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# Comparison of Movement Patterns in Captive-Released Eastern Hellbenders (*Cryptobranchus alleganiensis alleganiensis*) Using Three Different Release Methods

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Comparison of Movement Patterns in Captive-Released  
Eastern Hellbenders (*Cryptobranchus alleganiensis alleganiensis*)  
Using Three Different Release Methods

by

Julie A. Boerner

An Abstract of a Thesis  
in  
Biology

Submitted in Partial Fulfillment  
of the Requirements  
for the Degree of

Master of Arts

December 2014

Buffalo State  
State University of New York  
Department of Biology

## ABSTRACT OF THESIS

Comparison of Movement Patterns in Captive-Released  
Eastern Hellbenders (*Cryptobranchus alleganiensis alleganiensis*)  
Using Three Different Release Methods

Eastern hellbender (*Cryptobranchus alleganiensis alleganiensis*) population size has declined throughout much of its range. Previous captive-release headstarting programs have resulted in minimal success, presumably due to movement of captive-released animals away from the release site. This study aimed to increase the success of hellbender headstarting programs by assessing the effectiveness of three release methods. Releases were conducted in two stream sites within the Allegheny River drainage. Streams were similar; however stream A contained a higher boulder density. In each site, three salamanders were placed individually in cages, three salamanders were placed individually in nest boxes with the entrance blocked with screen, and three salamanders were released directly under cover rocks. Animals were monitored between 18 June 2013 and 12 October 2013 using radio telemetry. Results showed little difference in total movement and survivorship between stream sites or treatments. Number of movements was marginally significantly higher in stream B. The number of movements was not different between release types. Overall survival was low; only three animals were found alive for longer than six months. Four animals were never recovered, three were found dead, and eight transmitters were found. Movement was most dependent on the phase of the moon. Both distance and frequency of movement increased with greater moon phase. Captive-released animals generally moved further than what has been reported in wild hellbenders, with an average cumulative distance moved of  $653 \pm 138$  m (SE). The information gathered from this study could aid further captive-rearing projects, as well as inform monitoring and survey efforts.

State University of New York  
Buffalo State  
Department of Biology

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A Thesis in  
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By

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## **Introduction**

### *Conservation and Movement*

Declines in amphibian populations have been noted since the 1980s (Semlitsch 2002, Collins and Halliday 2005). Factors such as climate change, pollution, loss of habitat, and disease may be contributing to these declines (Blaustein and Bancroft 2007). However, with lack of information on described species, the exact causes are not well known (Collins and Halliday 2005). Amphibians are particularly vulnerable to environmental changes. Permeable skin, shell-less eggs, and complex life cycles put many species at risk of contaminants, changing temperature and precipitation levels, and UV radiation (Blaustein and Bancroft 2007). In this way, amphibians can serve as indicator species for environmental stress and are therefore valuable to conservation biologists (Collins and Halliday 2005). It is vital to the conservation of many amphibian species to acquire more data on their life histories, habitat requirements, and movement patterns (Semlitsch 2002, Collins and Halliday 2005, Blaustein and Bancroft 2007). Increasing the current knowledge base could help improve future management strategies.

Many amphibian populations are in need of active management. Relocation, repatriation, and translocation (RRT) programs, including captive-release programs, have been used to promote the survival of certain species. Relocation is the deliberate movement of individuals or populations from an area where they are threatened, particularly by development and other human activities, to an area with fewer threats to their survival. Repatriation is the release of individuals into an area formerly or currently occupied by the same species in order to recover or build the local population. Translocation is the release of individuals into areas not formerly occupied by that species. These new areas typically contain higher quality habitat than the species' natural habitat (Dodd and Seigel 1991). Success rates for captive-release and RRT

programs have been historically low (Stamps and Swaisgood 2007). In a 1991 study, Dodd and Seigel reviewed RRT programs for 25 amphibian and reptile species. Projects were considered successful if there was evidence of the establishment of a self-sustaining population. The outcome of a project was considered unclassified if monitoring time or the amount of data were inadequate to classify it as a success or failure. Projects were considered failures if a self-sustaining population was not established. Of the 26 projects reviewed, 19% were successful, 23% were unsuccessful, and 58% were unclassified. The authors noted that lack of knowledge of the species was a likely overall cause of failure. Germano and Bishop (2009) examined the results of 91 RRT projects published between 1991 and 2006. The project success rate for these studies was 41%. The most common reported cause of failure for amphibian releases was poor habitat, followed closely by homing and movement away from the release site. Other reported causes of failure included insufficient release numbers, human collection, and inclement weather. Germano and Bishop (2009) suggested that releasing animals at a younger growth stage may increase project success rates. In another RRT project review, Griffiths and Pavajeau (2008) found that of 21 reintroduced species, three displayed evidence of surviving in the wild (low success), 13 had bred successfully in the wild (partial-success), and five had established self-sustaining populations (high success). This constitutes an approximate 24% success rate. In general, less than half of RRT projects succeed. It is important to understand the reasons for failure of past projects before future efforts are undertaken.

The success rate of many RRT programs is often determined by habitat suitability. Bodinof et al. (2012) found that captive-reared juvenile Ozark hellbenders (*Cryptobranchus alleganiensis bishopi*) released at a site with densely arranged boulders showed 1.5-fold higher survival than hellbenders released at sparse boulder sites. Mean body condition (determined from

length and weight measurements recorded upon recovery) at the sparse boulder site was satisfactory, while body condition at the dense boulder site was excellent. In addition, hellbenders at the sparse boulder site had a greater proportion of physical abnormalities, leech parasites, and chytrid fungus (Bodinof et al. 2012). Nickerson (1980) simulated poor habitat condition in a laboratory experiment to assess effects of habitat suitability on wild-caught adult Ozark hellbenders. Thirty hellbenders were removed from optimal habitat in the North Fork of the White River, Missouri and placed in aquaria. Animals were subjected to lack of substrate, non-aerated water, and reduced food supply. Over the course of the experiment—approximately four months—hellbenders lost 32-65% of their capture weight. At the end of the laboratory experiment, animals were returned to the site of capture. After three months, all animals had increased dramatically in body weight, indicating the importance of suitable habitat on hellbender body condition (Nickerson 1980).

A concern associated with RRT projects is that animals will reject selected sites with suitable habitat due to unfamiliarity and move to areas with poor habitat. It is possible that low success rates of RRT projects may be due to this factor, and as a result, increased detectability may also lead to predation (Stamps and Swaisgood 2007). In a movement study of adult natterjack toads (*Bufo calamita*), Husté et al. (2006) found that out of 24 original animals, three died either from predation or human factors associated with movement. Reinert and Rupert, Jr. (1999) found that only 36.7% of translocated adult timber rattlesnakes (*Crotalus horridus*) survived. Predation, likely due to high movement rates, was the primary cause of death. In a study of translocated adult mountain yellow-legged frogs (*Rana muscosa*), Mathews (2003) observed a loss in body mass in animals due to movement and homing behavior. Confining animals in enclosures at release sites for a period of time may help them to acclimate to the new

environment. This acclimatization process often reduces distances traveled after release (Stamps and Swaisgood 2007). Semlitsch (2002) suggested that aquatic breeding amphibians should be headstarted in cages secured within natural stream habitats. In this way, they are raised in their natural environment, and should remain nearby once released. Techniques such as these may benefit hellbender headstarting projects.

### *Hellbender Biology*

The Eastern hellbender is one of two subspecies of the genus *Cryptobranchus*, the other being the Ozark hellbender. Eastern hellbenders are the largest salamanders in the western hemisphere, with adults averaging 50 cm in length and potentially reaching up to 74 cm (Mayasich et al. 2003). Hellbenders are fully aquatic. Though they have lungs, these are primarily used for buoyancy control. Lateral skin folds allow for cutaneous gas exchange, which is the primary form of respiration (Guimond 1970).

Hellbenders are typically found in swift, shallow, highly oxygenated streams with rocky substrate (Hillis and Bellis 1971, Mayasich et al. 2003). They are primarily nocturnal, though diurnal activity has been noted, mainly during the breeding season (Mayasich et al. 2003, Humphries 2007). Laboratory experiments have indicated that hellbenders may not be strictly nocturnal, but may have early morning peaks of activity (Noeske and Nickerson 1979). Crayfish are the main food source for adult hellbenders, though they will also consume fish and other organisms, as well as occasionally eggs and larvae of their own species (Peterson et al. 1989, Mayasich et al. 2003).

Breeding generally occurs in the fall, although the exact timing and length of the breeding season varies by population. In general, Eastern hellbender populations begin breeding in mid-late August through mid-September (Ingersol et al. 1991, Mayasich et al. 2003). Males

excavate nests beneath large, flat rocks or similar structures with an entrance facing downstream. Females enter these nests, often followed by transient males. Females usually release two strings of eggs, forming strands that twist together (Mayasich et al. 2003). A single female may deposit between 200 and 400 eggs, and several females may deposit eggs in the same nest (Topping and Ingersol 1981, Mayasich et al. 2003). Eggs are externally fertilized by the male. After oviposition occurs, the female leaves the nest. The male typically remains near the egg mass for up to three weeks (Mayasich et al. 2003).

At the time of hatching, larvae range from 27 to 33 mm in length and possess external gills. They develop rapidly within the first few weeks post-hatching. After one year, larvae average 70 mm in length and gills are reduced. Juveniles undergo incomplete metamorphosis at approximately eighteen months. At two years, gills are greatly reduced or absent (Mayasich et al. 2003). Age at sexual maturity has been estimated at 3-4 years (Smith 1907) and 5-6 years (Bishop 1941), though this is still debated and may depend on size (Mayasich et al. 2003). Sexually mature males can be identified during the breeding season by a swollen ridge of tissue around the cloaca. Gravid females exhibit a swollen abdominal region where the eggs are located (Topping and Ingersol 1981, Mayasich et al. 2003). Since hellbenders show negligible sexual dimorphism, this is the only known reliable way of telling the sexes apart (Mayasich et al. 2003, Makowsky et al. 2010). Eastern hellbenders have frequently been shown to exhibit unequal sex ratios, with males outnumbering the females (Hillis and Bellis 1971, Humphries and Pauley 2005, Foster et al. 2009, Burgmeier et al. 2011). Hellbenders are believed to live 30 or more years (Peterson et al. 1983, Mayasich et al. 2003). Little is known about dispersal in juveniles. However, a number of mark-recapture surveys have recorded movements in adults (Mayasich et al. 2003).

### *Current Status*

Historically, Eastern hellbenders were found throughout much of the Eastern United States in the Susquehanna, Ohio, Tennessee, and Mississippi drainage systems (Mayasich et al. 2003). Today, this range has been greatly reduced (Figure 1). Nickerson et al. (2003) compared hellbender populations in Tennessee's Little River and the North Fork of Missouri's White River and found populations to be almost entirely composed of adults. Burgmeier et al. (2011) obtained similar results when surveying a population in southern Indiana, in addition to finding extremely low population densities. A 20+ year study covering five rivers and both subspecies revealed that populations of hellbenders have declined by an average of approximately 77% (Wheeler 2002).

In 2011, the Ozark hellbender was listed as endangered under the Endangered Species Act (USFWS 2013). The Eastern hellbender is currently under consideration for federal endangered species listing. However, it is already listed as endangered in Maryland, Ohio, Illinois and Indiana, and as threatened in Alabama. In New York, it is listed as a species of special concern (NYDEC 2013). Both the Ozark and the Eastern hellbender are listed under Appendix III of the Convention on International Trade in Endangered Species of Wild Flora and Fauna (CITES). Under CITES, an export or re-export permit issued by the Management Authority of the State is required in the case of trade and permits may only be issued if the specimen was obtained legally (CITES 2013).

In New York State, the Eastern hellbender is found only in the Allegheny (within the Ohio) and Susquehanna River drainage basins (Figure 2). Populations in these basins show similar trends to other populations throughout the hellbender's range. In a survey of the Allegheny River drainage, Bothner and Gottlieb (1991) captured 293 hellbenders at eight different sites. In 2009, Foster et al. repeated this study and found that hellbenders were

extirpated from one of Bothner and Gottlieb's (1991) historical sites in the Allegheny River drainage. A total of 159 individuals were captured and ecological density had decreased in all sites but one (0-2/10 m<sup>2</sup> from <1 to 6/10 m<sup>2</sup> reported by Gottlieb (1991)). In addition, populations contained more mature individuals than juveniles (Gottlieb did not find any juveniles in his original surveys although Foster did).

Destruction or modification of habitat is considered one of the main threats to Eastern hellbender populations (Wheeler et al. 2002, Foster et al. 2009). Siltation and habitat modification reduces cover and foraging habitat, and also leads to pollution and other water quality issues (Mayasich et al. 2003). Overutilization and overharvesting for commercial, scientific, and educational purposes also may have had an adverse effect on populations in the past. Climate change is a growing concern, reducing water levels in some areas and increasing the amount of UV-B radiation penetrating the water column (Mayasich et al. 2003). Other potential threats to Eastern hellbender populations include introduced species, recreational fishing, pathogen outbreaks, and problems with implementation and enforcement of current regulations (Mayasich et al. 2003). Some combination of these factors could be causing the declines in hellbender populations observed recently.

#### *Hellbender Movement*

Several studies have been done regarding hellbender movement patterns. Burgmeier et al. (2011) found that adult Eastern hellbenders moved very little over the course of the year. When they did move, it was over relatively short distances (mean: 27.5 m) to nearby shelter rocks (and occasionally bedrock, downed trees, or root masses). Peterson (1987) observed that mean net movement of adult Eastern hellbenders did not differ significantly from 0 m/day. Hillis and Bellis (1971) performed a mark-recapture study on Eastern hellbenders in northwest

Pennsylvania. Out of a total of 152 individuals captured and marked between 11 June, 1968 and 3 August, 1968, 81 were recaptured between 5 August, 1968 and 21 September, 1968. Of these recaptured individuals, 54.8% moved 10 m or less from the original capture site. The remaining 45.2% were recaptured greater than 10 m from the original capture site, with a maximum movement distance of 160 m. Humphries and Pauley (2005) also performed a mark-recapture study of Eastern hellbenders in West Virginia. Based on 99 captures of 44 individuals, the mean intercapture distance observed was 35.8 m. In another mark-recapture study of Eastern hellbenders conducted in the Allegheny River, Foster et al. (2009) found all recaptured animals at the original site of capture except one female captured >1 km upstream from the original site.

Nickerson (1980) performed a movement study on Ozark hellbenders (*C. a. bishopi*). Thirty animals were removed from the North Fork of the White River (NFWR) in Missouri, and then returned to the same site 4-7 months later. After three months, the animals were found once again. Five of the 27 hellbenders (three died during transport) were recovered at the same site of release. The authors did not specify the fate of the other 22 animals. Nickerson and Mays (1973) also performed a mark-recapture study in the NFWR. A total of 569 individuals were tagged, and 58 were recaptured. Of these recaptures, 70% were less than 30 m from the original capture site and 34% were found at the original site (including at least two recaptured under the same rock of initial capture). Only three individuals were found more than 90 m from the original capture site. The authors did not speculate as to the fate of the 511 animals not recaptured.

Gates et al. (1985) found evidence of movement in translocated adult Eastern hellbenders. Animals were taken from the Allegheny River drainage in Pennsylvania and released into a Maryland stream. Dispersal was predominantly downstream, though individual distances varied widely (range: 0-2340 m). Movements were most frequent during high stream

discharge periods associated with storm events. Most individuals eventually established home ranges around one or more cover rocks. Bodinof et al. (2012) observed that captive-released juvenile Ozark hellbenders tended to move towards areas with cobble-boulder substratum (although 8% were found in bank crevices and root masses). These movements were primarily downstream, however one individual exhibited a net movement of approximately 500 m upstream.

#### *Buffalo Zoo Headstarting Program and Captive-Release*

Foster et al. (2009) found that hellbender numbers in the Allegheny River drainage had suffered a recent decline. This information prompted the New York State Department of Environmental Conservation (NYSDEC) to collect a hellbender egg mass from the Allegheny River main stem in 2009. Individuals were hatched from these eggs and raised in the Buffalo Zoo. In the summer of 2011, 46 of these animals were released. After four weeks, 37% were located. Another set of 100 individuals were released in the summer of 2012, however only eight were located again (Roblee, pers. comm.). This may indicate poor survival, significant movement, or non-detection. Recent hellbender release efforts similar to those of the NYSDEC have indicated that survivorship is most tentative in the first few weeks. Bodinof et al. (2012) performed a study on captive-reared Ozark hellbenders (*C. a. bishopi*) released into the North Fork of the White River, Missouri. Using radio telemetry, they found that short-term settlement, health, and survivorship were influenced by the distance to cobble-boulder substrata. In West Virginia, released captive-reared Eastern hellbenders moved frequently in the first few weeks (Greathouse, pers. comm.). In a similar study in Ohio, frequency of movement was greatest in the first 21 days post-release (Lipps, pers. comm.).

## *Objectives*

In conjunction with the hellbender headstarting program undertaken by the NYSDEC and the Buffalo Zoo, eighteen captive-reared animals were implanted with radio transmitters and released into two streams within the Allegheny River drainage.

The objectives of this study are as follows:

1. Determine if movement patterns and survival vary by release type.
2. Determine if movement patterns and survival vary by stream location.

## *Significance*

This study aims to determine the most effective release method for captive-reared Eastern hellbenders. Determining the most effective, cost-efficient, time-efficient method is highly important for informing future RRT programs. The success or failure of the various release methods used in this study, as well as the data collected on hellbender movement, will benefit future headstarting and release programs in New York State and in other areas of the Eastern hellbender's range. Movement has been cited as a cause of failure of many RRT programs. Monitoring is critical to the success of projects such as this.

## **Methods**

### *Study Animals*

Eighteen captive-reared juvenile Eastern hellbenders were released at two sites within the Allegheny River drainage and were monitored post-release. Hellbenders were raised at the Buffalo Zoo from eggs collected in 2009 from the Allegheny River main stem. Study animals were chosen from among the 302 remaining healthy individuals in the Buffalo Zoo headstarting program. The top 1% by weight (three individuals) were excluded to limit size-related bias and the bottom 30% by weight (99 individuals) were excluded to avoid risk of injury due to

transmitter size. Eighteen study animals were chosen from the remaining 200 individuals using a random number generator. Transmitters were implanted in early May by the Buffalo Zoo veterinary staff using methods previously developed by hellbender researchers (Stouffer et al. 1983, Briggler pers. comm., Lipps pers. comm.). Advanced Telemetry Systems (Isanti, MN, U.S.A.) model F1170 transmitters with a slow pulse rate (Pulser R: 30 ppm, Pulser W: 15 ms) were used. Transmitters weighed approximately four grams each. Animal weights ranged from 203 g to 338 g. These weights fall within the recommended transmitter:animal mass ratio of 3-5% outlined by Browne et al. (2011). At the time of surgery, mass and length of all animals were recorded. Approximate age of the animals at the time of surgery was four years. Genders were unknown. Hellbenders were monitored for approximately one month for healing prior to release. During this recovery period, animals were housed six per tank at the Buffalo Zoo.

#### *Study Sites and Treatments*

Stream locations to be used as study sites were chosen by the NYSDEC as ideal for hellbender release. The first site was located on a tributary near Portville, NY. The second site was located on a tributary near the town of Hinsdale, NY (Figure 3). Landowners were contacted prior to release for property access permission. IACUC approval was acquired prior to animal release (IACUC #26).

Crayfish surveys were conducted between 28 July 2013 and 29 July 2013 in stream A and between 26 July 2013 and 27 July 2013 in stream B. For each survey, three minnow traps were baited with cat food. Traps were tied off to rocks within the study site (one trap upstream, one trap midstream, and one trap downstream corresponding to treatment placement). Traps were set in the afternoon and left overnight (approximately 16 hours). After this time, each trap was inspected and total number of crayfish caught was determined. For each individual crayfish, sex

and species was determined. Each crayfish was also measured to the nearest mm with manual calipers placed around the carapace. Crayfish were released after data collection.

Boulder density and stream width surveys were conducted between 11 August 2013 and 12 August 2013 in stream A and between 19 August 2013 and 21 August 2013 in stream B. For each location, the stream bank was marked every 2 m between the downstream treatment site and the upstream treatment site. At each mark, the width of the stream from bank to bank was measured to the nearest 0.01 m. A random number generator was used to randomly sample 15% of the marked transects for boulder density. The total number of boulders (defined as a rock  $\geq 30$  cm at one axis, Bodinof et al. 2012) within 1 m of each randomly selected transect was recorded.

Each of the two stream locations received three identical study treatments. 1) Soft-release cage treatment: Three salamanders were placed in cages (one animal per cage) designed by Kenneth Roblee, NYSDEC. Cages were constructed of 3 cm x 3 cm vinyl coated steel mesh with a 2.5 cm x 2.5 cm vinyl mesh inlay, 1.5 m x 1.8 m and 0.3 m in height (Figure 4). Cages were staked in place in an approximately 0.5 m deep section of the stream with a large cover rock located in the center and open to natural stream substrate on the bottom. Cage edges were sealed with gravel and large rocks. 2) Soft-release nestbox treatment: Three salamanders were placed in nestboxes (one animal per box) with the entrance blocked with vinyl coated steel mesh. Nestboxes were constructed of concrete approximately 0.33 m x 0.43 m with an approximately 0.17 m x 0.51 m long tunnel attached (Figure 5). This design was originally developed by Jeff Briggler in Missouri and has been used in several states for hellbender nest habitat (Briggler and Ackerson 2012). The nestboxes used in this study were designed by Greg Lipps and constructed by Bluffton Precast Concrete, Bluffton, OH. Greg Lipps and the Columbus Zoo provided these nestboxes. Nestboxes were embedded in the stream locations two weeks prior to hellbender

placement. At the time of hellbender placement, the pH inside each nestbox was measured to ensure conditions were safe for study animals. 3) Hard-release treatment: Three salamanders per stream location were released directly under a suitable cover rock in the stream (one animal per rock).

All treatments were clustered within a 10 m radius (one of each type of release in each cluster, three clusters per site). Clusters were placed at least 20 m apart within each location (Figure 6). This distance was maximized as appropriate based on suitable areas in the stream reach. Sites for treatment clusters were chosen by looking for a suitable hard-release rock, suitable substrate near that rock for placement of both soft-release treatments, and as close to the center of the stream as possible.

#### *Release and Monitoring*

Study animals were placed in stream A on 18 June 2013 and in stream B on 20 June 2013. Animals were chosen randomly from the 18 implanted with transmitters for release type, cluster site, and stream location. Placement was done by hand. Initial data including velocity, depth, temperature, distance to shore, and GPS point were recorded at each treatment location (Figure 7).

Animals were located using a Communications Specialists (Orange, CA, U.S.A.) receiver (model R1000) and a Telonics rubber “H” type antenna (model RA23). In a pre-release equipment test on dry land, I was able to locate transmitters 100% of the time. Each salamander was located daily between 18 June 2013 and 22 August 2013 unless conditions prevented. Animals were then located once every other week between 23 August 2013 and 12 October 2013. Monitoring time of released animals occurred at the same time of day (morning or afternoon) and alternated between stream locations. Between monitoring sessions, all equipment

was decontaminated using a 10% bleach solution to ensure no pathogens were transferred between streams. Animals were located by following the transmitter signal to within a meter (or less) of their actual location.

Nestbox animals were released on 30 June 2013 by removing the mesh covering the nest opening. Nestboxes were left in place to minimize disturbance and to provide supplemental habitat. The initial plan was to release caged animals after three months in the stream. However, all but one animal escaped before this time (see Table 1 for exact escape dates). The remaining animal was released on 5 August 2013 by propping the cage up so that there was space for it to get through underneath but without actually disturbing the animal.

When an animal was located, data were recorded as outlined in Figure 7. Latitude/longitude was recorded using a Magellan SporTrak Pro Handheld GPS unit. Habitat in which the hellbender was residing was defined as “run,” “riffle,” or “pool.” Depth of animal location was recorded to the nearest cm. Length and width of the rock (or other substrate) under which the hellbender was residing was measured as the longest axis (length) and the corresponding longest perpendicular axis (width), recorded to the nearest cm. Cover type was recorded as “rock,” “log,” “snag,” “root mass,” “vegetation mat,” or “stream bank.” Definitions and example photos of cover objects can be found in Appendix III. The density of boulders (defined as a rock  $\geq 30$  cm at one axis, Bodinof et al. 2012) within 1 m of the rock (or other substrate) where the hellbender was currently residing was recorded as “low” (0-2 boulders), “medium” (3-5 boulders), or “high” (>5 boulders). Velocity was recorded using a Swoffer M2100 propeller type flow meter to the nearest 0.01 m/s. Distance to nearest shoreline was recorded to the nearest m. Water temperature was recorded as °C. In addition, lunar cycle data were obtained from the United States Naval Observatory (2012). Data were collected at

midnight, Eastern Standard Time. Daily moon phase was recorded on a scale of 0 (new moon) to 1 (full moon) to the nearest hundredths. In addition, data on lunar brightness as a function of lunar phase were obtained from Heiken et al. (1991) and recorded on a scale of 0 (no light) to 1 (100% light) to the nearest hundredths.

### *Analysis*

Mixed models were used to analyze what prompted animal movement (discrete, 0/1) and movement distance (continuous, meters) as a function of the measured variables. Individual animals and date were included as random effects to account for autocorrelation from repeated measurements. Model selection was based on Akaike's Information Criterion (AIC) (Akaike 1973) using maximum likelihood. Generally, the lowest  $\Delta$ AIC value indicates the best-fit model (Burnham and Anderson 2002). Velocity (m/s) was collinear with depth (cm) (variance inflation  $>6$ ; Fox and Weisberg 2014) and was left out of the final analysis. Lunar brightness was collinear with lunar phase and was also left out of the final analysis. The best-fit model retained depth (cm), cover type, and moon phase. A generalized linear mixed model (GLMM) assuming a binomial error distribution was used for animal movement. The GLMM was fit using the Laplace approximation in the "lme4" package (Bates and Maechler 2009) in the R statistical programming environment (R Development Core Team 2012). The coefficients and their interactions were analyzed using an ANCOVA with the "car" package, which calculates a Wald chi square (Fox and Weisberg 2014). Coefficients with a p-value  $\leq 0.05$  were considered 'significant', and coefficients with a p-value  $\leq 0.10$  were considered 'marginally significant' (*sensu* Hurlbert and Lombardi 2009). A linear mixed model (LMM) was used for movement distance. The coefficients and their interactions were analyzed using an ANCOVA with the pamer function in the "LMERConvenienceFunctions" (Tremblay and Ransijn, 2013).

Distance moved (m), number of movements, and number of days located also were analyzed by release type (hard-release, soft-release with cages, soft-release with nestboxes), release location, and animal weight using analysis of variance (ANOVA) models with the "lmtest" package (Hothorn et al. 2014) for the R statistical programming environment (R Development Core Team 2012). Since only two stream locations were used in this study, *t*-tests were used to analyze differences in crayfish numbers, boulder densities, and stream width between stream locations.

## Results

All animals behaved differently, regardless of stream location or treatment type (Figures 8-9). Each animal exhibited its own unique movement pattern. Appendix I and Appendix II contain GPS maps of individual movement patterns in stream A and stream B, respectively. Cumulative distance moved across all animals ranged from 13.9 m to 1,892.3 m, with a mean of  $653 \pm 138$  m (SE) (Table 1).

There was a significant “moon phase x cover type” interaction ( $F_{5,1}=2.74$ ,  $p=0.02$ ). For all cover types except root mass, distance moved from cover increased with increased moon phase. Distance moved decreased with increased moon phase from the root mass cover type (Figure 10). Animals also moved more frequently with increased moon phase ( $\chi^2_1=6.68$ ,  $p=0.01$ ). The mean ( $\pm$ SE) moon phase during which animals moved was  $0.63 \pm 0.04$ , whereas animals generally moved less frequently when the moon phase was less (mean= $0.43 \pm 0.02$ ; Figure 11).

Number of movements was marginally significant between stream locations, with a higher number of movements in stream B (mean= $5.67 \pm 0.65$ ) than in stream A (mean= $4.00 \pm 0.50$ ) ( $F_{1,11}=4.13$ ,  $p=0.07$ ; Figure 12). There was no significant difference in either cumulative movement (m) ( $F_{1,11}=0.18$ ,  $p=0.68$ ) or number of days located ( $F_{1,11}=0.14$ ,  $p=0.71$ ) between the

two stream locations (Figure 12). There was no difference in cumulative number of movements ( $F_{2,11}=1.40$ ,  $p=0.29$ ), cumulative movement (m) ( $F_{2,11}=1.74$ ,  $p=0.22$ ), or number of days located ( $F_{2,11}=0.58$ ,  $p=0.58$ ) between caged, hard release or nest box animals (Figure 13).

There was no significant effect of animal weight (g) on cumulative number of movements ( $F_{1,11}=0.06$ ,  $p=0.81$ ), cumulative movement (m) ( $F_{1,11}=0.05$ ,  $p=0.83$ ), or number of days located ( $F_{1,11}=0.02$ ,  $p=0.89$ ). The majority of movements were directed downstream, however eleven of the eighteen study animals exhibited some upstream movement (not exceeding 406 m) (Figure 14).

Though more crayfish were caught in stream A, variance between samples was high therefore there was no significant difference in number of crayfish caught between streams ( $t=1.18$ ,  $df=2$ ,  $p=0.36$ ). Stream A had a significantly higher boulder density (mean= $7.35\pm 1.84$ ) than stream B (mean= $2.80\pm 0.54$ ) ( $t=2.27$ ,  $df=19$ ,  $p=0.04$ ). Stream width was marginally significant between stream locations, with a larger average width in stream B (mean= $16.8\pm 0.6$  m) than in stream A (mean= $15.5\pm 0.4$  m) ( $t=-1.88$ ,  $df=64$ ,  $p=0.07$ ).

## **Discussion**

### *Movement*

Movement was most dependent on the phase of the moon. Generally, hellbenders moved more often and moved further with higher moon phase. This relationship was true for most cover types. It is unclear why the relationship was opposite for the root mass cover type. There may be different degrees of light penetration between cover types; however this would need to be studied further. Also, in this study I did not measure nor calculate luminosity. Local factors may have impacted the amount of light detected by the animals. The effect of lunar phase may be independent of light intensity and so further study of this result is warranted. Movement during

periods of high moon phase does not seem to fit what is known about animal movement. Mitchell and Hazlett (1996) found that crayfish movements (particularly in *Orconectes virilis* and *O. propinquus*) followed the lunar cycle, with less movement during a full moon and more during the new moon. Cloud cover did not alter this effect. The authors' proposed explanation of this behavior was reduction of predation risk. Detectability is increased on moonlit nights, and so predation risk would be higher. Therefore, crayfish would benefit from moving less during nights near the full moon (Mitchel and Hazlett 1996). Since crayfish, including *Orconectes propinquus*, are the main food source of hellbenders, it would be assumed that they would follow the same activity patterns. However, this was not the case.

FitzGerald and Bider (1974) observed effects of the lunar cycle on activity levels in the American toad (*Bufo americanus*). Toads were less active during the full moon and more active during the new moon. The authors proposed a dual mechanism to explain this relationship. Toads may benefit by eating insects that are more active under low light conditions, while remaining safe from nocturnal predators, themselves (FitzGerald and Bider 1974). Ralph (1957) also found a similar relationship in the red-backed salamander (*Plethodon cinereus*) and proposed the predation reduction explanation. Since hellbenders are the prey of many predatory species, it would be expected that they would avoid moving during times of increased visibility (such as during the full moon). Again, this was not the case.

Though little literature exists on movement patterns in relation to the lunar cycle in hellbenders, it is likely that wild hellbenders behave similar to other species. The relationship observed in these study animals was therefore unexpected. Though the reason for this is unclear, it is possible that the way in which these particular animals were raised had an effect. Hellbenders released in this study were raised in clear tanks and supplied with small cover

objects that did not effectively eliminate all outside light. Animals were never exposed to complete darkness. They were kept on a daily schedule of bright white light during the day, blue light to mimic moonlight in the late afternoon, and then the security lights in the building they were housed during the night. In the wild, hellbenders tend to stay under large boulders in deep areas of the stream or river where very little light would be able to penetrate. Captive-released animals may therefore not have been used to the darkness of a new moon and so would choose to move during brighter nights. It is important to note that this study did not measure moonlight intensity or the local effects of variables such as cloud cover, topography, and shading on light intensity. However, several studies have shown that these variables do not significantly alter the relationship between moon phase and animal movement (Fitzgerald and Bider 1974, Mitchel and Hazlett 1996, Ralph 1957). Further studies need to be done to assess the effect of the lunar cycle on movement in wild hellbenders in comparison to captive-reared hellbenders, as well as if this relationship applies to the majority of captive-reared animals. This could have important implications for hellbender conservation, such as how animals should be raised in captivity and when they should be released into the wild.

The majority of studies of wild adult hellbenders revealed short distance movement. Burgmeier et al. (2011) recorded an average movement distance of 27.5 m over the course of a year. Humphries and Pauley (2005) recorded an average of 35.8 m over the course of two years. Hillis and Bellis (1971) recorded a maximum movement distance of 160 m between 11 June, 1968 and 21 September, 1968. Peterson (1987) found that mean net movement did not differ significantly from 0 m/day between 5 September, 1985 and 7 November, 1985. Interestingly, larger movements have been recorded in translocated and captive-released hellbenders. Gates et al. (1985) observed cumulative distances moved ranging between 0 and 2,340 m in translocated

Eastern hellbenders monitored between August and December, 1981 and between June and September, 1982. Bodinof et al. (2012) observed a cumulative movement of 500 m in one individual between 2008 and 2009. In comparison, I found that study animals generally moved further than previous records. Cumulative distance moved ranged between 14 and 1,892 m with an average cumulative distance moved of  $653 \pm 138$  m and an average daily movement of  $11 \pm 2$  m over the course of approximately four months. Clearly, these animals are moving much further than wild animals that have never experienced captivity.

Timing of movement was similar to what has been previously observed in captive-reared hellbenders. Captive-reared Eastern hellbenders in West Virginia moved frequently in the first few weeks (Greathouse, pers. comm.). In Ohio, frequency of movement was greatest in the first 21 days post-release (Lipps, pers. comm.). Animals in this study had stopped moving by 21 August, 2014, constituting a 64-day post-release movement period. While this is a much longer length of time than in previous studies, most movements occurred in the first few weeks post-release, with animals eventually settling down later in the summer. This study presents observations on a small group of eighteen individuals. Further studies need to be conducted using more individuals to determine if these patterns apply to more captive-released animals.

### *Release Types*

There were no differences between the three different release types. Confining animals within the release site for a period of time reduces the distance moved after release (Stamps and Swaisgood 2007). However, I did not observe this in the captive-released animals used in this study. The effect of the lunar cycle on animal movement may suggest that the problem lies in the raising of captive animals before they are released. If this program was to be repeated, conditions should more closely match the natural environment in terms of lighting and substrate. Semlitsch

(2002) suggested that animals should be headstarted in cages secured within natural stream habitats. This may be a viable option.

Stamps and Swaisgood (2007) also hypothesized that animals may move further after release if the acclimatization process was in some way traumatic. It is possible that while hellbenders were confined in nestboxes and cages they became stressed either from lack of food, exposure to disease, or some other cause. This may have led to increased movement post-release resembling increased movement in hard-released animals. Since all but one caged animal (Animal A2) escaped before the planned release time, it is difficult to draw definitive conclusions on this release type. Currently, Kenneth Roblee of the NYSDEC is continuing the cage study using a different cage design that will hopefully decrease escapes. It remains to be seen whether this new design will decrease animal movement and increase survival post-release.

#### *Stream Locations*

Number of movements was higher in stream B than stream A, though there were no significant differences in total movement or number of days located between streams. Both locations were tributaries of the Allegheny River and were near each other, so no differences were expected. There appeared to be no differences in food resources between the two streams. However, stream A did have a significantly larger boulder density than stream B, which potentially could have contributed to the difference in number of movements. Bodinof et al. (2012) observed that captive-reared Ozark hellbenders typically selected sites with coarse substratum, and therefore more cover objects. Nickerson (1980) also described the importance of suitable habitat for Ozark hellbenders. When exposed to poor habitat conditions, hellbenders lost 32%-65% of their body weight. They then increased dramatically in body weight when placed back in habitat with suitable substrate (Nickerson 1980). Germano and Bishop (2009) found that

the most common reported cause of failure of amphibian RRT projects was poor habitat. There were fewer boulders in stream B, which may be the reason why animals in this location moved more often. Stream A was also visibly affected by siltation. These poor habitat conditions may have caused increased movement in study animals. In addition, stream B was wider than stream A, though this was only marginally significant and probably did not contribute to differences between the two stream locations.

### *Use of Cover Objects*

Contrary to what has been published about hellbenders, study animals used a wide variety of cover objects. Hillis and Bellis (1971) reported that Eastern hellbenders use large rocks in the stream for cover, and many sources have corroborated this. While I did observe hellbenders using rocks as cover, many other types of cover objects were used including logs, snags, root masses, vegetation mats, and stream banks. The use of many of these cover objects has not been comprehensively described in hellbenders. Burgmeier et al. (2011) observed occasional use of logs and root masses by wild adult Eastern hellbenders. Bodinof et al. (2012) also observed the use of bank crevices and root masses by captive-reared Ozark hellbenders. An egg mass was also discovered in a bank crevice in the Allegheny River drainage (Miller, pers. comm.). The discovery of hellbenders using types of cover objects which have not been thoroughly described in the species creates the potential for finding more wild hellbenders. Current surveys focus on what is widely considered as typical hellbender habitat— large, flat rocks in the center of swift, shallow, highly oxygenated streams (Hillis and Bellis 1971, Mayasich et al. 2003). If surveys were expanded to cover all possible cover objects, even the unconventional ones, new individuals may be found.

After animals were released from nestboxes, the nestboxes were left in the stream with the expectation that study animals would use them as cover. However, animals used them infrequently post-release. Between 14 July 2013 and 21 July 2013, Animal B1 (a cage animal) was located in a nestbox. The animal then moved out and into the same nestbox before vacating it completely on 25 July 2014. Between 15 July 2013 and 17 July 2013, Animal B3 (also a cage animal) was located in the same nestbox at the same time as Animal B1. It vacated the nestbox on 18 July 2013. This is not the only occasion two animals were found using the same cover object. Between 1 July 2013 and 2 July 2013, Animals B5 and B4 were located under the same root mass at the same time. This behavior is unusual in hellbenders, a usually solitary animal (Mayasich et al. 2003). Study animals were raised in groups of 6-10 animals at the Buffalo Zoo. It is therefore possible that these animals may have lost their solitary nature, at least in part, by being exposed to conspecifics during captive-rearing.

On two occasions study animals were seen partially exposed within the stream. On 2 July 2013, Animal A9 was located under a small log in the stream. A portion of the animal's tail was fully visible. On 15 July 2013, Animal A4 was located in a crevice within the stream bank. The animal's head was exposed. Appendix IV contains photos of these two events. Neither of these animals moved as I approached them. It is unclear whether these animals lacked fear, were unaware that they were exposed, or were otherwise incapable of performing antipredator behaviors. Animal A4 was ultimately lost to predation. Unfortunately, animal A9's signal was lost. If these animals did lack antipredator capabilities, this could be detrimental to the survival of captive-release animals. Exposing animals to predatory cues before release may also effectively decrease predation rates. Gall and Mathis (2009) observed innate predator recognition in lab-reared hellbender larvae and suggested that learning may also play a role in predator

avoidance. In future captive-release programs, larval hellbenders should be conditioned to predatory cues in order to test this hypothesis and potentially increase survival of headstarted animals.

### *Fate of Study Animals*

Before the end of the data collection period (12 October 2013), ten animals had died and the signals of three other animals were lost (See Table 1 for animal information). After thorough searching, these three animals (A3, A7, and B5) could not be relocated. It is unclear whether these animals were removed from the stream by a predator and carried far away, the transmitters stopped working, or the animals moved into the Allegheny River mainstem where they would be difficult to relocate.

In six of the ten confirmed deaths, only the transmitter was found. The transmitter appeared "licked clean," but there were no bite marks or scratches. Four of these transmitters (A1, A6, A8, and B7) were found out of the stream up to 20 m from the stream edge and buried under leaf litter. Raccoon (*Procyon lotor*) tracks were found near these transmitters, indicating that this was the likely predator. The other two transmitters (A4 and B9) were found in shallow water near the stream edge under the silt layer. The PIT tag of animal A4 was also found with the transmitter. Again, there were no bite marks or scratches on either of these transmitters. No other predatory evidence was found with these two transmitters, however, the way in which the transmitters were found was consistent with mink (*Mustela vison*) or river otter (*Lontra canadensis*), with the former being the likely predator. No signs of river otter presence were found in either stream location during the course of the study. Appendix V shows examples of the evidence found with these transmitters.

In two of the ten confirmed deaths, some remains were found along with the transmitter. The transmitter for Animal B3 was found approximately 4m from shore under the leaf litter, and some entrails were also found. The transmitter for Animal B1 was found less than one meter from the stream edge with some spinal remains (Appendix VI – a). No bite marks or scratches were found on the transmitters and no other predatory evidence was found. These two events seem consistent with raccoon predation, but cannot be confirmed.

In one of the ten confirmed deaths, the animal was found on the stream bank less than a meter from the water (B2). The body had been dried out by the sun and so a necropsy could not be performed (Appendix VI – b). No predatory evidence was found and it appears this animal may have beached itself. On two separate occasions, animals were found in a similar way, but were alive. The first of these events occurred on 25 July 2013. Animal B9 was found on land less than a meter from water but appeared completely healthy and was quickly released back into the stream. This animal was found dead almost a month later on 19 August 2013 (only the transmitter was found). On 4 August 2013, Animal A5 was also found on land right beside the water, but was showing signs of oxygen deprivation (transmitter incision line, toes, cloacal slit, and gill slits were reddened; Appendix VII). The animal was placed in a tank with stream water and an airstone and was monitored for approximately an hour. When the animal appeared healthy, it was released back into the stream. This animal survived to the end of the data collection. No predatory evidence was found on or around these animals and it appears that these animals may also have beached themselves.

In one of the ten confirmed deaths, the animal was found whole and apparently untouched by predators in a pool of water separated from the main stream flow (Appendix VI – c-f). The animal (B8) showed no outward signs of trauma. However, its skin was bluish-grey in

color. A necropsy of the animal performed by Dr. Kurt Volle of the Buffalo Zoo revealed an enlarged left atrium of the heart, and chytrid tests performed by the San Diego Institute for Conservation Research yielded a positive result, however the cause of death was undetermined (Volle, pers. comm.). The strange behavior observed in this animal does seem to be consistent with symptoms of chytrid fungus (*Batrachochytrium dendrobatidis* or "Bd"). Chytrid presents itself in a number of ways, including epidermal alteration and loss of functioning (i.e., respiration in many amphibians including hellbenders), congestion of organs (possibly leading to enlargement of organs such as the heart), and behavioral changes (Densmore and Green 2007). Many of the signs of chytrid infection are difficult to diagnose in living animals, and so may go unnoticed until death occurs. Bodinof et al. (2012) observed chytrid fungus in four out of 24 captive-released Ozark hellbenders. Only one of these four animals died (death recorded 70 days post-release). One animal was able to shed the fungus 264 days after it tested positive (Bodinof et al. 2012). If these study animals were indeed infected with chytrid, it may have affected their ability to absorb oxygen via the skin. This would explain the behavior of staying close to shore and beaching, as hellbenders are able to absorb a small amount of oxygen from the air, though inefficiently, directly into their lungs during hypoxic conditions (Smith 1907, Ultsch and Duke 1990). In addition, infected animals—particularly those staying close to shore and beaching themselves—would be more vulnerable to predation. This may account for the rather high predation rate by terrestrial predators. Though chytrid testing could only be performed on one animal in the current study, it is possible that many of these hellbenders were exposed to the fungus. Further studies are needed to determine the effects of chytrid on both wild and captive-released hellbenders. It is unclear why some animals are less affected by the fungus than others.

If the answer to this question is determined, measures could be taken to ensure more animals survive exposure to chytrid.

Five animals survived to the end of the data collection period. However, since 12 October 2013, one animal was lost and one animal died. Animal A9's signal was lost on 16 November 2013 and after thorough searching could not be relocated. Animal A2's transmitter was recovered on 5 November 2013 in thick underbrush a short way from the stream edge with no bite marks or scratches and no other evidence found (Roblee, pers. comm.). The three remaining transmitter signals (A5, B4, and B6) have stayed within a meter of the last recorded position since 19 August 2013. Recovery of these transmitters was not possible due to unsafe stream conditions and it is not known if the animals are still alive.

Bodinof et al. (2012) performed a similar study on captive-reared Ozark hellbenders. Between May 2008 and August 2009, 36 hellbenders were monitored. By the end of the study, 16 salamanders were alive, 13 had died, and seven were lost (Bodinof et al. 2012). Clearly, these animals fared far better than my 18 animals, with only five presumed alive after the initial four month monitoring period. It is unclear why certain individuals did better than others and why survival rates differ between studies done in separate locations.

### *Conclusions*

The Eastern hellbender salamander is a species in need of active management. Wheeler (2002) found that hellbender populations have declined by an average of 77% since the 1970s. Populations in the Allegheny River drainage of New York State, where this study was conducted, show similar trends (Foster et al. 2009). It is likely that this pattern of loss will continue unless measures are taken to reverse it. Current captive-release efforts may not be enough to ensure the survival of the Eastern hellbender. Existing populations face numerous

threats. The best conservation strategy would involve minimizing current threats while supplementing populations. Future headstart programs should focus on effectively mimicking natural conditions (i.e., natural moonlight and substrate) and potentially introducing juveniles to predatory cues. The release types used in this study were ineffective and should be tested again with new designs to help improve release success. In addition, supplementing cover rocks in release locations may decrease movement and increase survival post-release. Improving captive-release programs while implementing actions such as habitat reclamation hold the most potential to conserve the Eastern hellbender salamander.

Table 1. Summary data for each individual animal recorded between 18 June, 2013 and 12 October, 2013 (except when noted \*, indicating death or disappearance post-data collection)

Animal	Weight (g)	Length (cm)	Release Location	Release Type	Total No. of Movements	Total Movement (m)	No. of Days Located	Date of Escape	Date of Death/Disappearance
A1	252	34	Stream A	Cage	1	13.9	38	7/25/2013	7/26/2013 <sup>a</sup>
A2	304	34	Stream A	Cage	3	321.1	116		*11/5/2013 <sup>a</sup>
A3	222	34	Stream A	Cage	4	816.0	37	6/21/2013	7/25/2013 <sup>b</sup>
A4	236	34	Stream A	Hard	4	1548.2	37		7/25/2013 <sup>a</sup>
A5	234	33.5	Stream A	Hard	5	608.8	116		
A6	212	34	Stream A	Hard	6	236.3	45		8/2/2013 <sup>a</sup>
A7	261	35.5	Stream A	Nest	5	202.5	37		7/29/2013 <sup>a</sup>
A8	260	34.5	Stream A	Nest	5	615.3	54		8/11/2013 <sup>a</sup>
A9	258	34	Stream A	Nest	3	1042.0	116		*11/16/13 <sup>b</sup>
B1	234	33.5	Stream B	Cage	8	923.7	60	6/21/2013	8/19/2013 <sup>c</sup>
B2	220	33	Stream B	Cage	3	340.9	44	6/22/2013	8/3/2013 <sup>c</sup>
B3	288	34	Stream B	Cage	4	80.8	38	7/1/2013	7/28/2013 <sup>a</sup>
B4	260	33	Stream B	Hard	7	1892.3	114		
B5	312	35	Stream B	Hard	7	117.8	35		7/25/2013 <sup>b</sup>
B6	248	34	Stream B	Hard	4	1749.4	114		
B7	263	34	Stream B	Nest	4	146.1	32		7/22/2013 <sup>a</sup>
B8	338	34.5	Stream B	Nest	6	826.8	44		8/3/2013 <sup>d</sup>
B9	203	33	Stream B	Nest	8	272.4	60		8/19/2013 <sup>a</sup>

<sup>a</sup> Transmitter found without body – presumed eaten

<sup>b</sup> Disappeared

<sup>c</sup> Body found on land – cause of death unknown

<sup>d</sup> Cause of death unknown – chytrid positive

Figure 1. Current distribution of hellbenders by subspecies (NYDEC 2013).

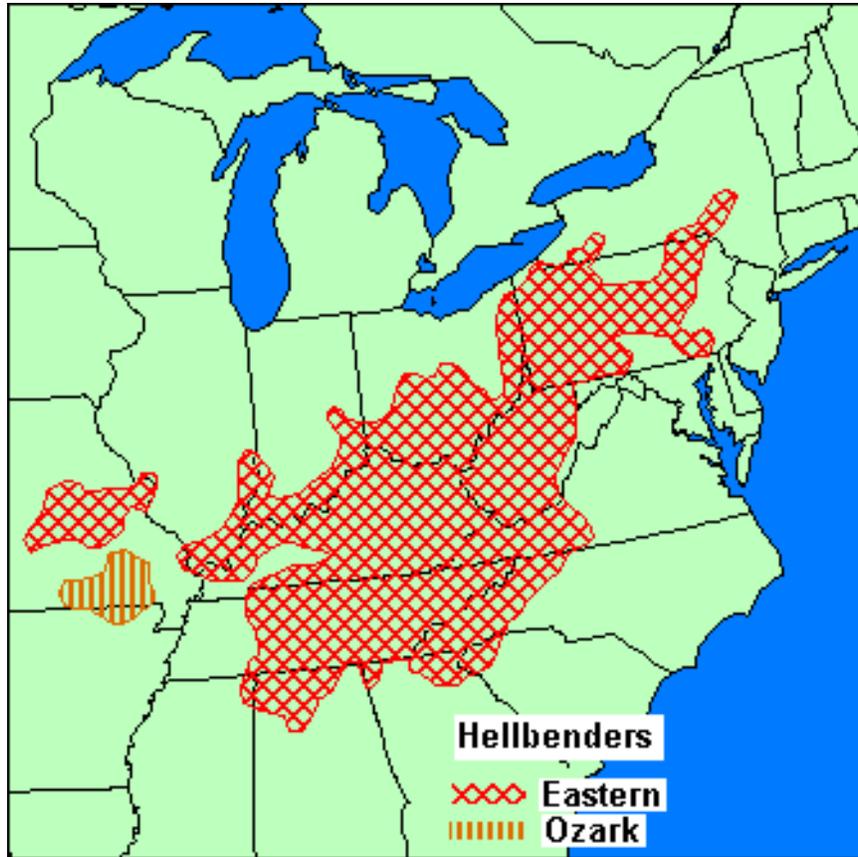


Figure 2. Range of the Eastern hellbender in New York State as reported by the New York State Amphibian and Reptile Atlas Interim Report. Data were collected from 1990-2007 (NYDEC 2013).

New York State  
Amphibian and Reptile Atlas

1990-2007 Data  
=== Interim Report ===

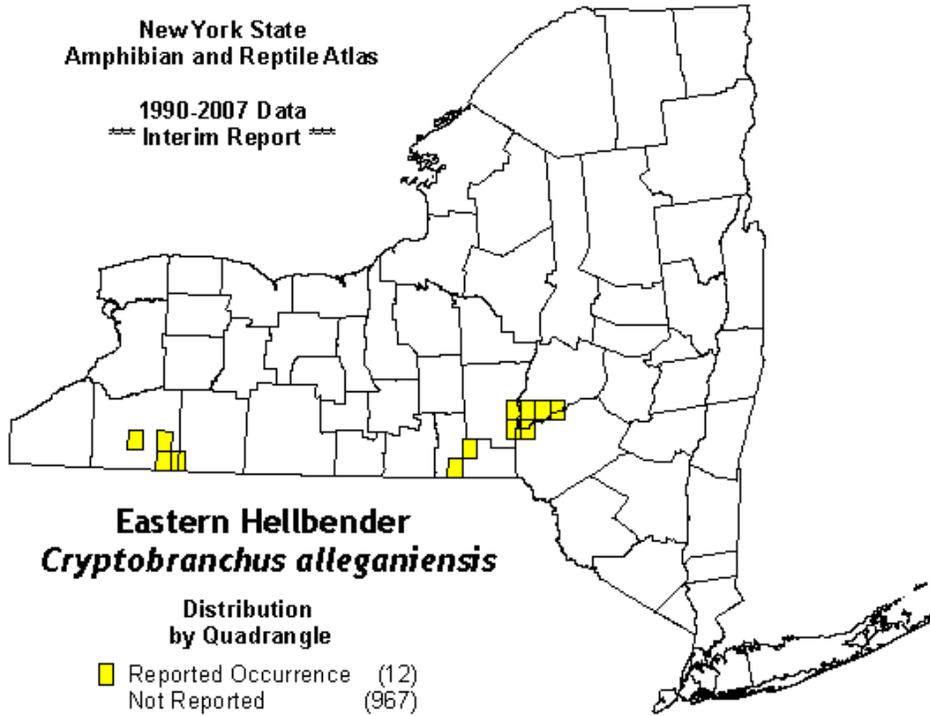


Figure 3. Map depicting approximate study site locations.

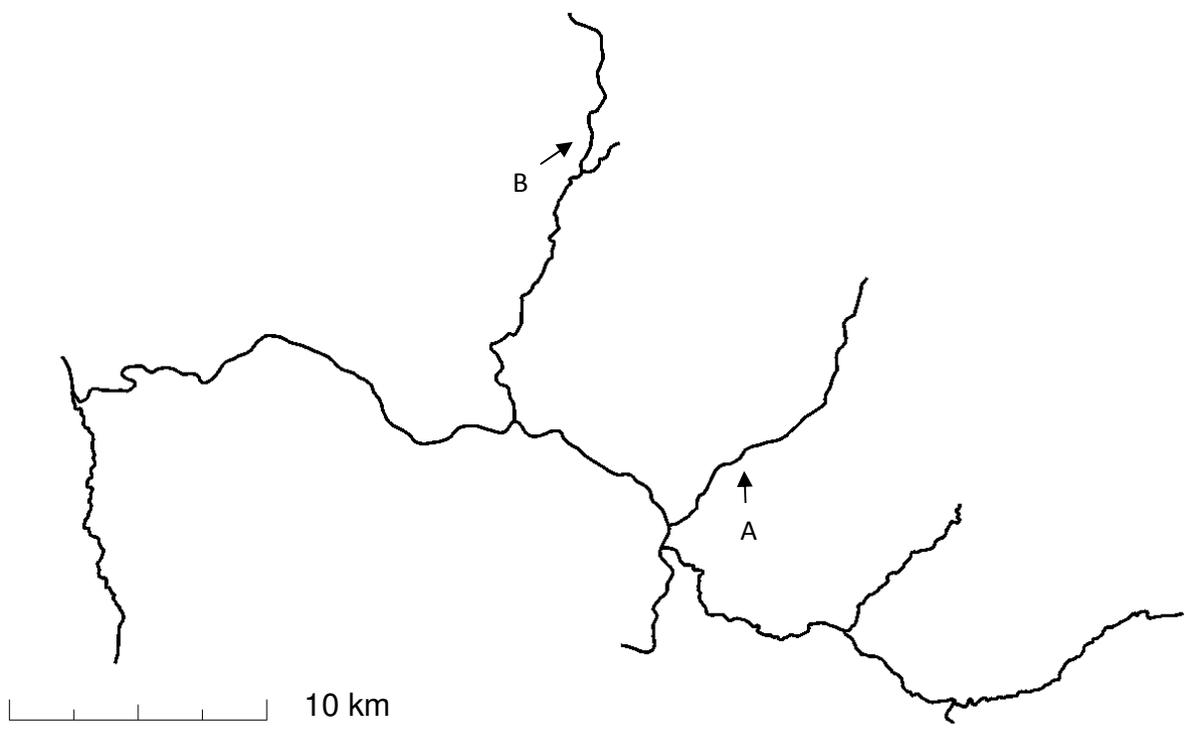


Figure 4. Schematic of cage construction (a), dry cage (b, photo by Amy McMillan), and cage in stream (c, photo by Ren Koithan).

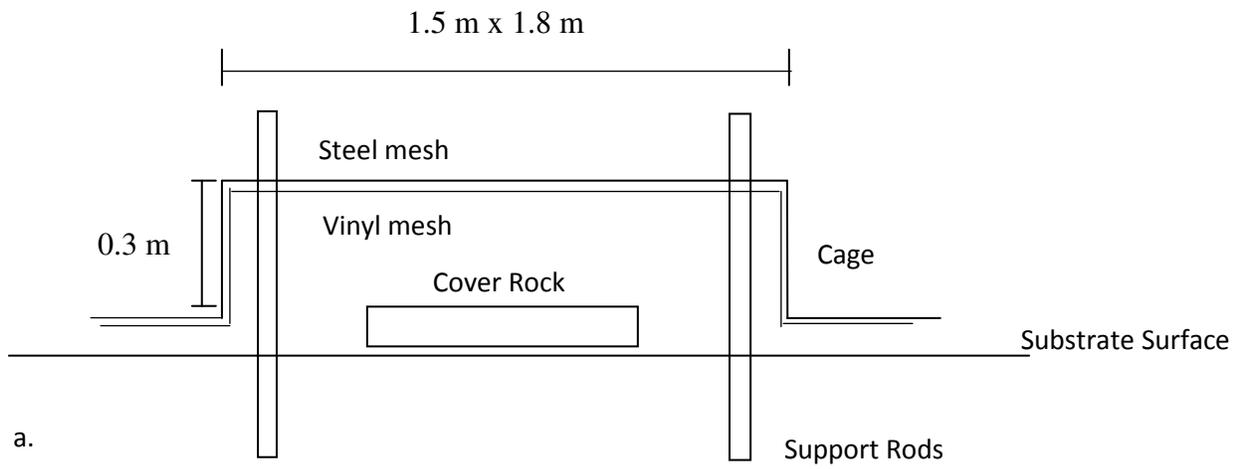
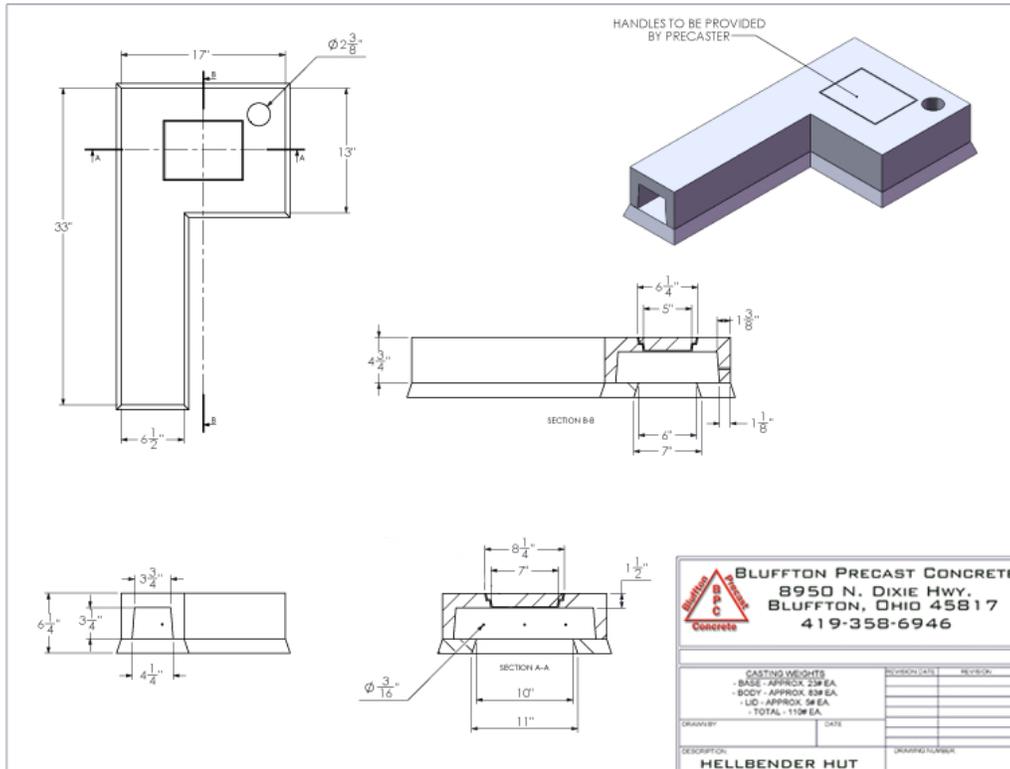


Figure 5. Schematic of nestbox construction (a), dry nestbox (b, photo by Greg Lipps), and nestbox in stream (c).



a.

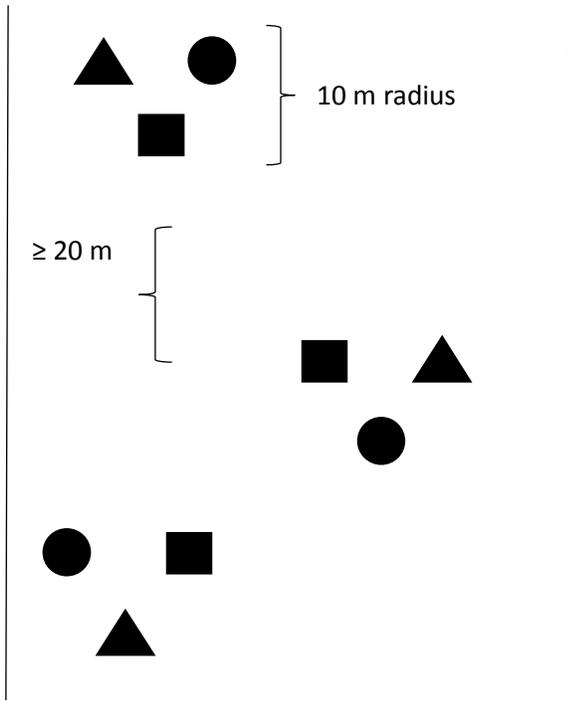


b.



c.

Figure 6. Schematic of treatment placement within a stream. Shapes represent individual treatment types.



Stream Flow



Figure 7. Sample data collection sheet.

Date: \_\_\_\_\_

**Eastern Hellbender Data**

Initials: \_\_\_\_\_

164.	Time	Moved Yes No	Latitude	Longitude	Waypoint	Habitat	Depth (cm)	Velocity (m/s)	Temp (°C)
						Pool Riffle Run			
	<b>Notes:</b>						<b>LxW (cm)</b>	<b>Bldr</b>	<b>Shore (m)</b>
								Low Medium High	

Figure 8. Cumulative distance moved in meters over time per animal for the “stream A” release location recorded between 18 June 2013 and 12 October 2013. Line termination indicates death or disappearance of animal. “a” = nestbox animals released, “b” = date of major rain event, “c” = remaining caged animal released.

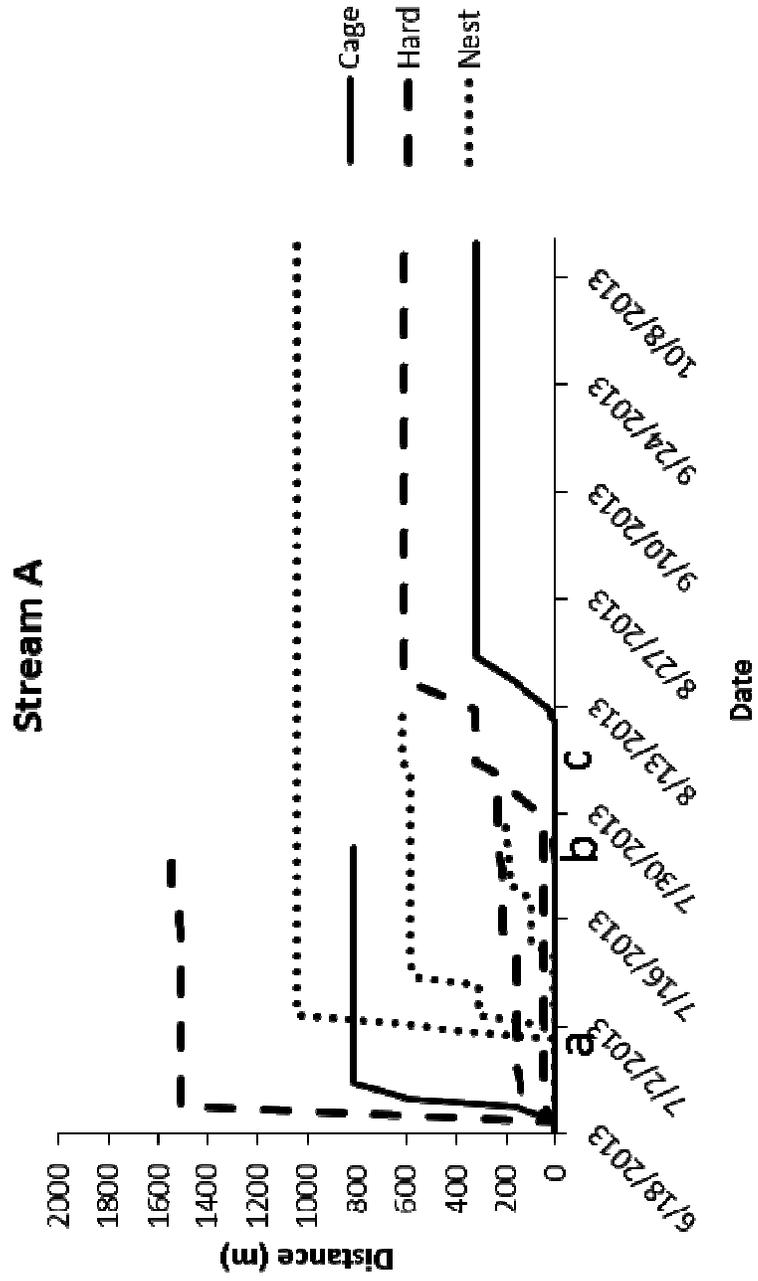


Figure 9. Cumulative distance moved in meters over time per animal for the “stream B” release location recorded between 20 June 2013 and 12 October 2013. Line termination indicates death or disappearance of animal. “a” = nestbox animals released, “b” = date of major rain event.

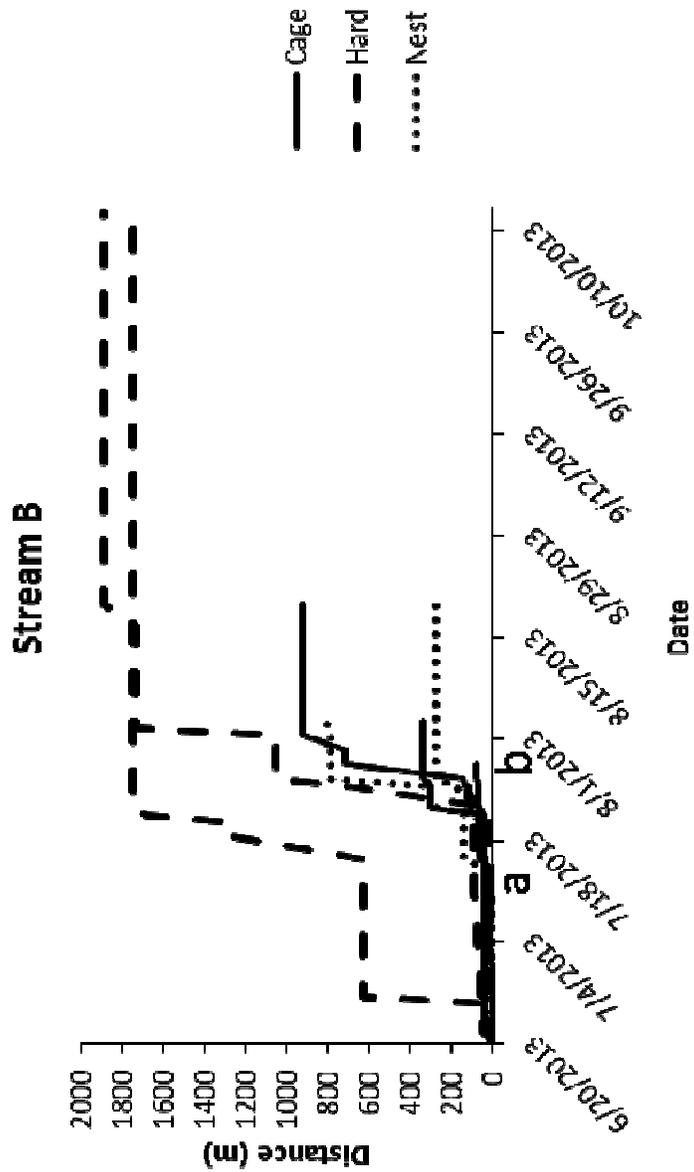


Figure 10. Interaction plot of the effects of cover type and fraction of the moon illuminated on distance moved from cover recorded between 18 June 2013 and 12 October 2013 with number of observations.

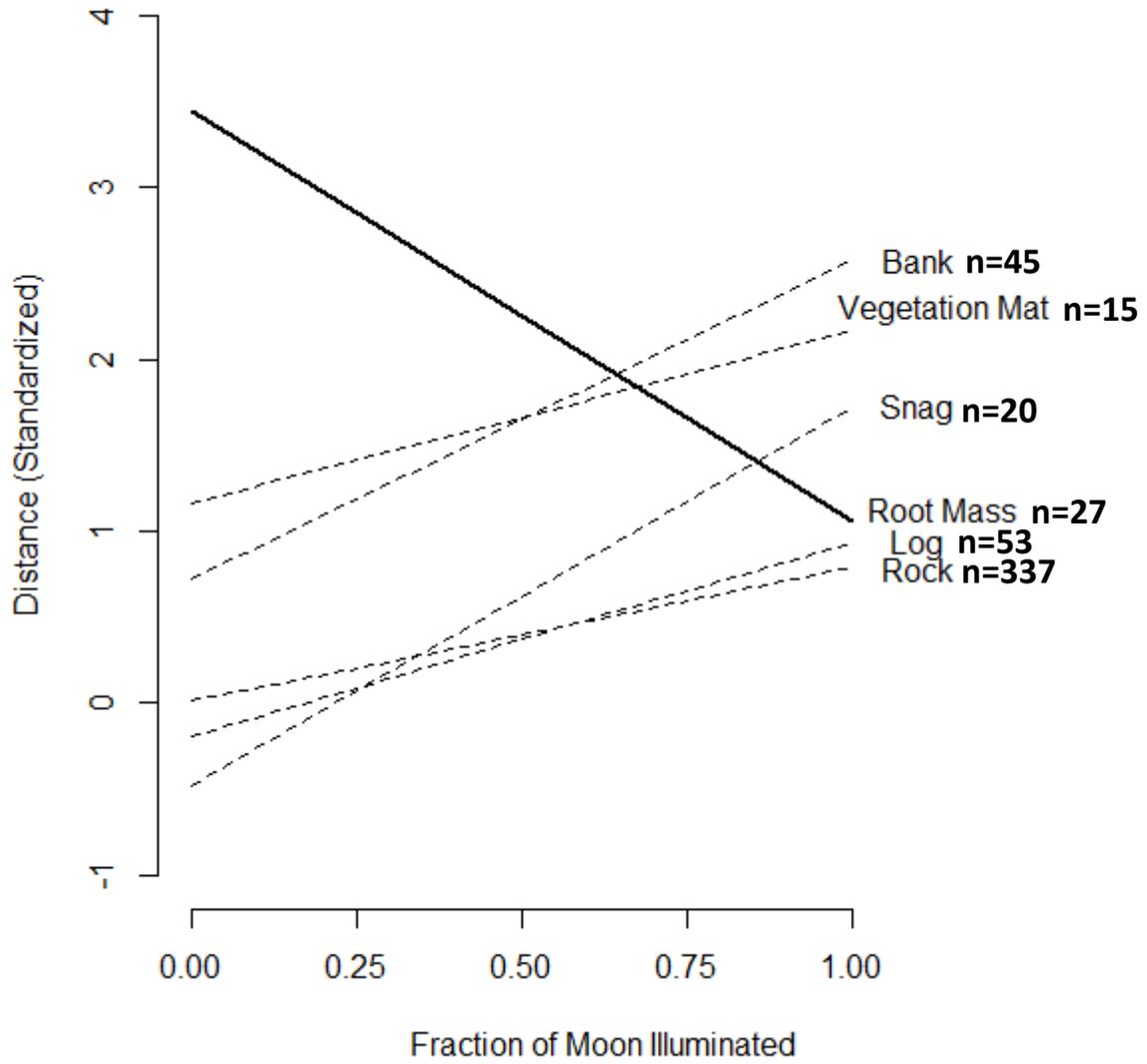


Figure 11. Barplot of average moon phase during periods of animal movement and no movement recorded between 18 June 2013 and 12 October 2013 with standard error bars and number of observations.

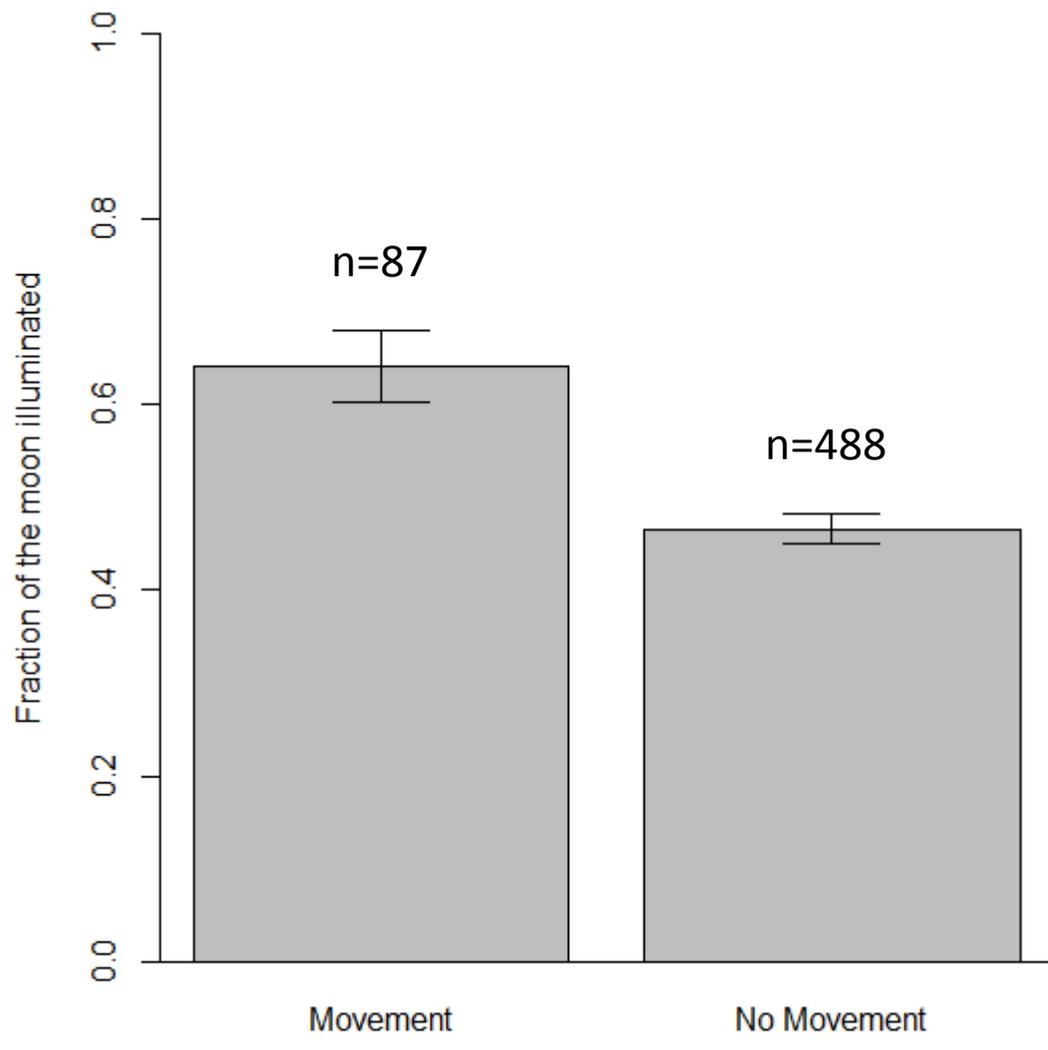


Figure 12. Stripchart of cumulative number of movements (A), cumulative movement in meters (B), and number of days located (C) by release location recorded between 18 June 2013 and 12 October 2013 with standard error bars.

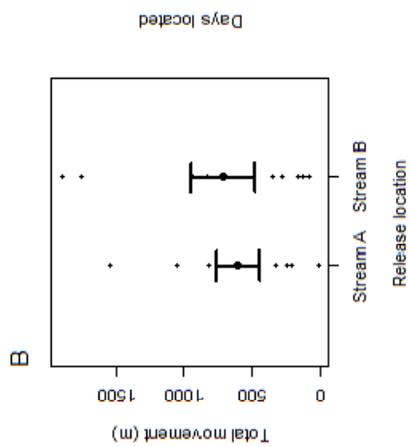


Figure 13. Stripchart of cumulative number of movements (A), cumulative movement in meters (B), and number of days located (C) by release type recorded between 18 June 2013 and 12 October 2013 with standard error bars.

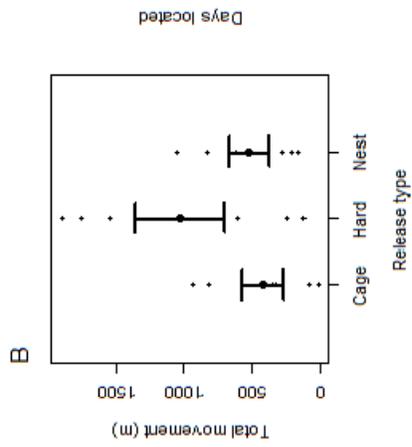
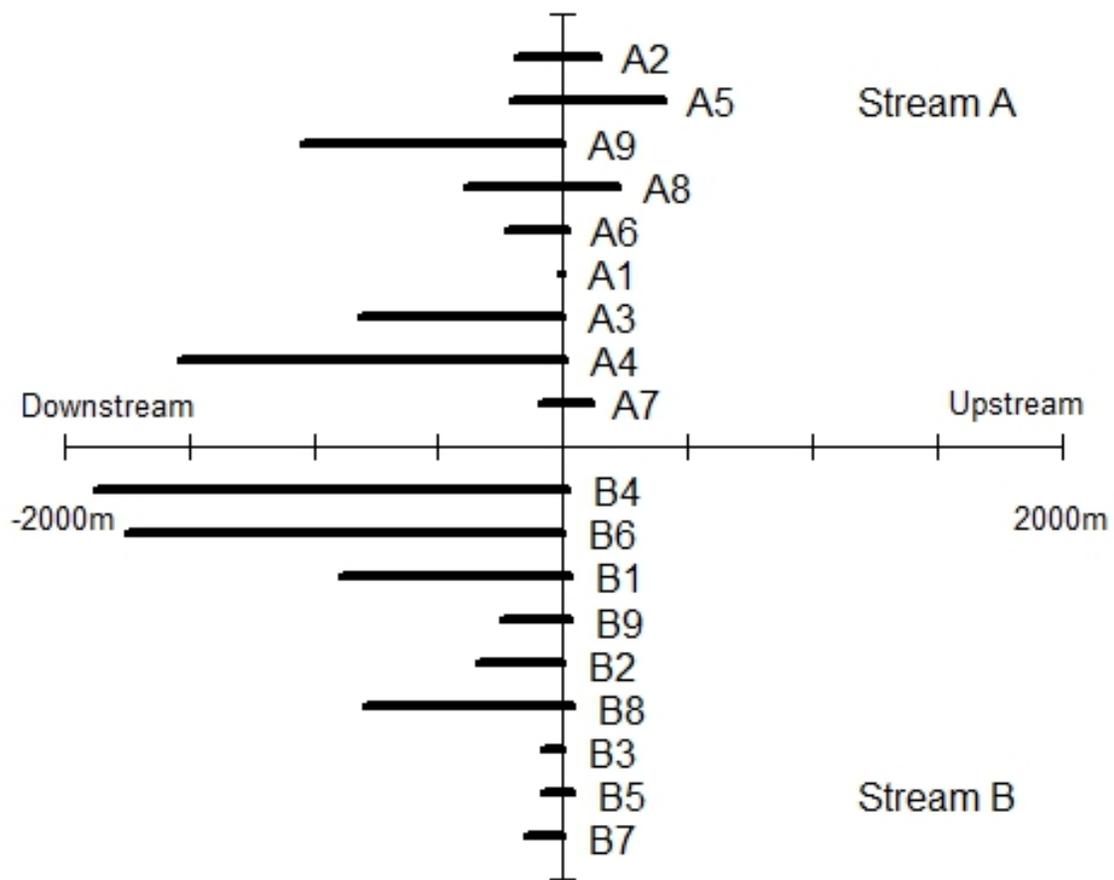


Figure 14. Cumulative dispersal distance of individual animals in meters upstream (positive) and downstream (negative) recorded between 18 June 2013 and 12 October 2013 arranged by stream location and number of days located (greatest to fewest days located from top to bottom in each stream).



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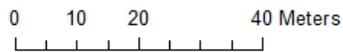
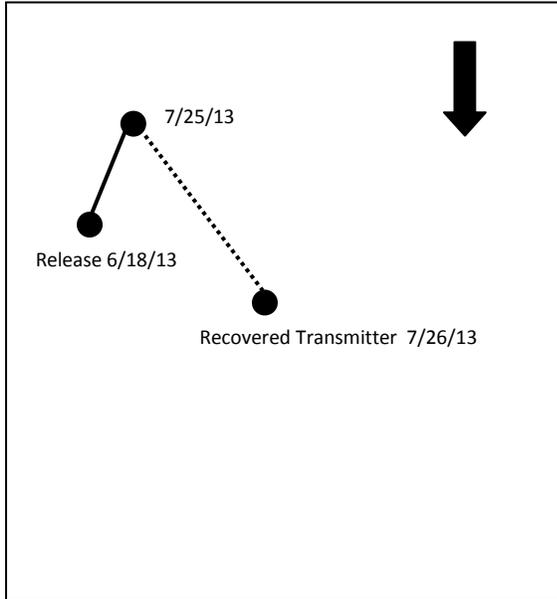
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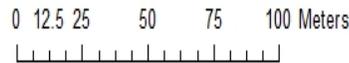
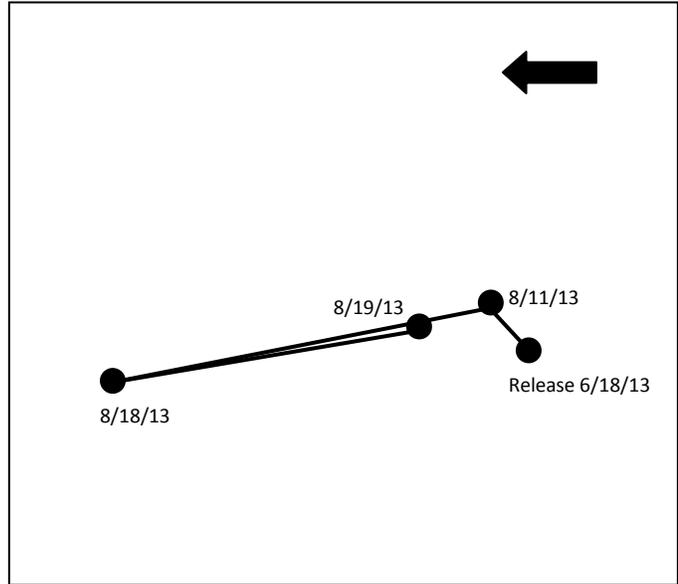
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**Appendix I – Maps of each individual animal's movements in Stream A. Solid lines indicate animal movement. Dotted lines indicate movements left out of analysis (i.e. moved by a predator, re-release, etc.). Arrows indicate direction of stream flow.**

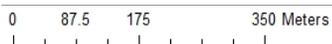
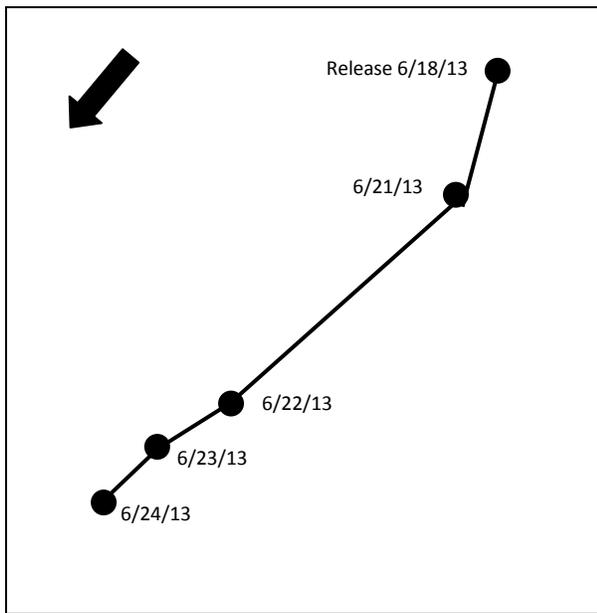
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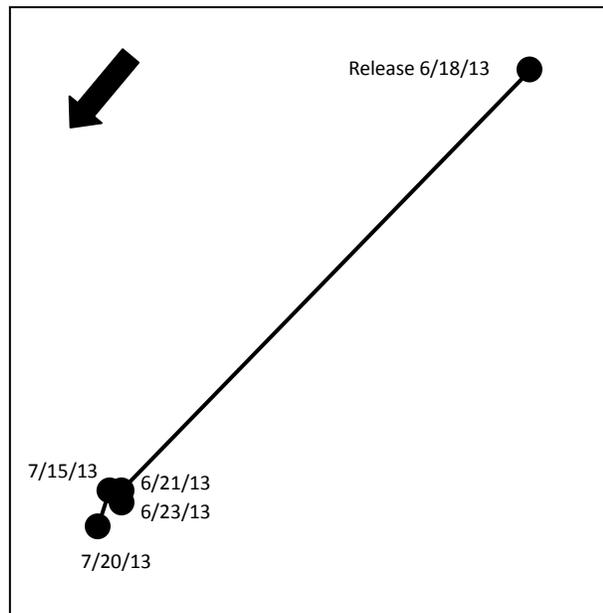
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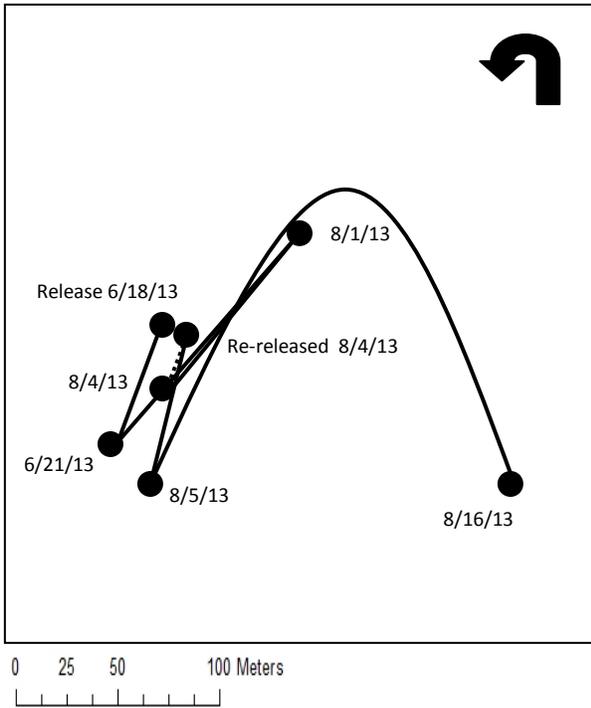
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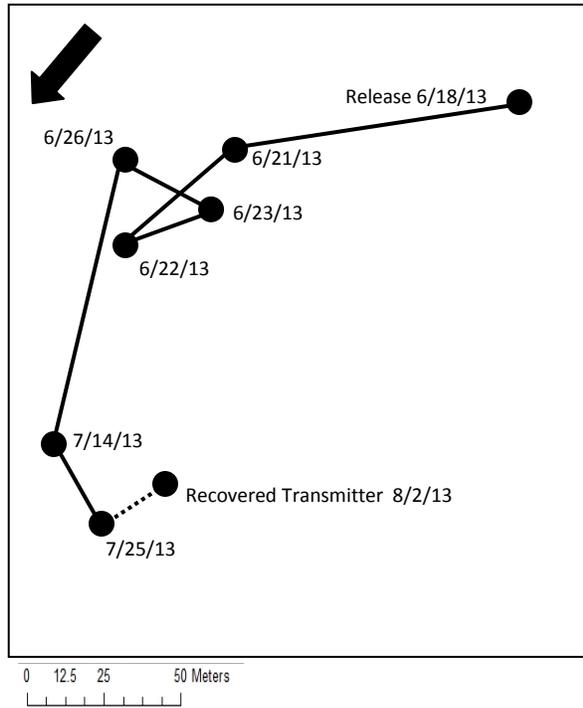
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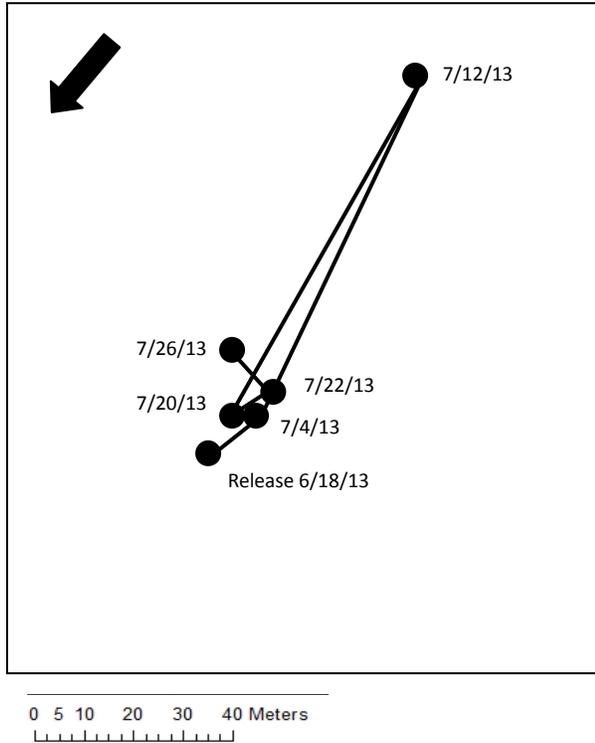
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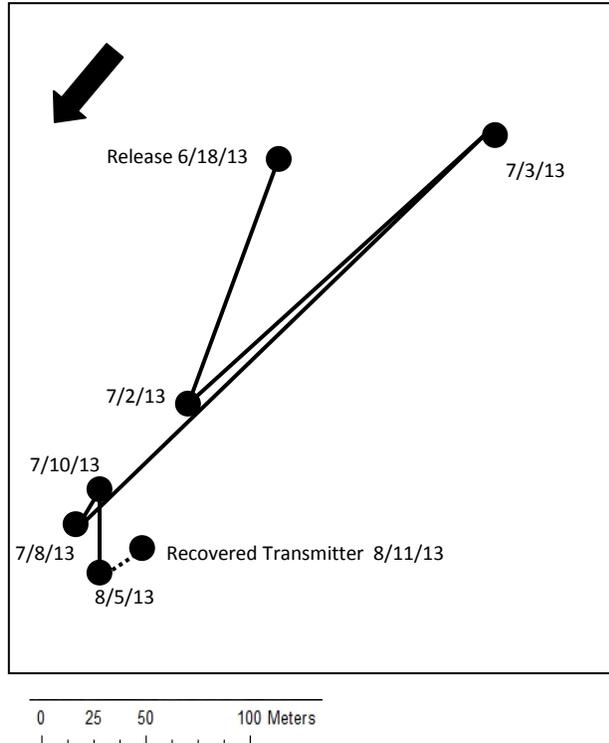
**Animal A6**



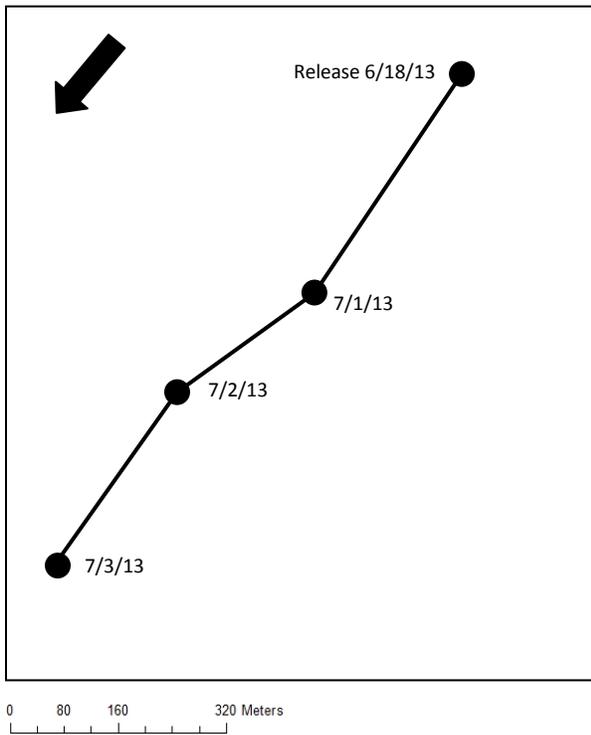
**Animal A7**



**Animal A8**

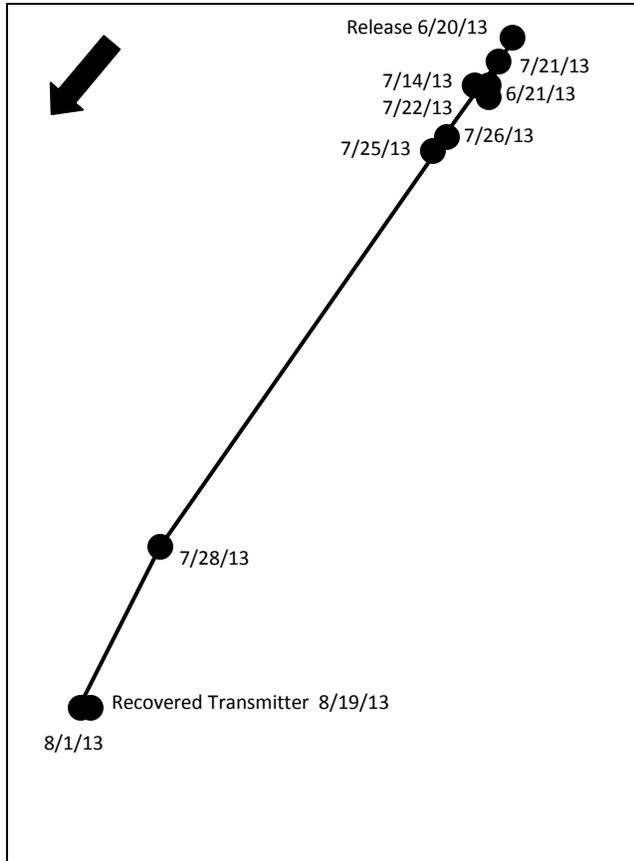


### Animal A9

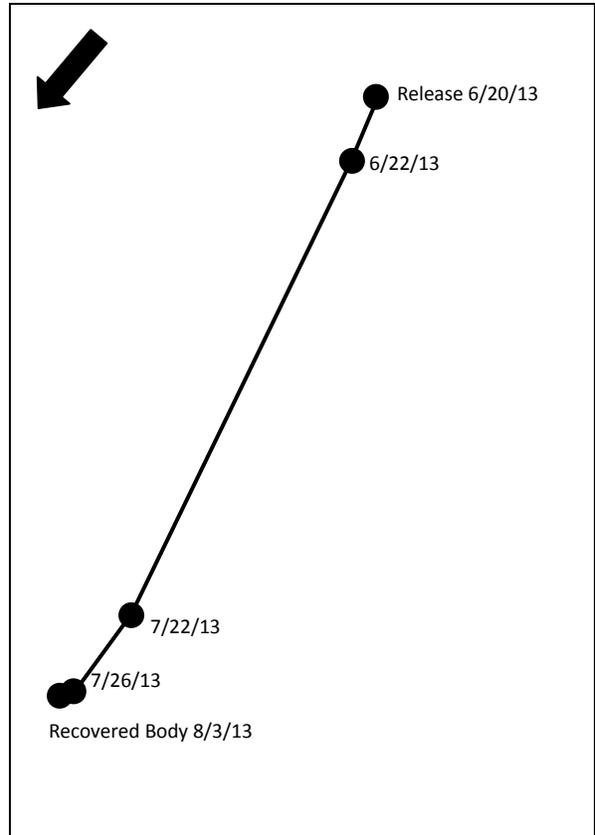


**Appendix II – Maps of each individual animal's movements in Stream B. Solid lines indicate animal movement. Dotted lines indicate movements left out of analysis (i.e. moved by a predator, re-release, etc.). Arrows indicate direction of stream flow.**

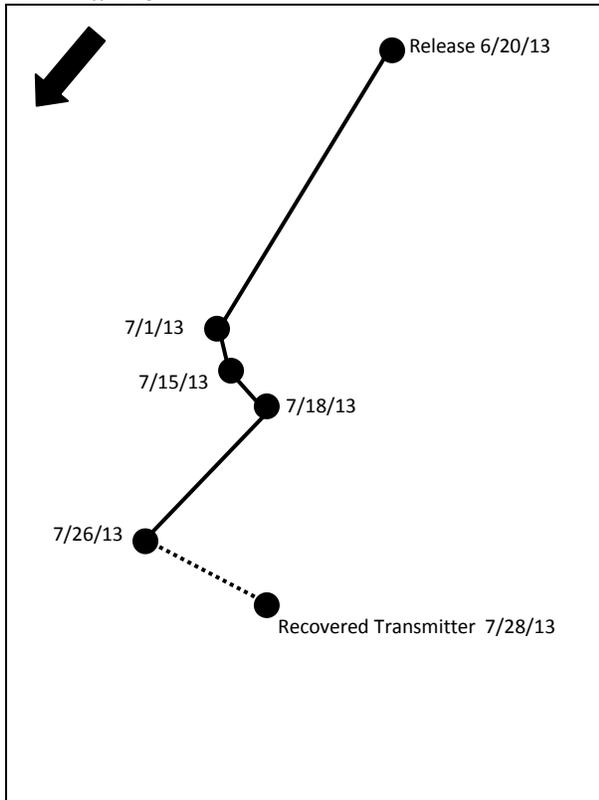
**Animal B1**



**Animal B2**

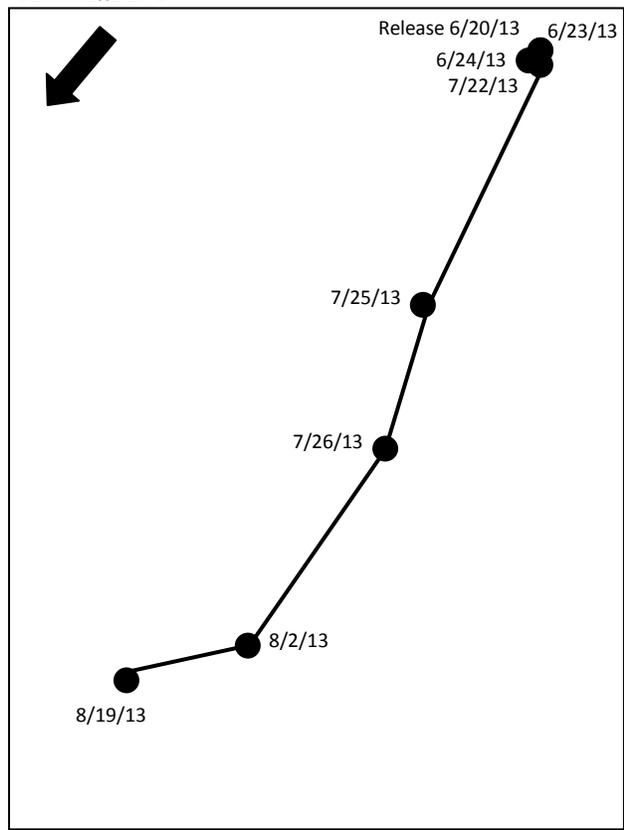


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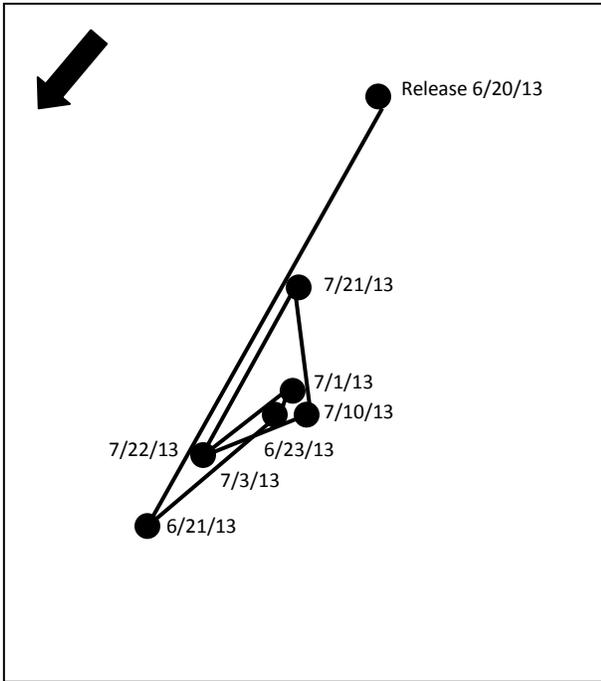
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**Animal B4**



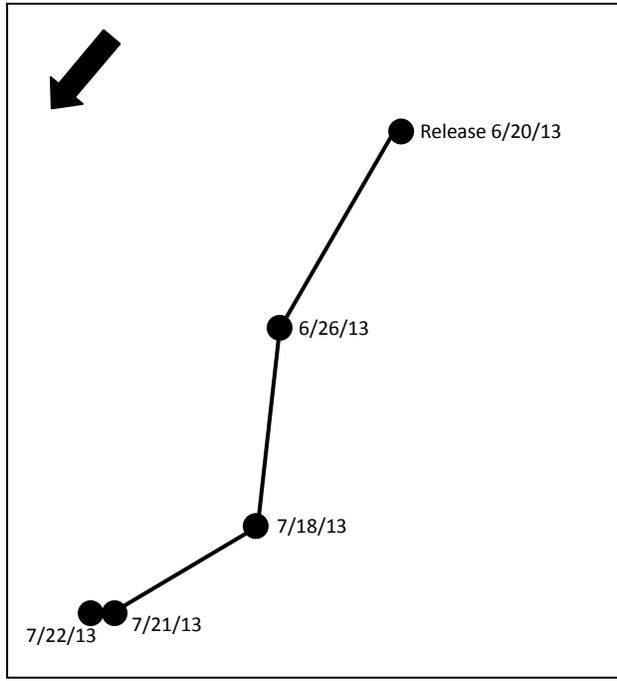
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**Animal B5**



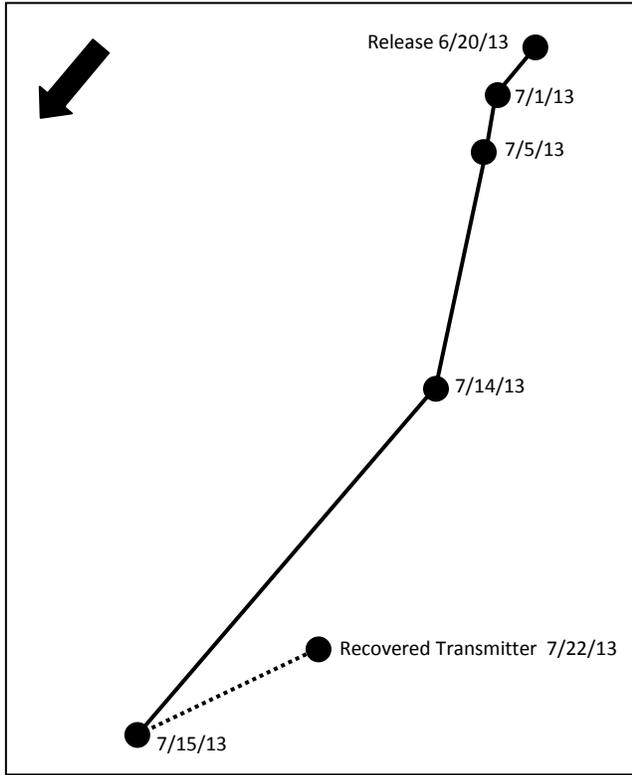
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**Animal B6**



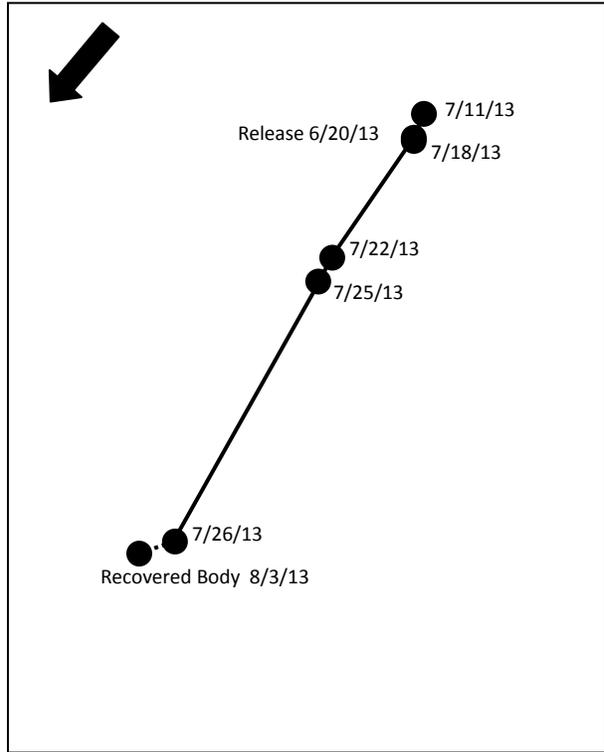
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**Animal B7**



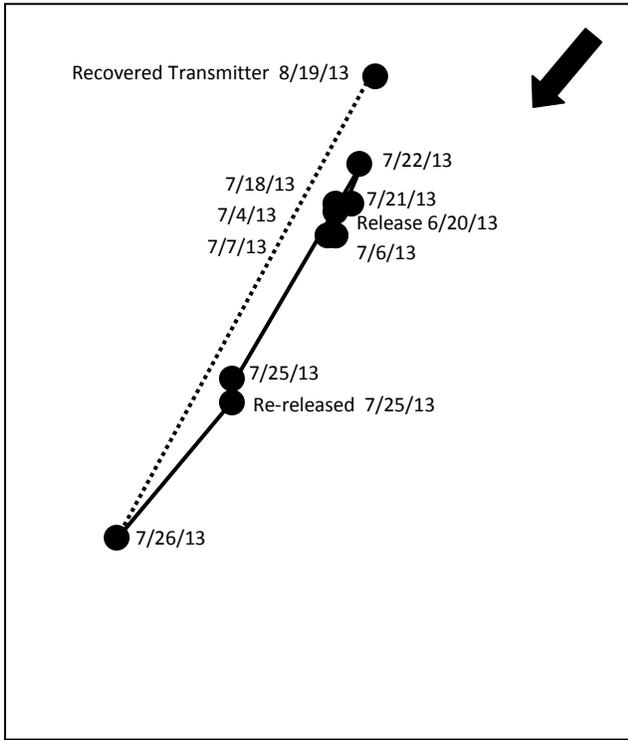
0 5 10 20 Meters

**Animal B8**



0 130 260 520 Meters

# Animal B9



0 40 80 160 Meters

**Appendix III- Definitions and examples of cover objects used by study animals between 18 June, 2013 and 12 October, 2013. a – log, b – snag, c – root mass, d – vegetation mat, e – bank, f – rock**

<b>Cover Type:</b>	<b>Definition:</b>
<b>Log</b>	a log within the stream with no other debris around it
<b>Snag</b>	a pile of various debris within the stream
<b>Root mass</b>	a tangle of roots from a living tree growing at the edge of the stream
<b>Vegetation mat</b>	a dense covering of living plants usually growing at the edge of the stream
<b>Stream bank</b>	an opening either in or beneath the bank of the stream



**Appendix IV – Images of visible animals. a-b – Animal A4 (head visible – 15 July, 2013), c-d – Animal A9 (tail visible – 2 July, 2013)**



**Appendix V – Evidence of study animal death/transmitter recovery recorded between 18 June, 2013 and 12 October, 2013. a-e – transmitter recovery locations, f – example of raccoon tracks found near multiple transmitter recovery locations.**



**Appendix VI – Study animal carcass recovery. a – Animal B1 remains recovered on stream bank on 19 August, 2013, b – Animal B2 remains recovered on stream bank on 3 August, 2013, c-f – Animal B8 body recovered in shallow pool on 3 August, 2013**



**Appendix VII – Animal A5 found out of water on 4 August, 2013. a – location where animal was found, b – condition of animal (reddened toes can be seen), c – reddened toes and cloacal slit, d – reddened toes and gill slit.**

