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## MEASURING SNAKES ACROSS THE DECADES. ARE TUBE-RESTRAINT MEASUREMENTS COMPATIBLE WITH AN EARLIER METHOD?

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**Abstract.**—Over the years, a wide diversity of established techniques with varied utility for the measurement of snake snout-vent length (SVL) have developed dependent upon the needs of investigators, especially for venomous species. Tube-restraint has emerged as a standard for safe and ethical handling while remaining inexpensive and pragmatic for field studies. While the adoption of this technique addresses concerns for accuracy and the ethical treatment of animals, it presents challenges when drawing comparisons with historical data gathered using alternative techniques. In the process of conducting a population study on Western Rattlesnakes (*Crotalus oreganus*) in British Columbia, Canada, we sought to determine whether length data gathered with the tube-restraint technique and the more historically prominent noose-restraint poles could be directly compared. We conducted paired measurements (n = 74) on rattlesnakes using both the noose-restraint and tube-restraint techniques and observed only marginal differences between methods. Following previous authors, we advocate that tube-restraint replaces other less-benign measurement techniques while providing comparable data. Further research is warranted, however, to investigate the accuracy of tube-restraint when compared with other historical methods, particularly a comparative evaluation of the technique amongst snake taxa with varying body forms.

**Key Words.**—historical; measurement; morphology; rattlesnake; snake; snout-vent length; SVL; tube

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### INTRODUCTION

A diversity of measurement techniques have been used through time to collect data on snake body length (snout-vent length or SVL; see Greenbaum 2003 and Tsai et al. 2018 and references within), offering flexibility to investigators, particularly those working on venomous animals. Tube-restraint has emerged as a favorable candidate for a universal length measurement technique for snakes as it is widely acknowledged as a consistently safe standard for the handling of venomous species (Murphy 1971; Murphy and Armstrong 1978; Lock 2008; Johnson 2011; Hogan 2015). This method requires readily available, inexpensive equipment, is practical for field studies, and drastically reduces the chance of injury to snakes and handlers. While tube-restraint for non-venomous snakes is not necessary to ensure handler safety, it offers protection to animals against potentially harmful manipulation of the sensitive head and cervical vertebrae, ensures measurement consistency across taxa, and is a practical handling method for veterinary services and taking caudal blood samples for genetic analyses. Tube-restraint requires several tubes of varying diameter; however, these are inexpensive and easily transportable.

While the historical shift in techniques reflects

an increasing concern for accuracy and the ethical handling of animals, it creates the problem of comparing contemporary data with those taken in the past using a different technique. To ensure such comparisons are robust requires a statistical assessment of lengths using different measurement methods. There are several studies that have investigated the accuracy and precision of various body length measurement methods (Madsen and Shine 2001; Blouin-Demers 2003; Bertram and Larsen 2004; Setser 2007; Cundall et al. 2016), but with more attention put towards accuracy within the method, rather than making comparisons between methods.

In the process of studying a population of Western Rattlesnakes (*Crotalus oreganus*), we needed to compare contemporary data on snake lengths to those collected nearly 35 y prior at the same site. We used the tube-restraint method to measure snake lengths, while historical data were collected using noose-restraint poles (see Schmidt and Davis 1941; Conant 1958; Bellairs 1967; Fowler 1978; but specifically Gregory et al. 1989), hereafter referred to as the noose-stretch method. To our knowledge a specific comparison of tube-restraint and noose-stretch methods does not exist. Understanding the relationship between these two techniques would allow historical morphological data to be compared with measurements from current populations.

## MATERIALS AND METHODS

We measured SVLs of Western Rattlesnakes in a population located in Vernon, British Columbia, Canada, in the fall of 2018. We first measured each snake using the tube-restraint method where, following Murphy (1971), we coaxed the snake to enter a clear acrylic tube until approximately one-third of the anterior end of the snake was inside. We carefully selected the appropriate tube size for the individual to ensure it could not turn its head around within the tube and to prevent contortion during handling. Once the snake was restrained, a handler guided the head of the snake into the distal end of the tube and secured the anterior section of the snake within the tube while another handler measured its length using a flexible measuring tape to trace along the dorsal surface along the vertebral ridge, starting from the snout and ending at the opening of the cloacal vent. We considered tube measurements precise when at least two recorded measurements were within 5 mm; thus, final tube measurements represent the mean of at least two tracings (see Blouin-Demers 2003).

After allowing each snake a 5-min rest period within holding baskets, we measured SVL on the same animal using the noose-stretch method. We approximated the methodology used in earlier studies on the same population of snakes (Macartney 1985, 1989; Macartney and Gregory 1988; Macartney et al. 1988, 1990) by using a noose-restraint pole following Gregory (1989). We placed the head of each snake in the noose, then slowly and carefully extended it along a meter stick to obtain a SVL measurement. We only conducted noose-stretch measurements once per individual to mitigate stress and injury potential. To avoid user bias and unnecessary additional measurements, the same investigator made all measurements.

To ensure unbiased comparisons, we ideally would have measured individual snakes using both techniques multiple times, with consecutive measurements being recorded by different investigators blind to prior measurements. Unfortunately, we were not able to hold our free-ranging study animals in captivity for extended periods (ethical considerations and permitting restrictions for species-at-risk), nor could we reliably recapture individual snakes for re-measurement except during sequential periods of den egress, between which times snake growth would have occurred. We thus could not completely eliminate the possibility that subconscious bias by the investigators would affect the repeatability of the two methods. Similar approaches have been taken to compare different measurement techniques, however (Madsen and Shine 2001; Measey et al. 2003; Bertram and Larsen 2004). Using a similar measurement technique, Rivas et al. (2008) suggest that measurements gathered independently by

two experienced researchers are generally consistent. Finally, two field researchers worked side-by-side during the two types of measurement on each snake, acting as a double check on the length value being recorded.

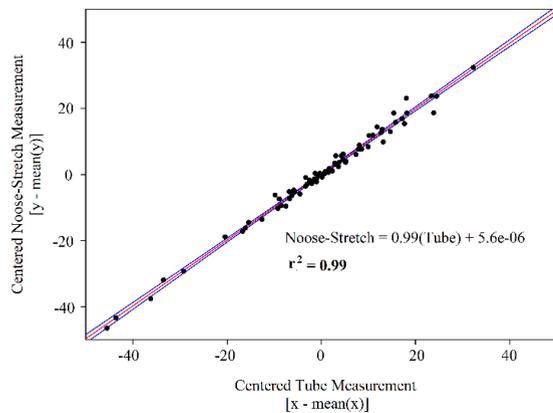
We used R 3.6.1 (R Development Core Team 2019) for all statistical analyses. We compared tube-restraint and noose-stretch methods using several statistical tests. First, we used a paired *t*-test to estimate the mean difference in SVL between measurements of both methods on the same snake. Second, we used Linear Regression to assess the relationship between noose-stretch and tube-restraint measurements. We used the tube restraint measurement as the predictor variable and the noose-stretch measurement as the response. Both measurements were centered by subtracting the mean of each measurement method from measurements of individual snakes; this allowed us to estimate the difference between measurements for a snake of average size as the *y*-intercept. Lastly, we grouped measurements into ecologically relevant size classes of juvenile (250–550 mm SVL), subadult/adult (550–750 mm SVL), adult (700–800 mm SVL), and large adult (800–1,050 mm SVL) and we used a single factor ANOVA to assess measurement discrepancy between size classes. All data met parametric assumptions for testing, and for all tests,  $\alpha = 0.05$ .

## RESULTS

We obtained paired measurements of SVL for 74 unique individuals. The mean measurement difference between methods was slight ( $3.2 \text{ mm} \pm 1.5 \text{ mm}$  standard deviation, or 0.4% of mean body length in the tube sample). Paired measurements were not significantly different ( $t = -1.84$ ,  $df = 73$ ,  $P = 0.071$ ; mean difference =  $-0.33$ ; 95% confidence interval [CI] =  $-0.68, 0.028$ ), with noose-stretch measurements generally being larger than tube-restraint. There was a strong relationship between measurement methods ( $r^2 = 0.99$ ,  $F_{1,73} = 6,502.3$ ,  $P < 0.001$ ; Fig. 1) and there was no difference at the origin (95% CI of *y*-intercept,  $-0.36 \leq \beta_0 \leq 0.36$ ) and no change in measurement difference between methods with changes in snake size (95% CI of slope,  $0.97 \leq \beta_1 \leq 1.02$ ). Measurement discrepancy was not significantly different between size classes based on residual values from regression analysis ( $F_{3,69} = 2.33$ ,  $P = 0.082$ ). The most severe measurement discrepancies (top 5%;  $n = 4$ ) were animals with SVLs of 927, 921, 885, and 635 mm (Fig. 1).

## DISCUSSION

Our study indicates that tube-restraint SVL measurements were consistent with those obtained using the noose-stretch method. All differences



**FIGURE 1.** Relationship of zero-centered measurements of snout-vent length (SVL) of Western Rattlesnakes (*Crotalus oreganus*) obtained via tube-restraint and noose-stretch methods ( $n = 74$ ) in British Columbia, Canada. The red line represents the mean slope, and blue lines represent 95% confidence limits.

in measurements were relatively small, although measurement difference tended to be greater for longer snakes, suggesting additional care should be taken when measuring particularly long animals. We attribute these greater discrepancies to variability in snake flexibility and cooperation during stretch measurements (Madsen and Shine 2001; Foster 2012; Astley et al. 2017) and measurement error during tube-restraint measurements for particularly long snakes. Our results did suggest that the measurement differences between methods were almost significant; however, we believe that for the purposes of comparing data collected using the different methodologies (i.e., to determine changes in population structure) this relationship is satisfactory. Furthermore, the differences in size obtained by different measurement methods are likely miniscule relative to ecologically relevant differences in size structure among populations or over time.

There has long been a call for a universal model of snake length measurement (Seigel and Ford 1988). We support this call for standardization and advocate for the adoption of tube-restraint as a universal standard for snake body length measurements. When appropriate, comparisons of data collected using this method versus those used historically should continue to be evaluated, particularly for species with varying body forms (i.e., Viperidae versus Colubridae, shorter snakes versus relatively longer ones, etc.).

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