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## FACTORS INFLUENCING *IN-SITU* DETECTION OF PIT-TAGGED HELLBENDERS (*CRYPTOBRANCHUS ALLEGANIENSIS*) OCCUPYING ARTIFICIAL SHELTERS USING A SUBMERSIBLE ANTENNA

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**Abstract.**—Secretive species are difficult to study and often of conservation concern, as exemplified by the Eastern Hellbender (*Cryptobranchus alleganiensis*). Traditional methods for sampling Hellbenders involves moving rocks, which damages essential habitat. Use and installation of artificial shelters has made studying Hellbenders less dangerous for the animal and less disruptive to stream habitat; however, researchers using shelters generally capture occupying animals to identify them. We tested the ability of a submersible portable Passive Integrated Transponder (PIT) antenna to accurately detect PIT-tagged Hellbenders in shelters. We tested the effects of the presence and depth of cover rocks on top of shelters, PIT tag location within the shelter, and tag orientation on detection efficiency of Hellbenders. For the 32 shelters occupied by a tagged individual with cover rocks in place, the scanner accurately detected 31% of the animals versus 88% when cover rocks were removed. The detection efficiency of the scanner dropped below 50% once cover rock depth exceeded 11 cm. Tags placed near the interface of the entrance tunnel and chamber, or along the chamber walls, had higher detection efficiencies than those in other locations within the shelter. Vertically oriented tags were 18% more likely to be detected than horizontally oriented tags. Our study demonstrates that while this technology has certain limitations, it shows potential as a research tool for studying Hellbenders and other taxa without the need to frequently handle individuals.

**Key Words.**—amphibians; cryptic species; mark-recapture; non-invasive survey; occupancy; passive integrated transponder tag

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### INTRODUCTION

The ability to detect and identify unique individuals is an important aspect of long-term research and monitoring efforts. Many amphibian taxa make this task challenging because they are difficult to re-locate or permanently mark. Toe-clipping has been used to uniquely mark individuals but is traumatic and is ineffective for species that can rapidly regenerate digits (Ferner 1979; Davis and Ovaska 2001). Other techniques include the use of visible implant elastomers (Nauwelaerts et al. 2000), alphanumeric tags (Osborn et al. 2009), external or internal radio transmitters (Richards et al. 1994; Rowley and Alford 2007), gastric transmitters (Larson et al. 2013), pressurized fluorescent markings (Schlaepfer 1998), and harmonic direction finders (Rowley and Alford 2007; Borzée et al. 2018). Unfortunately, some of these technologies can be inconvenient to use over extended periods (i.e., years), due to markers fading over time (Schlaepfer 1998), tag migration or flipping post-implantation (Heard et al. 2008; Brannelly et al. 2013), and loss of transmission signal. Unlike the aforementioned marking techniques, Passive Integrated Transponder (PIT) tags provide several key advantages for permanently marking individuals (Gibbons and

Andrews 2004). They do not require battery power, have high retention rates across multiple taxa (> 95%; Brown 1997; Gries and Letcher 2002; Dare 2003; Unger et al. 2012), and can be implanted without inhibiting the normal functioning of the animal (Gibbons and Andrews 2004). However, this method typically requires re-capture of individuals to be scanned with handheld PIT tag readers.

We tested the feasibility of a novel method for monitoring and electronically recapturing PIT-tagged individuals that eliminates the need for physical handling. Hellbenders (*Cryptobranchus alleganiensis*) are an ideal study species to test this proposed methodology because they reside under large inaccessible boulders, making them especially difficult to capture and monitor. Hellbenders are fully aquatic salamanders and one of the largest amphibians in North America (up to 74 cm in total length; Fitch 1947). They are long-lived (25+ y), and inhabit cold, fast-flowing streams in Appalachia, southern portions of the Great Lakes states, western Kentucky, and the Ozark region of Missouri and Arkansas (Nickerson and Mays 1973; Taber et al. 1975). Traditional survey methods involve lifting rocks to capture and monitor Hellbenders (Nickerson and Krysko 2003; Browne et al. 2011). This method, however, is physically intensive, potentially dangerous for both surveyors and Hellbenders,

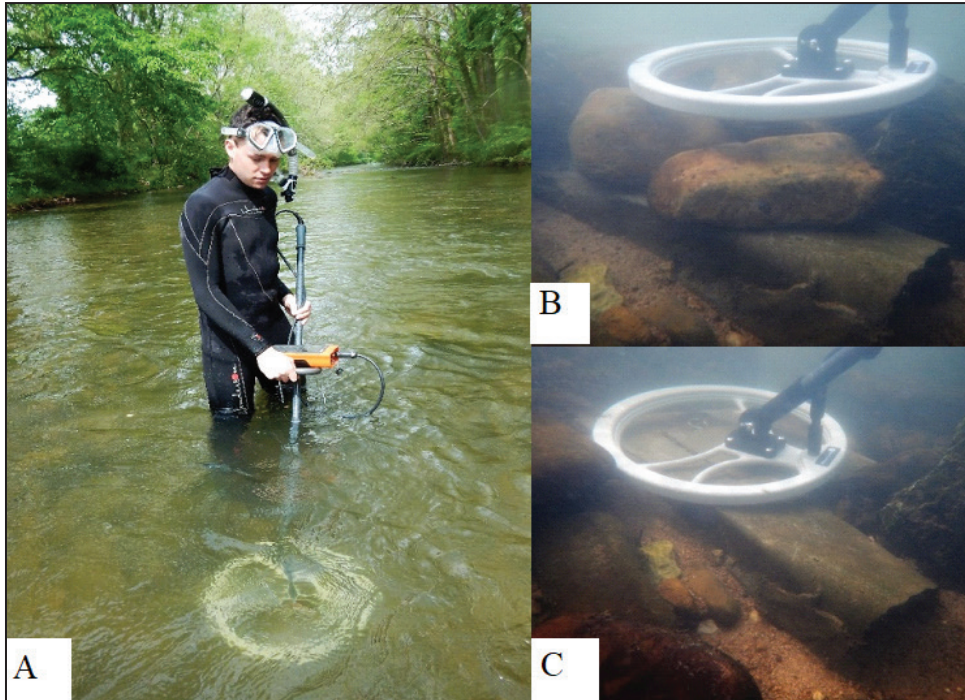


FIGURE 1. (A) Operational use of PIT tag scanning equipment to detect Hellbenders (*Cryptobranchus alleganiensis*). (B) Scan of artificial shelter with cover rocks in place. (C) Scan of artificial shelter with cover rocks removed. (Photographed by Alex Grimaudo).

and can destroy benthic stream habitat (Nickerson and Krysko 2003; Browne et al. 2011).

Due to the recent advent of artificial shelter technology, it is now possible to study and sample Hellbenders without displacing rocks (Briggler and Ackerson 2012; Jachowski 2016). These artificial shelters mimic the natural rock cavities used by Hellbenders and are occupied by Hellbenders for both shelter and nesting (Briggler and Ackerson 2012; Ettling et al. 2013; Jachowski 2016; Button 2019). Despite this promise, current manual techniques for sampling artificial shelters are still invasive, requiring disturbance of artificial shelter habitat (i.e., lid removal, blocking tunnel, and tactile investigation) to verify presence of Hellbenders. Additionally, researchers using traditional handheld PIT tag readers must remove Hellbenders from shelters to identify previously tagged individuals. Given the range-wide implementation of artificial Hellbender shelters (Briggler and Ackerson 2012; Ettling et al. 2013; Jachowski 2016; Settle 2017; Button 2019), our goal was to develop a technique for identifying PIT-tagged Hellbenders inside artificial shelters in a rapid and minimally disruptive manner.

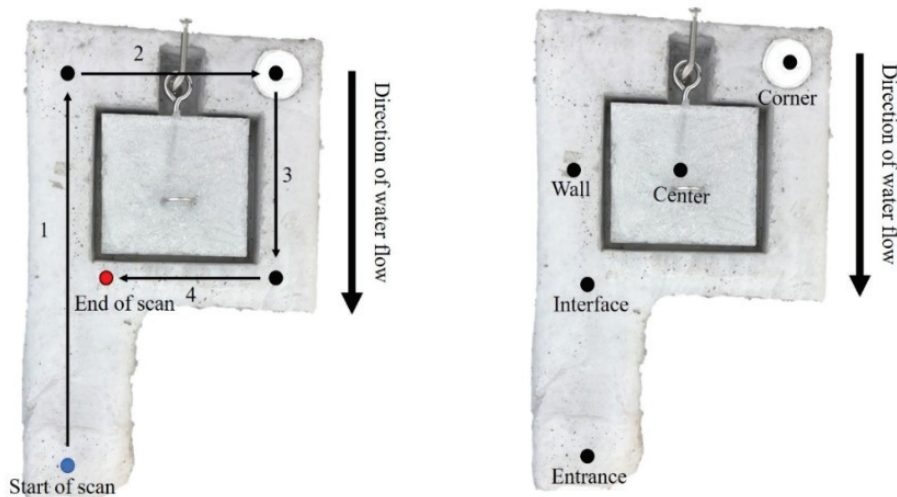
Recent advances in portable antenna systems and more powerful PIT-tag readers allow for remote detection of PIT-tagged animals beyond the range of traditional handheld readers. This technology has been used to successfully detect and identify tagged animals in both aquatic and terrestrial study systems (Zydlewski et al. 2006; Connette and Semlitsch 2013; Ousterhout and Semlitsch 2014) but remains untested as a method to study

Hellbenders in artificial shelters. In this study, we sought to determine the efficacy of using a submersible PIT tag antenna to remotely identify individual Hellbenders occupying artificial shelters, and to determine factors (e.g., tag implantation orientation, scanning distance, tag location within artificial shelters) that influence the performance of the antenna.

## MATERIALS AND METHODS

**Study sites.**—Our study area is located in southwestern Virginia, USA, in three streams within the upper Tennessee River Basin. We deployed 10 artificial shelter arrays within these three streams, each comprised of approximately 30 artificial shelters installed at an average density of one shelter per 160 m<sup>2</sup>. These arrays varied in length from 206–376 fluvial m.

**PIT-tag technology.**—Within our study area, every subadult and adult Hellbender ( $n = 906$ ) captured during previous surveys had a 12.5 mm PIT tag (HPT12, Biomark, Boise, Idaho, USA) inserted into the tail horizontally along the musculature. We used a Biomark HPR plus PIT-tag reader with a BP-plus handheld antenna (Biomark, Boise, Idaho, USA), referred to hereafter as the scanner, to test the feasibility of *in-situ* detection of tagged Hellbenders in artificial shelters (Fig. 1A). The HPR plus reader head is fully waterproof, capable of reading 134.2 kHz, HDX, FDX-B PIT tags, and has an antenna head width of 38 cm. When coupled with the



**FIGURE 2.** (A) Scanning procedure for each artificial shelter within streams: starting at the tunnel entrance (blue dot): (1) proceeded along the tunnel wall side of the shelter; (2) then moved along the back side of the shelter; (3) down the opposite side of the shelter; (4) ending with a scan across the front of the shelter (red dot); resulting in a concentric scan of the shelter with the scanner head on top of the shelter. All areas within and surrounding the arrows were scanned and the procedure was repeated as many times as possible within the 30 s time limit. (B) PIT tag locations within the artificial shelter tested in second experiment to assess the effects of location and PIT-tag orientation on tag detection efficiency.

BP-plus handheld antenna, the manufacturer states that the HPT12 tags are detectable at distances of 30.48–42.86 cm (Biomark, 2013. Biomark HPR Plus and HPR Reader. Available from <https://www.biomark.com> [Accessed 22 December 2017]). PIT tag read distances, however, can be affected by PIT tag orientation relative to the antenna face and other extrinsic factors, such as proximity to metal, power lines, and other sources of electromagnetic interference (Biomark, 2013. *op. cit.*).

**Artificial shelters.**—We constructed Hellbender artificial shelters from a mixture of sand, Portland cement, and Quikrete (The QUIKRETE Companies, Atlanta, Georgia, USA), which covered an interior metal frame made of galvanized hex-mesh and hardware cloth (Jachowski 2016). Each shelter consisted of a rectangular cavity chamber, accessible by a single tunnel entrance oriented facing downstream (modified boot design; Briggler and Ackerson 2012). Each artificial shelter weighed approximately 25 kg, with external chamber dimensions of 40 × 38 × 11 cm (length × width × height) and tunnel dimensions of 24 × 11 × 10 cm. A removable lid allowed researchers access to the main chamber to view and capture occupants (Fig. 2). Shelter installation methods were similar to Briggler and Ackerson (2012), with artificial shelters placed immediately downstream of large boulders (primary axis > 60 cm) that acted as anchor rocks during high stream flow events. We also surrounded the shelters with a mixture of stream sediment, gravel, small boulders, and placed large cover rocks on top of shelters to secure and camouflage them. We performed three experiments using these shelters between August and November 2017.

**Experiment 1: artificial shelter occupancy.**—To test the ability of the scanner to accurately detect Hellbender PIT tags and artificial shelter occupancy, we scanned 58 artificial shelters with the scanner before manually confirming shelter occupancy. Upon reaching an artificial shelter, we blocked the entrance of the shelter to prevent an occupant from escaping and performed the first scan of the shelter with cover rocks and installation rocks in place (with rocks; Fig. 1B), following a standardized and predetermined scanning-path procedure (Fig. 2A). We ended the first scan of each shelter upon detection of a PIT tag, or after 30 s of continuous scanning with no tag detection. If a tag was detected, we recorded the time to detection. We chose 30 s as the cutoff time for a complete scan because during an early pilot of this scanning procedure (June 2017; 22 unique shelters with PIT tagged occupants), all detections occurred within about 30 s. This short-duration scanning time also ensured that the scanning procedure could be conducted in approximately the same amount of time that it takes to open the shelter to ascertain presence/absence. After completing the scan with the cover rocks, we quantified the vertical depth of the cover rocks on top of the shelter using a mean of three representative points distributed across the surface of the shelter. We subsequently removed the cover rocks and performed a second scan (without rocks) with the scanner flush to the artificial shelter surface (Fig. 1C), following the same scanning and data collection procedure as the with rocks scan. Finally, we physically determined occupancy by removing the lid and reaching inside the shelter. We hand-captured any Hellbenders present and scanned them using a handheld pocket PIT-tag reader (Destron Fearing, Langeskov, Denmark). For data

analyses, we coded each with rocks and without rocks scanning event as a 1) true positive, 2) true negative, 3) false positive, or 4) false negative. True positive means the scanner detected a tag, the shelter was occupied, and the occupant ID matched the scanner read. True negative means the scanner did not detect a tag and the shelter was unoccupied or was occupied by an untagged individual. False positive means the scanner detected a tag, but the shelter was unoccupied, or the occupant tag did not match the scanner tag ID (meaning another animal was in close proximity to the shelter, but not occupying it, which the scanner had detected). False negative means the scanner did not detect a tag, but the shelter was occupied by a tagged individual. To align with the terminology of other studies employing similar PIT tag scanning technology (Zydlewski et al. 2006; Connolly et al. 2008), we use the term detection efficiency to refer to the proportion of true positives obtained for shelters occupied by a previously tagged individual.

**Experiment 2: PIT tag location and orientation.**—

To test whether PIT tag orientation or location within an artificial shelter influences scanner accuracy and success, we conducted a second experiment varying the PIT tag location (Fig. 2B) and orientation (horizontal or vertical) within unoccupied artificial shelters (n = 8). Shelters did not have cover rocks for this experiment. Orientation refers to the position of the ends of the PIT tag relative to the scanner head, horizontal (a parallel orientation; a common direction a tag would be oriented in a resting tail of a Hellbender relative to the scanner antenna), and vertical (a perpendicular orientation). We placed a loose PIT tag in a small plastic vial that was wedged into 3.175 mm thick clear vinyl tubing to secure the PIT tag within the artificial shelter during each manipulation trial (Fig. 2B). At each of the eight unoccupied artificial shelters, we performed three scans for each orientation-by-location combination (n = 30 scans per shelter; 240 total scans). We randomized the order of placements (location by orientation) for each shelter. The same person performed all scans, and never knew the location or orientation of the tags. Artificial shelter scan and data collection procedures were identical to those for the artificial shelter occupancy experiment (Fig. 2A).

**Experiment 3: scanner distance.**—We conducted a third experiment to assess the maximum detection distance of the scanner through rock material for horizontal and perpendicular tag orientations. We conducted this test at a local stone supplier (Old Dominion Flagstone Inc., Blacksburg, Virginia, USA). This controlled setting enabled a more rigorous test of the ability of the scanner to detect PIT tags through rocks. The flagstones used in this experiment were made of sandstone (Tennessee Blue grey stone) that had been cut in thin ( $\leq 5$  cm) flat sections. We secured the PIT tag horizontally or vertically beneath the flagstones. We sequentially stacked one flagstone at a time on top of the PIT tag. After each stone was added, we performed 10 30-s scans from the top. We measured the total height of obstructing rock, and the time it took to detect the tag. This process continued with each additional stone until the scanner failed to detect the PIT tag. In total, we tested nine distances ranging from 5.0–37.3 cm of flagstone material.

**Statistical methods.**—We carried out all statistical analyses in R (Version 3.2.2; R Core Team 2016) and used the lme4 package (Bates et al. 2015) and base package to build our models. For our artificial shelter occupancy experiment, we used a McNemar’s test to determine if the proportion of detections for the scans with rocks was significantly different from the proportion without rocks. We modeled the relationship between average cover rock depth and probability of detection with a binomial Generalized Linear Model (GLM).

We developed a binomial Generalized Linear Mixed Model (GLMM) to examine the main effects of PIT tag location and orientation within the shelters on the proportion of detections obtained during our PIT tag manipulation. We treated artificial cover shelter as a random effect to account for non-independence between measurements at the same artificial shelter. We selected an additive model to evaluate the combined influences of tag location and orientation on PIT tag true positive reads because this model performed better than a model with an interaction term between these variables ( $\Delta AIC = 4.1$ ; Table 1), and because there was no evidence of a significant interaction between the two variables. To determine whether location within the shelter influenced detection efficiency, we randomly set aside 16.7% of our

**TABLE 1.** The Binomial Generalized Linear Mixed Models developed to assess the effects of orientation (horizontal and vertical) and location within shelters (entrance, interface, wall, center, and corner) on the ability of a scanner to accurately detect PIT tags of Hellbenders (*Cryptobranchus alleganiensis*). Structure of each model is shown along with model Akaike Information Criterion (AIC) and significant *P*-values.

Model	Model Structure	AIC	$\Delta AIC$	<i>P</i> -values		
				Orientation	Interface	Wall
Additive	Orientation + Location + (1 Shelter)	213.2	0.0	0.005	0.011	0.016
Interactive	Orientation * Location + (1 Shelter)	217.3	4.1	0.070	0.015	0.012

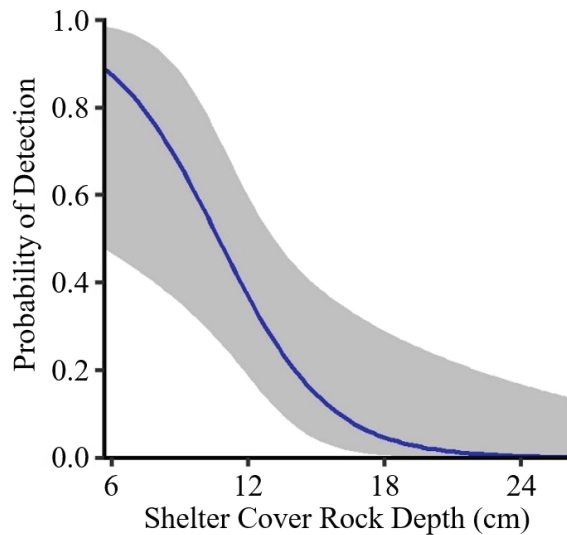


FIGURE 3. Relationship between artificial shelter cover rock depth and probability of detection (detection efficiency, true positive reads) of Hellbenders (*Cryptobranchus alleganiensis*). Shaded area denotes upper and lower bounds of 95% confidence interval around predicted probability.

data ( $n = 40$  scans, eight from each location), and treated these data as a reference group for comparing the relative detection efficiency of each of the five locations within the shelter.

For the scanner distance experiment, we developed two binomial GLMs to evaluate how the separate parameters of PIT tag orientation and overall distance between the scanner head and PIT tag affected detection efficiency. In addition to the GLMs, we used a Wilcoxon signed rank test to determine whether PIT tag orientation impacted the time to obtain a detection.

## RESULTS

**Artificial shelter occupancy.**—We scanned and verified occupancy status of 58 artificial shelters (Table 2). Overall, 32 artificial shelters were occupied by PIT-tagged animals. Presence of cover rocks significantly reduced detection efficiency. In with rocks scans, we obtained 31.25% detection efficiency (10 of 32), compared to 87.50% without rocks (28 of 32) on the same shelters ( $\chi^2 = 16.06$ ,  $df = 1$ ,  $P < 0.001$ ). Four Hellbenders went undetected by the scanner in both sets of scans. The average time to detection with rocks was 12.2 s and without rocks was 5.2 s. Combining both cover rock treatments, detection in 36 of 38 Hellbenders occurred within 20 s.

When considering only scans in which cover rocks were present, artificial shelters with detections had a lower average depth of cover rocks (10 cm) than artificial

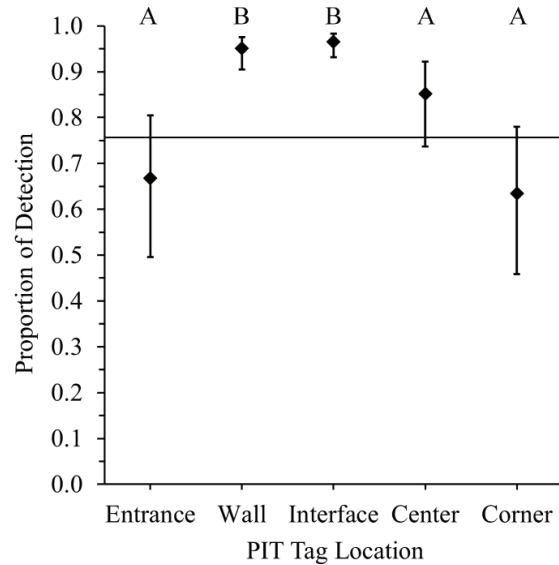


FIGURE 4. Effect of PIT tag location within the artificial shelter on proportion of detection (detection efficiency, true positive reads) of Hellbenders (*Cryptobranchus alleganiensis*). The horizontal line signifies the detection efficiency of a randomly selected reference group. Detection at locations marked A were not significantly different from the reference group proportion of detections. Detection efficiency at locations marked with B were significantly greater than the reference group ( $P < 0.05$ ). Error bars represent 95% confidence intervals.

shelters without detections (13.6 cm;  $Z = -2.95$ ,  $P < 0.005$ ). The probability of detecting a PIT-tagged animal in an artificial shelter was  $< 50\%$  when rock depth was  $\geq 11$  cm (Fig. 3). Across all 58 scanned artificial shelters, the average cover rock depth was 13.4 cm (range, 4.67–26.2 cm). The maximum cover rock depth with a detected Hellbender was 14.8 cm.

**PIT tag location and orientation.**—After 240 scans among five locations within artificial shelters (Fig. 2B), two of the locations (the interface and wall) had significantly higher detection efficiency than the randomly selected reference group (0.97 and 0.95 vs. 0.76; interface:  $Z = 2.553$ ,  $P < 0.050$ ; wall:  $Z = 2.401$ ,  $P < 0.050$ ; Fig. 4). PIT tag orientation also had a significant effect on the ability of the scanner to detect a Hellbender (Fig. 5). The vertical PIT tag orientation had significantly greater detection efficiency than the horizontal orientation (0.91 vs. 0.77;  $Z = 2.786$ ,  $P < 0.010$ ).

**Scanner distance.**—PIT tag orientation also had a significant effect on the ability of the scanner to detect the PIT tag in the scanner distance experiment, with vertically oriented tags having a significantly greater detection efficiency than horizontal tags ( $Z = 3.439$ ,  $P < 0.001$ ). The scanner detected the PIT tag in the horizontal

**TABLE 2.** Number and percentage of true versus false positive and negative scanner reads of PIT-tags of Hellbenders (*Cryptobranchus alleganiensis*) with and without cover rocks in place. Actual occupancy status of shelters was confirmed by manually checking and removing the Hellbender from each shelter. Manual checks revealed 32 shelters were occupied with a PIT-tagged Hellbender and 26 shelters were empty. In scans labeled With Rocks, one of three false positive scans occurred with an occupied box.

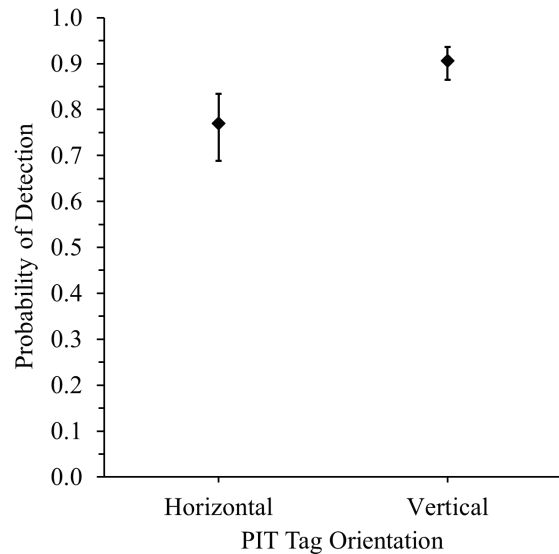
Classification	With Rocks	Without Rocks
True Positive	10	28
True Negative	24	25
False Positive	3	1
False Negative	21	4
Totals	58	58

and vertical orientation 100% of the time up to distances of 25.8 cm and 34.8 cm, respectively, and failed to detect the tag once the distance between the tag and scanner exceeded those values. The overall distance between the scanner antenna and PIT tag significantly affected the ability of the scanner to detect the tag, with shorter distances having higher detection efficiencies than longer distances ( $Z = -5.746, P < 0.001$ ). For the distances where both PIT tag orientations were detected, the average time to detection was shorter for the vertical orientation (1.06 s), than for the horizontal orientation (2.35 s;  $V = 599, P < 0.001$ ).

### DISCUSSION

The primary aim of this study was to assess whether PIT-tagged Hellbenders could be reliably detected in artificial shelters without having to remove cover rocks and open the shelters. We found that without cover rocks the scanner detected tagged Hellbenders 87.5% of the time. This efficiency is comparable to that reported in an aquatic PIT telemetry study (82%) of Slimy Sculpins (*Cottus cognatus*) implanted with 12.5 mm PIT tags (Cucherousset et al. 2005). Kelly et al. (2017) reported detection efficiencies of 79% for Mottled Sculpin (*Cottus bairdii*) and 16% for Creek Chub (*Semotilus atromaculatus*) using equipment comparable to this study. They attribute the significantly lower detection efficiency for Creek Chub to its use of deep pool habitats that are not ideal for this scanning technology. Additionally, it is important to note that the vast majority (92%) of our without rocks detections occurred within 20 s. Qualitatively, this indicates that our high detection efficiency without rocks can be obtained as rapidly with this scanning methodology as a visual and tactile shelter occupancy check.

Detection efficiency with cover rocks in place (31.25%) contrasted strongly with the higher detection efficiency observed after cover rock removal (87.5%). This



**FIGURE 5.** Effect of PIT tag orientation on proportion of detection (detection efficiency, true positive reads) of Hellbenders (*Cryptobranchus alleganiensis*). Error bars represent 95% confidence intervals.

discrepancy is most likely attributable to the inability of the scanner to read PIT tags through thick rocks; however, it is also possible that Hellbenders move around in their shelters when they sense vibrations caused by cover rock removal, and that these movements also increase scanning efficiency. The former explanation, however, is more plausible given that the detection efficiency of the scanner was significantly influenced by increasing cover rock depth, dropping below 50% when rock depth surpassed 11 cm. This effect of rock depth was further corroborated in the scanner distance experiment.

In the controlled scanner distance experiment, the maximum detection distances of the scanner (25.8–34.8 cm) were within the ranges of previous studies using the same or similar technologies conducted on fish and snake species (Cucherousset et al. 2005; Oldham et al. 2016; Kelly et al. 2017), suggesting that limitations we observed using the scanner are widely applicable. Knowing the maximum possible read distance through rocky material under controlled conditions is important for understanding the limitations of the scanner, and for comparison to performance under more complex field conditions. For qualitative comparison, the maximum rock depth obtained from with rocks scans in the artificial shelter experiment, summed with the average artificial shelter height (14.83 cm + 11 cm = 25.83 cm), was the same as the maximum detection distance in the controlled scanner distance experiment with horizontal tag orientation (25.80 cm). The 31% overall detection efficiency we observed, however, when scanning *in-situ* shelters with cover rocks contrasted with the 100% detection efficiency of the scanner at similar distances in the controlled distance experiment. We hypothesize this difference may be due

to surface irregularities that the scanner has to overcome when scanning across cover rocks in the field, requiring that the antenna be placed at varied angles when scanning shelters. This likely interferes with the ability of the device to obtain a signal at the same efficiency as the controlled environment, where the antenna head could be maintained at a constant level plane relative to the tag throughout scanning. Because we obtained much higher detection efficiency in the field once cover rocks were removed (87.5%), we find it unlikely that construction elements of the shelter (e.g., metallic components), or environmental factors (e.g., water chemistry), account for the performance gap in our field test relative to the controlled experiment. These results also suggest that this technology could be employed to detect Hellbenders under natural boulders < about 25 cm thick, thus avoiding disturbance of individuals using natural habitat.

In both the PIT tag location/orientation experiment and the scanner distance experiment, we found that vertical PIT tag orientation had significantly greater detection efficiencies than horizontally oriented tags. Thus, it is possible that detection efficiency would have been greater during the artificial shelter occupancy experiment if tags had been implanted vertically in the tails of Hellbenders. Furthermore, the scanner distance experiment revealed that PIT tag orientation significantly affects both detection efficiency and the time required to obtain a detection, with vertically oriented tags being read 55% quicker. Consequently, researchers may reconsider how they PIT-tag Hellbenders if hoping to employ a similar scanning technology in their study system; however, prior to doing so, the feasibility, safety, and retention rate of vertically implanted tags would have to be tested. Studies in terrestrial plethodontid and ambystomatid salamanders, have also shown horizontal tag orientation negatively affects detection efficiency (Cucherousset et al. 2008; Connette and Semlitsch 2013; Ousterhout and Semlitsch 2014).

We also found that PIT tag location within the artificial shelter influenced the detection efficiency of the scanner, with two PIT tag locations (interface and wall) yielding approximately 20% higher detection than the randomly generated reference group. Due to the size of the scanner relative to the shelter and the scan pattern, it is possible that the interface and wall are actively scanned for a greater period of time than the other locations, leading to increased detection efficiency at these locations. As Hellbenders do not use the same area of the artificial shelter throughout the year, understanding this nuance in the performance of the scanner is important for the development of standardized scanning procedures. For example, during the pre-breeding period (early to mid-August), male Hellbenders tend to occupy shelter tunnels and guard them as prospective nesting sites (pers. obs.). During this time of year, the procedure could be modified to focus on the tunnel to increase detection efficiency for

guarding males.

We found this technology to be an effective tool for surveying Hellbenders in artificial shelters after removing large cover rocks. However, it should be noted that in systems with many untagged individuals, performing shelter checks using this technology alone (in the absence of confirmatory visual and tactile investigation of shelters) will result in an underestimation of occupancy. Our experiments suggest that we could increase the utility of the scanner even further by eliminating the need for cover rocks and by considering implanting PIT tags vertically. As a result, we are experimentally testing alternative methods for stabilizing and camouflaging artificial shelters within the context of a variety of stream conditions, shelter designs, and shelter installation locations. If further refined to maximize detection efficiency, this technology could become a valuable tool for long-term population monitoring surveys, monitoring of males guarding nests, and assessment of localized movement patterns and shelter use.

*Acknowledgments.*—We thank all of the generous landowners who allow us to access their land study this species and J.D. Kleopfer for his support throughout the study. We would like to thank Clara Frazier for assistance in the field along with Matt Lacey, Tiffanie Pirault, Alex Grimaudo, John Hallagan, Cathy Jachowski, Valentina Alaasam, Jeronimo Gomes Da Silva Neto, and Hank Vogel for their help in constructing and installing artificial shelters. This research was supported by an undergraduate research fellowship from Dr. Dennis Dean and the Fralin Life Science Institute at Virginia Tech, as well as the Virginia Department of Game and Inland Fisheries. All protocols were approved under the Virginia Department of Game and Inland Fisheries handling permit (Virginia DGIF Permit No. 060465) and the Virginia Tech Institutional Animal Care and Use Committee (VT IACUC No. 16-162).

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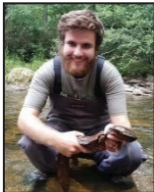
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**JOHN R. CONNOCK** received his B.S. in Fish and Wildlife Conservation in May 2018 from Virginia Tech, Blacksburg, Virginia, USA. He has worked in the Hopkins lab since his sophomore year at Virginia Tech and obtained a Fralin Undergraduate Research Fellowship that allowed him to conduct a senior thesis investigating the use of portable PIT tag scanning technology to remotely survey for Eastern Hellbenders. His research interests include amphibian and reptile ecology, population ecology, and habitat management. He is now a Masters Student at Murray State University, Murray, Kentucky, USA. (Photographed by Brian F. Case).



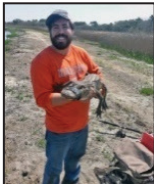
**BRIAN F. CASE** received a B.A. in Biology from Bucknell University, Lewisburg, Pennsylvania, USA, in 2015. His interests encompass the impacts of human activity (e.g., forest removal, agriculture, impervious development) on the healthy functioning of freshwater systems and amphibian species. After graduation, he spent two years in environmental policy interning in the U.S. Congress, as a blogger for Oceana, and as a fellow for the federal legislative team of The Nature Conservancy. He is currently a graduate student in the Hopkins lab studying the physiological and paternity correlates of hellbender reproductive biology, nest success, and paternal care. (Photographed by Jordy Groffen).



**SKY T. BUTTON** is a Master's student in the Hopkins Lab. His interests broadly focus on using community, reproductive, physiological, and climate change ecology research on amphibians and reptiles as tools to inform more efficient conservation decision-making. For his thesis, Sky is continuing the Hopkins lab's research on Hellbender conservation, examining extrinsic factors linked to reproductive success in the Eastern Hellbender (*Cryptobranchus alleganiensis alleganiensis*). He is also investigating ways to improve artificial shelter placement in streams to augment existing Hellbender breeding habitat in areas where recruitment has been anthropogenically reduced. (Photographed by William A. Hopkins).



**JORDY GROFFEN** obtained two M.Sc. degrees in Animal Sciences, specializing in ethology, welfare and adaptation physiology (Wageningen University in the Netherlands and Swedish University of Agricultural Sciences in Sweden) in 2012. His main research interest is in ecology of herpetofauna, invasive species and parasites. Jordy currently studies Hellbender ecology at Virginia Tech, Blacksburg, Virginia, USA, and has previously studied crocodiles, frogs, salamanders, flying foxes, toads, snails, and freshwater turtles. (Photographed by Brian F. Case).



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