via hemipene extrusion (Blanchard and Finster 1933; Gregory 1983).

TABLE 1. Comparison of different methods used to estimate the snout–vent length (SVL) of rattlesnakes (to nearest mm). Sample size is 21 for all measures. Estimating SVL using a standard approach (stretching animal along a meter stick) provided SVL estimates that were significantly higher than those produced using a squeeze box. Three of the squeeze-box methods involved multiple measurements on the same snake being used to generate a mean value that was used as an estimate of snake SVL. The standard deviation of these multiple measurements also was calculated for each snake, and the average of these standard deviation values is provided.

Method Used to Estimate SVL	Estimated SVL (± SE)	Comparison Standard SVL (paired t-test)	Mean SD
Standard SVL (meter-stick stretch)	696 (± 40.4)	—	—
One tracing, one measurement	603 (± 35.7)	$P < 0.001$	—
One tracing, three measurements	602 (± 35.9)	$P < 0.001$	3.9
Three tracings, one measurement each	597 (± 36.2)	$P < 0.001$	14.4
Three tracings, three measurements each	596 (± 36.3)	$P < 0.001$	13.6

We used four methods to calculate squeeze-box estimates of SVL, which collectively represent the varying degrees of time and effort that might be invested by an investigator measuring SVL in the field: (1) one tracing, one measurement: one measurement obtained from the single tracing; (2) one tracing, three measurements: mean of three measurements from the single tracing; (3) three tracings, one measurement each: one measurement taken on each of three tracings; (4) three tracings, three measurements each: mean of three tracings used to calculate mean of the three measurements. The grand mean of these three means was used as the datum in analyses.

We compared the estimates of snake SVLs obtained from the four squeeze-box methods to those obtained from the standard method using paired t-tests. Because the latter three methods each produced three initial estimates of SVL per snake (prior to calculation of the grand mean), we were able to calculate the standard deviation (SD) of these measures for each snake. We used the mean SD obtained under each of these squeeze-box methods to assess the variation caused by the inclusion of multiple tracings of the same animal. With only one measurement of standard SVL per snake we could not estimate the variance associated with multiple measurements, however a recent study using a similar method of determining SVL (flexible tape run along the venter of suspended, stretched snakes) found repeated SVL measurements (10 measurements on 20 snakes) to be quite precise as indicated by small deviations of the individual measurements from the mean (mean coefficient of variation = 1.10 ± 0.10%) (Blouin-Demers 2003). Repeated standard SVL measurements in garter snakes also have low variance, even between researchers, once a reasonable amount of experience with the technique has been established (Larsen, unpubl. data).

Estimates of snake SVL provided by the standard method were significantly higher than those provided by any of the four squeeze-box methods (in all cases, $P < 0.001$; Table 1). The average SVL estimates provided by the four squeeze-box methods were similar (± 1.2%), however, variation in the estimates of SVL for each snake expectedly increased as multiple tracings and/or measurements were added (Table 1). Accuracy in estimating SVL (using standard estimates for reference) was not improved by providing additional tracings and/or measurements. A simple regression shows the relationship between measurements of SVL obtained using standard and squeeze-box methods (Fig. 1).

To further illustrate the use of squeeze-box estimates of SVL, we compared a size-frequency histogram developed using squeeze-box data from the one tracing, three measurements calculation (Fig. 2A), with a second histogram constructed using data from the standard method (Fig. 2B). Both frequency distributions have three peaks, however a Kolmogorov-Smirnov two-sample test (Sokal and Rohlf 1969) indicated that the differences in the two distributions were marginally significant ($D_{28,28} = 0.36$; $P = 0.055$).

Our study indicates that using the squeeze box resulted in significantly lower SVL estimates than those obtained using the standard approach irrespective of tracings or measurements. However, because the correlation between squeeze-box estimates

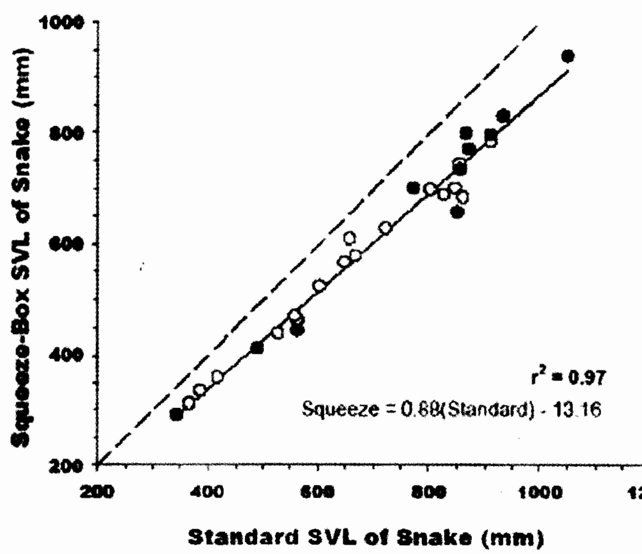

FIG. 1. Relationship between estimates of Northern Pacific Rattlesnake (*Crotalus oreganus*) snout–vent length (SVL) obtained using a squeeze box and values obtained by stretching the snake along a meter stick (standard method; N = 28). Estimates of snake SVL taken using the squeeze box consistently underestimated SVL. Squeeze-box estimates were the average of three measurements from the same tracing of one snake. Data from female snakes are represented by white circles, males by black circles. The dashed line represents a 1:1 relationship.

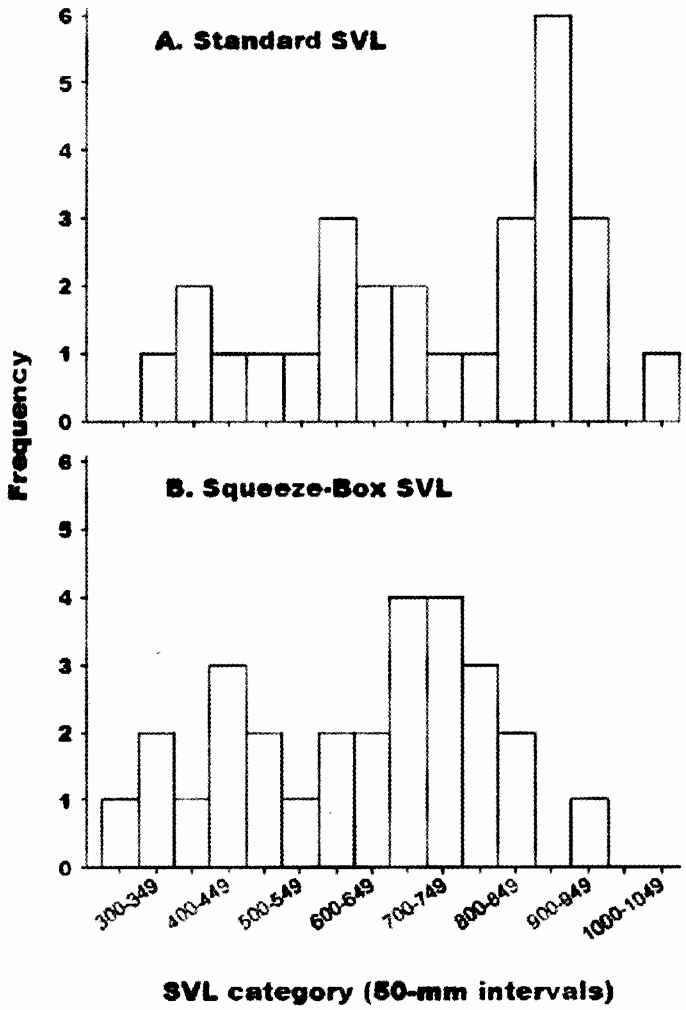

FIG. 2. Differences in size-frequency histograms of Northern Pacific Rattlesnakes (*Crotalus oreganus*) generated from the same animals using A. the standard snout-vent-length (SVL) determined by stretching a snake along a meter stick, and B. using a squeeze box. Each SVL is the mean of three measurements from the same tracing of the same animal.

and standard measurements was strong ($r^2 = 0.97$), it is possible to accurately predict standard SVL from squeeze-box SVLs. Also, because the length estimates obtained from the squeeze box were reasonably precise (i.e., high repeatability), repeated measures of snakes in the squeeze box are unnecessary. The one tracing, three measurements method described here appears to represent a good compromise between maximizing accuracy and precision and minimizing measurement time. Taking three measurements on the same tracing should preclude gross errors in SVL estimation without adding to the stress of the animal. Collecting additional tracings and measurements would increase processing time without improving the accuracy or precision of the estimates.

Different methods exist for measuring the length of snake tracings and may influence accuracy. Quinn and Jones (1974) placed a string along the snake tracing and then measured the string against a meter stick. Cross (2000) used a flexible metric tape for measuring the length of the snake tracing. Regardless of the tool used to record the length of the snake tracing, some error is to be expected. For example, use of a metric tape to measure tracings from tightly coiled animals can be problematic (C. L. Cross, pers. comm.). For these reasons, repeated measurements of each tracing are desirable.

The size of the squeeze box in relation to the snakes being measured also may be an important factor in determining the accuracy of SVL estimates. Squeeze boxes that are large relative to the snakes being studied may be more desirable. In this study there was a trend, albeit weak, of squeeze-box estimates for larger snakes to be less accurate (Fig. 1). This suggests that a larger box might increase the accuracy of measurements, presumably because the snakes would be less contracted during the tracings. The ratio of squeeze-box floor space (cm^2) to maximum snake SVL (mm) was approximately 1:1 in this study. Cross (2000) reported a high degree of accuracy for his squeeze-box estimates of SVL for cottonmouths measured in a squeeze box with a ratio of 1:3.4 (Cross 1998). Relatively large squeeze boxes may be more desirable, but logistics will dictate what size box can be transported in the field. For example, our squeeze box needed to fit into a backpack as it often was carried over long distances.

Given the potential usefulness of squeeze boxes, additional work on the accuracy and precision of this tool is warranted, preferably spanning a variety of taxa. Even the 'standard' method that we used in this study could be examined in more detail: we used data from this method as the benchmark for assessing the validity of length estimates based on squeeze-box measurements, but perhaps an even more effective and less stressful method exists. In general, investigators will need to determine the level of accuracy and precision in SVL measurements required to reach the goals of their respective studies. Detailed studies on life-history trade-offs experienced by snakes may require very accurate SVL data. Conversely, monitoring programs may not need to 'correct' SVL estimates from squeeze boxes, providing the methodology remains constant over time and among researchers.

Acknowledgments.—This study was conducted during the course of a rattlesnake inventory project, funded by the Habitat Conservation Trust Fund of British Columbia. Additional support for the project was provided by Forest Renewal British Columbia, through Aspen Planers Ltd. Research permits were provided by British Columbia Parks, British Columbia Ministry of Environment, Lands and Parks, and the City of Kamloops. Data collection in the field was facilitated by S. Chaput, L. Gomez, P. Gregory, D. Haughland, V. Larsen, and C. Thevarge. C. Cross and A. Savitsky provided us with comparative data on the sizes of squeeze boxes and cottonmouths, and P. Gregory and S. Rollans provided useful comments on a draft of the manuscript. We are grateful for the input of A. Price and an anonymous reviewer.

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