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12-2013

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Mueller, Julias S.; Cheek, Brandon D.; Chen, Qingman; Groeschel, Jillian; Brewer, Shannok K.; and Grabowski, Timothy B., "A Simple Device for Measuring the Minimum Current Velocity to Maintain Semi-Buoyant Fish Eggs in Suspension" (2013). *The Prairie Naturalist*. 54.  
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## A Simple Device for Measuring the Minimum Current Velocity to Maintain Semi-Buoyant Fish Eggs in Suspension

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**ABSTRACT** Pelagic broadcast spawning cyprinids are common to Great Plains rivers and streams. This reproductive guild produces non-adhesive semi-buoyant eggs that require sufficient current velocity to remain in suspension during development. Although studies have shown that there may be a minimum velocity needed to keep the eggs in suspension, this velocity has not been estimated directly nor has the influence of physicochemical factors on egg buoyancy been determined. We developed a simple, inexpensive flow chamber that allowed for evaluation of minimum current velocity needed to keep semi-buoyant eggs in suspension at any time frame during egg development. The device described here has the capability of testing the minimum current velocity needed to keep semi-buoyant eggs in suspension at a wide range of physicochemical conditions. We used gellan beads soaked in freshwater for 0, 24, and 48 hrs as egg surrogates and evaluated minimum current velocities necessary to keep them in suspension at different combinations of temperature ( $20.0 \pm 1.0^\circ\text{C}$ ,  $25.0 \pm 1.0^\circ\text{C}$ , and  $28.0 \pm 1.0^\circ\text{C}$ ) and total dissolved solids (TDS;  $1,000\text{ mg L}^{-1}$ ,  $3,000\text{ mg L}^{-1}$ , and  $6,000\text{ mg L}^{-1}$ ). We found that our methodology generated consistent, repeatable results within treatment groups. Current velocities ranging from 0.001–0.026 needed to keep the gellan beads in suspension were negatively correlated to soak times and TDS and positively correlated with temperature. The flow chamber is a viable approach for evaluating minimum current velocities needed to keep the eggs of pelagic broadcast spawning cyprinids in suspension during development.

**KEY WORDS** minimum current velocity, pelagic broadcast-spawning cyprinids, semi-buoyant eggs, velocity chamber

At least 12 species of Great Plains cyprinids produce non-adhesive semi-buoyant eggs that are broadcast into the water column during spawning (Perkin and Gido 2011). This reproductive strategy presumably allows eggs to complete development while avoiding the risk of abrasive damage from shifting sandy substrates characteristic of Great Plains rivers and streams (Moore 1944). However, these pelagic broadcast-spawning cyprinids are in decline throughout the Great Plains and most are of conservation concern (Hoagstrom et al. 2011, Perkin and Gido 2011). Anthropogenic modifications of the landscape, fragmentation of rivers through dam and reservoir construction, and altered flow regimes have all impacted pelagic broadcast-spawning cyprinids (Dudley and Platania 2007, Durham and Wilde 2009, Hoagstrom et al. 2011, Perkin and Gido 2011).

In response to declining populations of pelagic broadcast-spawning cyprinids, current research has focused on the minimum distance eggs require for drifting in order to complete development (Perkin and Gido 2011). However, to date investigations have examined transport dynamics at coarse scales through analysis of patterns of species presence and absence or recovery of egg surrogates released during the spawning season of these species. For example, Perkin and Gido (2011) evaluated the minimum distance required for egg transport of eight species of pelagic broadcast-spawning Great Plains cyprinids by determining the minimum stream fragment

length in which populations of these species have persisted through time. At a finer scale, studies on Rio Grande silvery minnow (*Hybognathus amarus*) and Pecos bluntnose shiner (*Notropis simus pecosensis*) suggest that flow variability due to channel morphology and complexity may entrain eggs and shorten the fragment length or reduce water requirements for persistence (Kehmeier et al. 2007, Kinzli and Thornton 2009, Widmer et al. 2012). There has been little attention directly given to evaluating transport dynamics that might influence the distance required for successful reproduction (e.g., minimum current velocity required to keep eggs in suspension) and physicochemical conditions (e.g., temperature, total dissolved solids [TDS]) that might influence buoyancy of eggs.

Recent development of high-resolution models for evaluating drift trajectories of particles like semi-buoyant fish eggs has encouraged the evaluation of velocity and discharge requirements for successful reproduction. In this study, we describe a simple chamber and method for assessing current velocities necessary to keep semi-buoyant fish eggs in suspension and provide the initial validation of this approach using gellan beads as egg surrogates (Reinert et al. 2004, Kehmeier 2007). The objectives of our study are to describe the creation and operation for a flow chamber that can be used to keep semi-buoyant eggs in suspension, while also providing results that show how this device is capable of obtaining repeatable and accurate results under different physicochemical conditions.

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## METHODS

### Design and Construction

We designed a flow chamber consisting of four main components: a submersible pump, a riser stem, a ball valve, and a clear PVC observation and measurement chamber (Fig. 1). We used a Laguna Powerjet fountain and waterfall pump kit (United Pet Groups Inc., Cincinnati, Ohio, USA) rated at  $56.8 \text{ L min}^{-1}$  at 1.7 m of head pressure. The riser stem, fitting, and control valve included with the pump were connected with a standard 2.09-cm diameter PVC ball valve. The use of two valves allowed for both fine adjustments and maintenance of very low current velocities. Our lowest obtained velocity was  $0.001 \text{ m s}^{-1}$ , but the flow chamber has the capabilities of lower velocities if needed. We cut a 2.09-cm diameter clear PVC pipe to a length of 120 cm and attached to the ball valve, forming the flow chamber column. We drew two circles around the PVC pipe to delineate a length of 100 cm centered 10 cm from the top and bottom of the PVC pipe. We drew smaller hash marks every 1.0 cm on the pipe within this 100-cm length. We used these hash marks to calculate current velocity by calculating the ratio between the time in seconds (s) for the tube to fill to that point and the linear distance (cm) from the bottom of the chamber to the hash mark. Additionally, we used hash marks to estimate discharge. We estimated discharge by multiplying the area of the tube by the average velocity within the tube. The flow chamber weighed approxi-

mately 3.2 kg and could be quickly broken down into two pieces, making it portable and lightweight.

### Operation

Prior to placing objects into the column, we closed the ball valve to avoid a sudden jet of water when the device is turned on. We then placed the flow chamber into a 37.8-L plastic tub. Tubs measured approximately 35 cm in depth, 47 cm in width, and 47 cm in height, which was sufficient to keep the pump fully submerged and was wide enough to catch water as it spilled over the top of the column. We verified the vertical position of the column using a level or plumb bob to ensure accurate and consistent measurements in all trials.

We started the pump and opened the ball valve approximately 3–7% of a full turn to allow the column to slowly begin filling. We added gellan beads as the water reached the 50-cm mark on the pipe then allowed the column to continue filling while adjusting the ball valve to keep the objects at equilibrium ( $\pm 5 \text{ cm}$ ) within the column. When water spilled over the top of the column, we started a 1-min timer and observed the gellan beads to ensure they remained stationary in the column without any adjustments to the ball valve. If the gellan beads remained stationary, then we recorded the vertical position of each bead. If the objects had moved within the column, then we adjusted the ball valve and repeated the process until the objects were able to remain at equilibrium

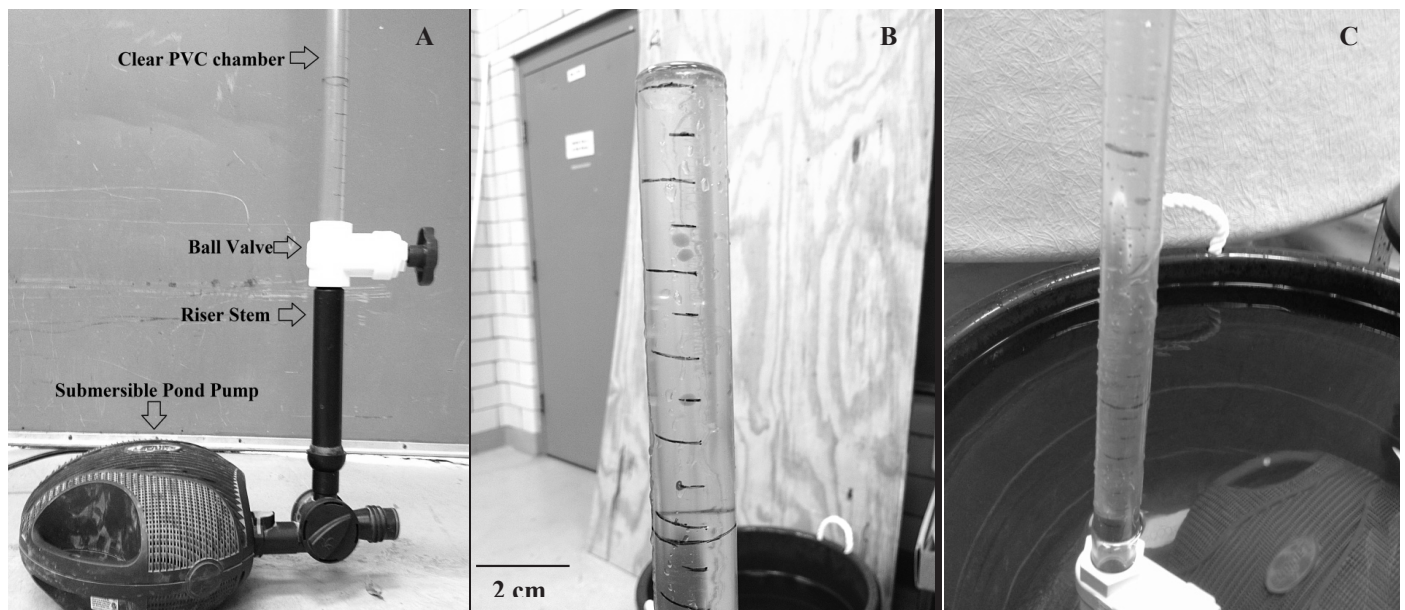


Figure 1. A) The flow chamber, created using a submersible pond pump, riser stem, ball valve, and clear PVC chamber, was used to measure the minimum current velocity needed to keep gellan beads in suspension. B) The clear PVC chamber with circles drawn around the pipe to represent 10 cm and hash marks drawn to represent 1 cm. C) The flow chamber fully submerged in a plastic tub regulated at a given temperature and total dissolved solid treatment level. Trials were conducted during October 2012 in laboratory facilities at Texas Tech University, Lubbock, USA.

during the 1-min test. Once this step was complete, we unplugged the pump and no further adjustments were made to the valves for the remainder of the trial. If there was a need to recover the test objects, we removed the pipe, valve, and riser stem from the pump and turned them upside down over a dip net. After the water was drained out of the pipe, we repeated the previously described filling procedure and recorded the time it took for the chamber to fill to each recorded position.

### Experimental Design

We used red gellan beads (Technology Flavors and Fragrances Inc., Amityville, New York, USA) to test the influence of water temperature and TDS on the minimum velocities required to keep egg surrogates in suspension. Originally used in food and drinks, studies have shown these beads have a similar shape and specific gravity (SG) as some fish eggs and do not change physically with the addition of salinity (Reinert et al. 2004, Kehmeier et al. 2007). However, gellan beads are packaged in a preservative solution that can affect their SG. Reinert et al. (2004) soaked gellan beads in water for 0, 24, and 48 hrs and found that SG was negatively correlated with soak time, presumably due to the absorption of water. Therefore, the gellan beads were soaked in water for the same three time frames (0, 24, 48 hrs) to minimize the influence of the preservative solution on the results of the trials.

For each soak time treatment group, we ran trials at three different water temperatures:  $20.0 \pm 1.0^\circ \text{C}$ ,  $25.0 \pm 1.0^\circ \text{C}$ , and  $28.0 \pm 1.0^\circ \text{C}$ . The temperature of the water was maintained by the addition of one or more 50-W submersible heaters (Aqueon Products, Franklin, Wisconsin, USA). For each soak time and temperature combination, there were also three different TDS treatment levels:  $1,000 \text{ mg L}^{-1}$ ,  $3,000 \text{ mg L}^{-1}$  and  $6,000 \text{ mg L}^{-1}$ . Total dissolved solid treatment levels were established through the addition of the appropriate amount of Instant Ocean seawater mix (Spectrum Brands, Cincinnati, Ohio, USA). We determined the treatment levels based upon values observed in the Canadian River during the spawning season of pelagic broadcast-spawning cyprinids native to the drainage (Pigg et al. 1999) and set up a separate tub for each temperature  $\times$  TDS treatment group. We placed three gellan beads from each soak time  $\times$  temperature  $\times$  TDS treatment group in the appropriate flow chamber and conducted three replicate trials per treatment group using the procedure described above. We performed power analyses (Zar 1999) to determine the appropriate number of trials per temperature, TDS, soak time treatment group. The majority of the beads remained in close proximity to one another (1–2 cm) within the column and in these cases we used a mean vertical position for our velocity calculations. There were a few instances (Table 1) where one of the three beads would become stationary at a distance from the others (3–5 cm). We conducted an additional trial in these cases to ensure this was a result of physical or SG differences that were found among gel-

lan beads (Reinert et al. 2004). Trials where the beads were widely separated within the chamber (6+ cm) were eliminated from analysis as visual examination indicated these beads were damaged or otherwise deformed.

We used analysis of covariance (ANCOVA) to evaluate the effects of soak time, temperature, and TDS on the minimum current velocity necessary to keep the gellan beads in suspension with soak time as a covariate and temperature and TDS as independent variables. We used Dunnett's multiple comparison procedure as a post-hoc test to evaluate differences among treatment groups. We performed all analyses using SAS 9.2 (SAS Institute, Inc., Cary, North Carolina, USA) with a significance value of  $\alpha = 0.05$ .

### RESULTS

Our methodology generated consistent, repeatable results within treatment groups (Table 1). In general, the coefficient of variation of the mean minimum current velocities for each treatment group did not correlate with soak time ( $r = 0.02$ ,  $P = 0.93$ ), temperature ( $r = 0.22$ ,  $P = 0.28$ ), or TDS ( $r = 0.30$ ,  $P = 0.13$ ). Our minimum detectable difference in mean minimum current velocity among treatment groups was approximately  $0.002 \text{ m s}^{-1}$  based upon a post-hoc power analysis (Zar 1999). As expected, soak time had an influence on the minimum current velocity necessary to keep gellan beads in suspension ( $F_{3,83} = 6.56$ ,  $P < 0.05$ ; Fig. 2). Treatments groups soaked in water prior to testing required lower current velocities to remain in suspension than those that received no pre-test soak ( $t_1 \leq -16.80$ ;  $P < 0.05$ ; Fig. 2). However, there was no difference between the 24-hr and 48-hr soak times ( $t_1 = 0.65$ ,  $P = 0.87$ ).

Overall, soak time, TDS, and the interaction between soak time and temperature explained approximately 83% of the variation in current velocity required to keep gellan beads in suspension. There was an inverse relationship between TDS and current velocity that was independent of soak time and temperature ( $F_{1,83} = 23.08$ ,  $P < 0.05$ ). However, temperature interacted with the three soak times differently, independent of TDS ( $F_{3,83} = 5.05$ ,  $P < 0.05$ ). The interaction of temperature and soak time exhibited a positive relationship to minimum current velocity ( $\beta_1 = 0.0004$ ,  $t_1 = 3.18$ ,  $P < 0.05$ ), while the interaction of the other two soak times with temperature did not produce an effect (24 hrs:  $\beta_1 = -0.004$ ,  $t_1 = -1.65$ ,  $P = 0.10$ ; 48 hrs:  $\beta_1 = -0.003$ ,  $t_1 = -1.85$ ,  $P = 0.07$ ).

### DISCUSSION

Our results demonstrate that the flow chamber described in this paper can be used to estimate minimum current velocities required to keep semi-buoyant eggs in suspension at any time during egg development and to provide the data necessary for modeling transport dynamics at fine spatial and temporal scales. The results of our study are consistent with

Table 1. Number of trials ( $n$ ) per soak time, temperature, and total dissolved solid (TDS) treatment group with the mean ( $\pm$  SD) and range of minimum current velocities necessary to keep gellan beads in suspension. Trials were conducted during October 2012 in laboratory facilities at Texas Tech University, Lubbock, USA.

Soak time (hrs)	Temperature ( $^{\circ}$ C)	TDS (mg L $^{-1}$ )	$n$	Mean current velocity (m s $^{-1}$ )
0	20	1,000	4	0.017 $\pm$ 0.006
		3,000	3	0.015 $\pm$ 0.005
		6,000	3	0.015 $\pm$ 0.005
	25	1,000	4	0.017 $\pm$ 0.006
		3,000	3	0.018 $\pm$ 0.001
		6,000	3	0.013 $\pm$ 0.006
	28	1,000	3	0.026 $\pm$ 0.003
		3,000	3	0.021 $\pm$ 0.009
		6,000	3	0.016 $\pm$ 0.007
24	20	1,000	3	0.006 $\pm$ 0.001
		3,000	3	0.005 $\pm$ 0.001
		6,000	3	0.002 $\pm$ 0.001
	25	1,000	5	0.005 $\pm$ 0.002
		3,000	3	0.002 $\pm$ 0.000
		6,000	3	0.001 $\pm$ 0.001
	28	1,000	3	0.003 $\pm$ 0.001
		3,000	3	0.001 $\pm$ 0.001
		6,000	3	0.001 $\pm$ 0.001
48	20	1,000	3	0.007 $\pm$ 0.001
		3,000	3	0.006 $\pm$ 0.001
		6,000	4	0.003 $\pm$ 0.001
	25	1,000	4	0.004 $\pm$ 0.003
		3,000	3	0.001 $\pm$ 0.000
		6,000	3	0.001 $\pm$ 0.001
	28	1,000	3	0.005 $\pm$ 0.003
		3,000	3	0.001 $\pm$ 0.000
		6,000	4	0.001 $\pm$ 0.000

gellan bead behavior as predicted by basic physics and work conducted by Reinert et al. (2004). An increase in TDS results in an increase in the density of the water medium, which would increase the buoyancy of the gellan beads and thus reduce minimum current velocity necessary to keep them in suspension. Additionally, soaking the beads in water reduced their specific gravity as they absorbed water and resulted in lower current velocities needed to keep them in suspension. However, it seemed that the density of water inside and outside the gellan beads were influenced similarly by temperature, resulting in relatively minor effect of temperature we observed on minimum current velocity.

Our device is capable of testing fish eggs under different physicochemical conditions such as, temperature and TDS; however, the chamber may over simplify stream conditions. For example, minimum current velocities obtained using this device may be relative as the chamber only measures horizontal rather than vertical movement. Furthermore, current velocity in a stream is not the only factor that is keeping these eggs in suspension. Turbulence created by interacting with channel features and ripples created by sand bed features may cause the velocity to be pushed in more of a quasi-vertical motion rather than simple horizontal as in a pipe with laminar flow. In essence, the vertical direction of the pipe for this

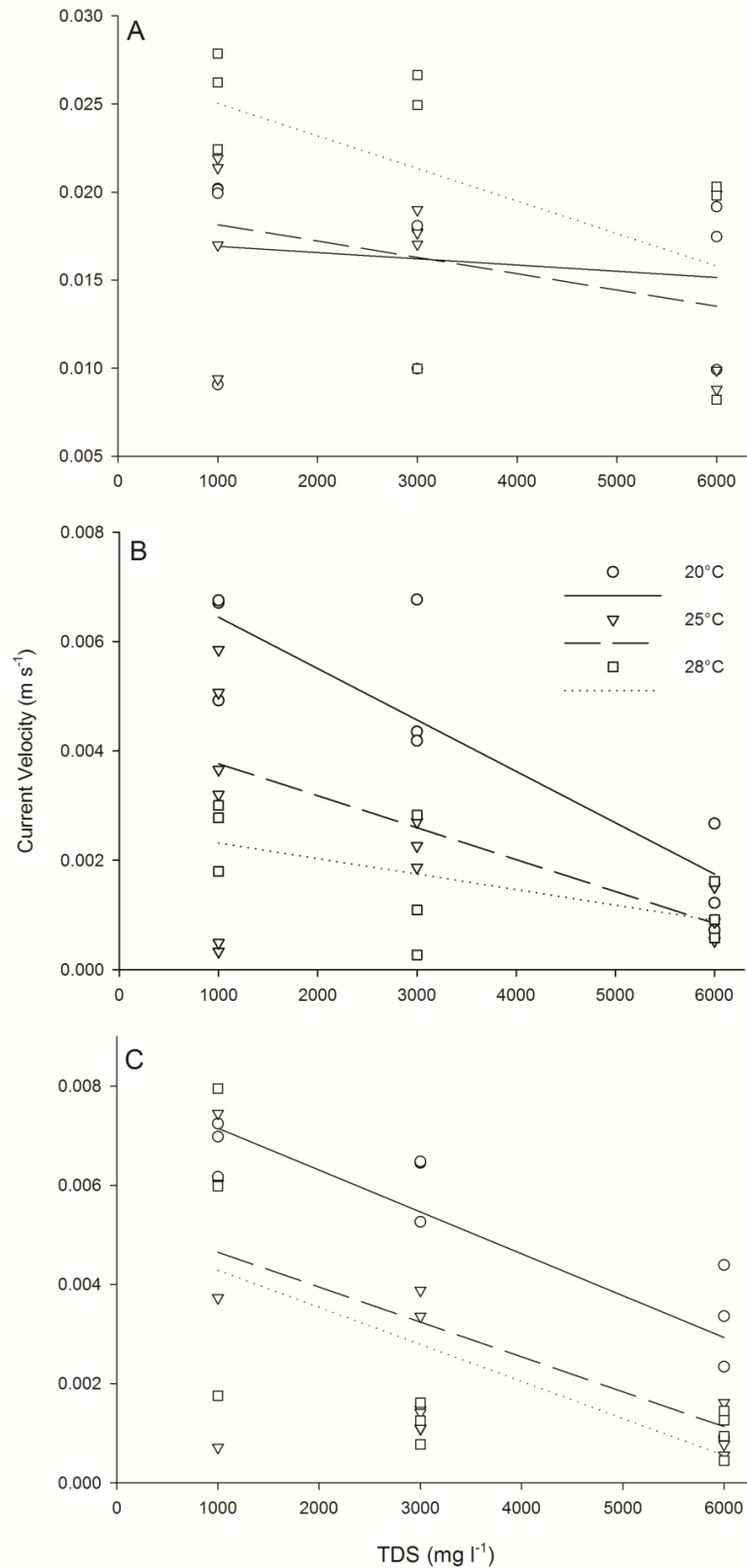


Figure 2. The effect of temperature and total dissolved solids (TDS) on the minimum current velocity required to keep gellan beads soaked 0-hrs (A), 24-hrs (B), and 48-hrs (C) in suspension. Trials were conducted during October 2012 in laboratory facilities at Texas Tech University, Lubbock, USA.

chamber calculates uplift velocity. Regardless, this device may be appropriate for questions related to magnitudes of differences when talking about stream velocity.

### MANAGEMENT IMPLICATIONS

Past studies have demonstrated the importance of discharge for pelagic broadcast-spawning fishes in freshwater systems (Reinert et al. 2004, Medley et al. 2007); however, there remains a lack of knowledge on the interactions between semi-buoyant eggs, the physicochemical environment, and the flow dynamics within a river channel. Researchers using this chamber can obtain both estimates of the minimum current velocity needed to keep semi-buoyant eggs in suspension and how physicochemical conditions influence this velocity. Understanding how changes in physicochemical conditions affect egg buoyancy may provide insight for determining the fragment length necessary for successful reproduction, especially as streams become increasingly fragmented due to the creation of dams and impoundments.

### ACKNOWLEDGMENTS

Funding was provided by the U.S. Fish and Wildlife Service Great Plains Landscape Conservation Cooperative. B. Durham and two anonymous reviewers provided comments and suggestions to improve this manuscript. Cooperating agencies for the Texas Cooperative Fish and Wildlife Research Unit are the U.S. Geological Survey, Texas Tech University, Texas Parks and Wildlife, and the Wildlife Management Institute. Cooperating agencies for the Oklahoma Cooperative Fish and Wildlife Research Unit are the U.S. Geological Survey, Oklahoma State University, Oklahoma Department of Wildlife Conservation and the Wildlife Management Institute. Use of trade, product, or firm names is for descriptive purposes only and does not imply endorsement by the U.S. Government.

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*Submitted 9 January 2013. Accepted 10 July 2013. Associate Editor was Melissa Wuellner.*