# Work smarter, not harder: Comparison of visual and supplemental survey methods for the eastern massasauga rattlesnake

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Abstract:

Monitoring programs of rare and endangered species are of greatest conservation need. Herpetofuana are a taxa characterized by low detection rates and require the best survey methods for effective monitoring. The eastern massasauga rattlesnake is declining in every state or province that it is found and would benefit from better survey techniques. In the past, studies concerned with population demography have relied on intensive visual mark-recapture survey methods. Other common snake capture techniques have seen little use in eastern massasauga rattlesnake population studies. We explored the effectiveness of using artificial cover objects and funnel traps in supplementing visual survey methods. Funnel traps were about 3 times as efficient as visual surveys ($p = 0.0009$), and when capturing only males were about 4 times as efficient ($p = 0.0036$). Wooden coverboards were about twice as efficient as visual surveys when capturing females ($p = 0.0286$). In addition to massasauga surveys: carpets, coverboards, and funnel traps were more efficient at assessing the diversity of a site ($p = 0.0019$, 0.0011, 0.0002 respectively). We suggest the use of these supplemental techniques would provide for more robust data sets that will yield more powerful results.

Introduction:

Monitoring populations of rare and endangered species has become a priority for many management and conservation agencies (Guisan et al. 2006). However many species of plants and animals, particularly rare and endangered species, have detection probabilities of less than one (McArdle 1990, Parris et al. 1999, Kery 2002 and 2003, Slade et al. 2003, Tyre et al. 2003, Bailey et al. 2004, Wintle et al. 2004 and 2005, de Solla et al. 2005). Low detection rates of rare or endangered species can be the result of many factors including, inherently imperfect detection methods (McArdle 1990), small population sizes (Morse et al. 1988), operator error, random chance, misidentification and difficult conditions such as dense vegetation or weather (Wintle et al 2005). Also animal behavior and efficiency of survey method can reduce detection probabilities (Gu and Swihart 2004). In addition, adequate detection can require large amounts of time and money (Mackenzie et al. 2002), which land managers might simply not have. Because of low detection in these species more efficient and cost effective methods could be highly beneficial to conservation agencies.

One taxon that is characterized by low detection probabilities is herpetofauna. Cryptic behavior, camouflaging color patterns, and generally short active seasons make detection of individuals a difficult task (Mazerolle et al. 2007). These characteristics can make estimates of population size and vital rates inaccurate or hard to obtain (Mazerolle et al. 2007, Gu and Swihart 2004). Typical sampling methods for reptiles and amphibians include drift fences in combination with pitfall or funnel traps (Fogarty and Jones 2003, Ribeiro-Júnior et al. 2008), use of artificial cover objects (Crosswhite et al. 1999), auditory call surveys (Crouch and Paton 2002), night driving (Parris 1999), and visual encounter surveys (Karns 1986, Heyer et al. 1994). Factors influencing a choice of one method over another include habitat type (Doan 2003), behavior of animals (Crosswhite et al. 1999), and simply what is known to work (Karns 1986, Heyer et al. 1994). Depending on the data needed (e.g., one species vs. all species, males vs. females), the efficiency of the method is extremely important (Fogarty and Jones 2003).For these reasons comparison of survey methods for individual species is of the utmost importance if researchers and land managers are to effectively monitor populations.

The eastern massasauga rattlesnake (*Sistrurus catenatus catenatus*) is the focal species for this study. Eastern massasauga rattlesnakes (EMR) have declined across their entire range (Szymanski 1998, Johnson et al. 2000), and have recently been proposed for threatened listing under the U.S. Endangered Species Act. Because they are ambush predators, massasaugas are reclusive, largely sedentary, and cryptically colored, making detection extremely challenging (Parker and Plummer 1987). This species is typically found in wetland habitats from spring to mid-summer and upland meadows in latesummer (Bailey et al. 2011). Snakes return to wetlands before winter in search of hibernacula that typically consist of crayfish and small mammal burrows (Harding and Holman 2006). Extant populations of eastern massasaugas are found in small isolated patches of suitable habitat surrounded by heavily modified landscapes (Szymanski 1998). For many remaining EMR population, dynamics and long term viability are uncertain. This uncertainty is in large part due to low EMR detection rates, which makes mark-recapture studies time and effort intensive (Parker and Plummer 1987). In the past, the general survey method used in population studies has been mark-recapture based on visual encounter surveys (Casper et al. 2001). However, a comparison on a per effort basis for different survey techniques is lacking for the eastern massasauga rattlesnake. Techniques such as funnel traps in combination with drift fences and use of artificial cover objects (e.g. carpet square and wooden boards) can be effective at capturing massassaugas, yet the efficiency and individuals capture rates for the different techniques have never been directly compared (Casper et al. 2001). Comprehensive data comparing capture success on a per effort basis could warrant the use of these supplemental techniques as standard survey protocol for EMRs (Casper et al. 2001). Here we explore the use of artificial cover objects, and drift fences in combination with funnel traps in supplementing visual surveys by comparing capture rates (snakes/hour) between the various survey methods. Our objective was to identify the most efficient survey method, to improve detection efforts, and enhance population surveys. We hypothesize that supplemental survey methods will have a higher capture rate (i.e. snakes per hour) and higher species per hour than visual encounter surveys.

Study Site:

Our study was conducted at Pierce Cedar Creek Institute (PCCI) in Barry County, MI. Pierce Cedar Creek Institute is a 267 ha preserve, approximately 40% of which is classified as wetland. The study area encompassed approximately 19.9 ha composed primarily of prairie fen, wet meadow, and nearby upland prairie and old field plant communities surrounding Brewster Lake, Cedar Creek and adjoining tributaries (Figure 1). Eight snake species are known to inhabit PCCI including eastern garter snakes (*Thamnophis sirtalis*), northern ribbon snakes (*Thamnophis sauritus septentrionalis*), eastern milk snakes (*Lampropeltis triangulum triangulum*), eastern hog-nosed snakes (*Heterodon platirhinos*), northern water snakes (*Nerodia sipedon*), blue racers (*Coluber constrictor foxii*), brown snakes (*Storeria dekayi*), and eastern massasauga rattlesnakes (*Sistrurus catenatus*).

Materials & Methods:

In May 2015, we assembled four trapping arrays throughout the wetland where massasaugas are known to occur. Arrays were comprised of one drift fence, two funnel traps, seven carpet squares and seven coverboards. Drift fences were composed of pre-fabricated 0.9 m tall x 30.5 m long silt fencing with attached wooden posts. Fences were buried approximately 15 cm into the soil and a funnel trap was placed at each end to capture snakes. Funnel traps were constructed using hardware cloth and aluminum screening (see figure 2). All cover objects (i.e., carpet squares and wood boards) were 0.6 x 1.2 m. Wood cover boards were ¼ inch thick plywood. Alternating cover objects were placed along each side of the drift fences 10 m from the fence and 5 m apart (Figure 1). Drift fences were placed in areas of large continuous patches of habitat (Karns 1986). We sampled traps for 42 days between 21 May and 14 August. We recorded the number and species of snakes found using each trap. Trap effort was determined by averaging the time it took each of three researchers to check each individual trap type at each trapping array. Number of snakes encountered and trap effort were used to calculate catch-per-unit-effort.

In order to compare survey methods visual surveys were also conducted in the same areas that traps were placed (see figure 1). Surveys were generally conducted between 0900 h and 1200 h and/or between 1300 h and 1600 h. Each surveyor recorded their search effort (i.e. total time spent actively looking for snakes). Effort data were used to calculate catch-per unit effort (snakes/hour). Capture locations were recorded using handheld Garmin GPS units. Encountered snakes were captured and handled in compliance with the GVSU IACUC (permit # 13-02-A) following the Guidelines for Use of Live Amphibians and Reptiles in Field and Laboratory Research.

All captured EMRs were sexed by probing for the presence of hemipenes. Massasaugas were marked permanently by injecting a subdermal passive integrated transponder (PIT) tag, and temporarily by applying colored nail polish on rattle segments. After processing, snakes were released at their capture site on the same day of capture. All captured massasaugas were documented with photos. We sanitized lab equipment according to disinfection protocols recommended by Pierce Cedar Creek Institute to prevent the spread of Snake Fungal Disease (i.e. *Ophidiomyces ophiodiicola*). Other snake species were not marked, so we report the number of species captured using each method only.

Data Analysis:

Snake capture data were measured in snakes/hour so that all methods were comparable. Captured snakes were not marked as unique and recaptured snakes were used in data analysis. Capture data was pooled into weekly survey periods to ensure that all survey units and trapping arrays were included for each survey period. This resulted in 12 survey periods. Data were tested for normality using Shapiro-Wilk tests. Data were non-normal and contained count variables, therefore we used Poisson regressions (PROC GLIMMIX, SAS v. 9.4; Carruthers et al. 2008) to test for differences in capture efficiency between the capture methods. We used Pearson $\chi^2$ to test for dispersion and validate the use of Poisson regression. All Pearson $\chi^2$ values were less than 1.5, illustrating that Poisson regression is an appropriate analysis for our count data (Carruthers et al. 2008). All *Thamnophis* species were pooled because these snakes could not always be identified to species. Our dependent variables were the number of snakes/hour, with separate analyses for male EMR, female EMR, total EMR, and the total number of species/hour (i.e., species richness). Independent variables were capture type (carpet, board, funnel, visual) and week was included as a random effect. Visual surveys were used as the standard and all other methods were compared to visual surveys. All analyses were performed in SAS v. 9.4. Significance was assumed at $p<0.05$.

Results:

For EMRs, drift fences with funnel traps had the highest catch per unit effort with 34 snakes/hour, followed by coverboards (15 snakes/hour), visual surveys (5 snakes/hour), and carpets (1 snakes/hour) (Table 1). The funnel trap captures were biased toward males, which represented 90% of the captures (n=10). Conversely, board captures were biased toward female EMRs, with 100% of board captures (n=8) being females. Visual encounter surveys were also biased toward females, with 80% of captures (n=83) being females. We only captured one EMR (a male) using carpets (Table 1). For total EMR analyses, only funnels found significantly more snakes/hour than visual surveys (β = 1.74, p=0.0009, Table 2), and this result was mostly driven by male captures as funnels also capture significantly more male EMRs than visual surveys (β = 3.09, p=0.0036). Boards captured significantly more female EMRs/hour than visual surveys (β = 1.29, p=0.029). Board (β = 2.74, p=0.0011), carpet (β =2.60, p=0.0019), and funnel survey methods (β = 3.22, p=0.0002) all had significantly higher numbers of snake species captured, when compared to visual surveys. Boards were the only technique that captured milk snakes (0.28 snakes/hour), funnels; blue racers (0.26 snakes/hour), visual; hog-nosed (0.005 snakes/hour), and carpets did not capture any unique species. All trapping techniques were more effective at capturing *Thamnolphis* (approximately 5 snakes/hour) while visual found less than one snake/hour. Carpets and boards captured roughly 2 brown snakes/hour while visual found .009 brown snakes/hour and funnels found none. Northern water snakes were only captured by funnel and visual methods but at rates of less than one snake/hour.

Table 1: Total counts for various eastern massasauga rattlesnake survey methods. Effort is measured in hours surveyed.

| Survey method | Total EMR (n) | Male EMR (n) | Female EMR (n) | Total number of species | Total effort (hours) |
|---|---|---|---|---|---|
| Carpet | 1 | 1 | 0 | 3 | 8.4 |
| Board | 8 | 0 | 8 | 4 | 7.2 |
| Funnel | 10 | 9 | 1 | 4 | 3.8 |
| Visual | 83 | 17 | 66 | 5 | 203.4 |

Table 2: P-values for poisson regression comparing trapping methods to visual surveys. Values listed with an * are significant at p<0.05.

| Analyses Type | Board | Carpet | Funnel |
|---|---|---|---|
| Total EMR | 0.0970 | 0.1262 | 0.0009* |
| Male EMR | 0.9957 | 0.9155 | 0.0036* |
| Female EMR | 0.0286* | 0.9925 | 0.6405 |
| Species Richness | 0.0011* | 0.0019* | 0.0002* |

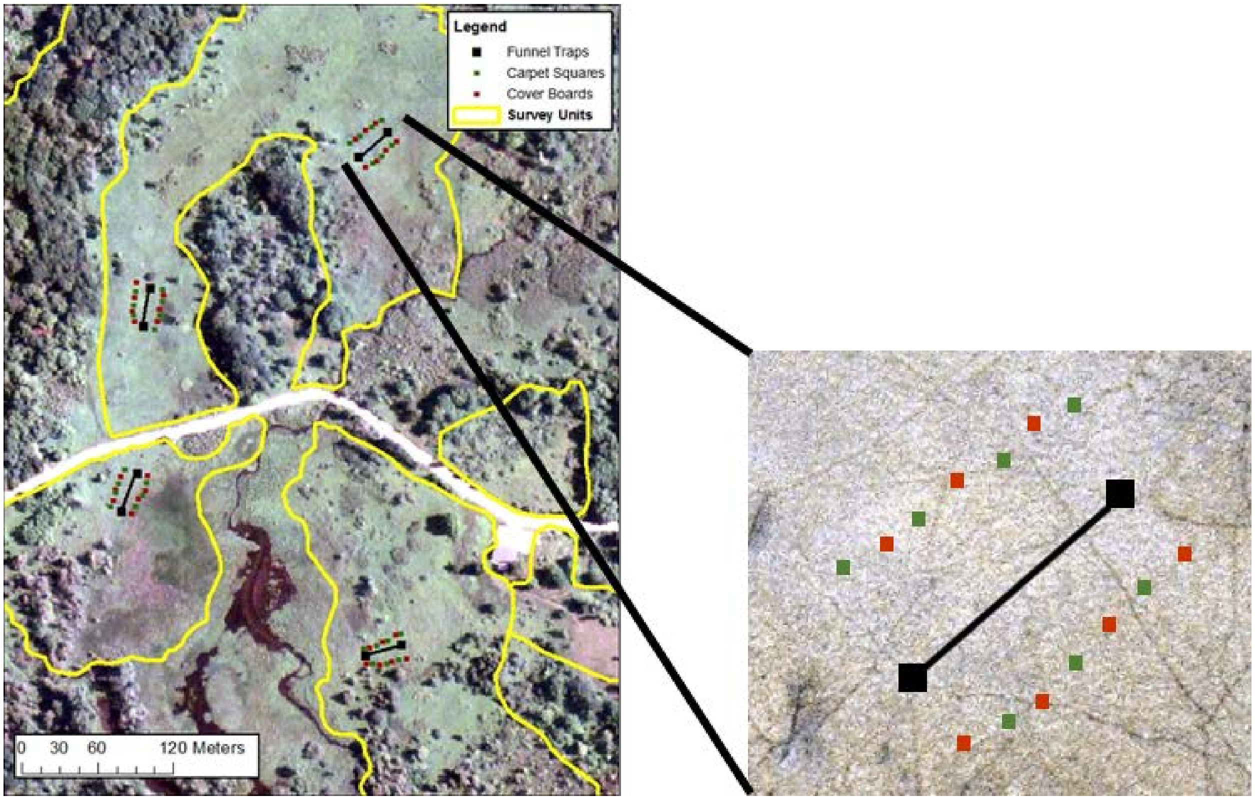


Figure 1: Map of study site at Pierce Cedar Creek Institute located in Hastings, Michigan. Areas outlined in yellow are survey units. Not all survey units were used for comparison of supplemental survey methods. Green squares indicate carpet squares, red squares indicate coverboards, black squares indicated funnel traps, and black lines indicate drift fences.

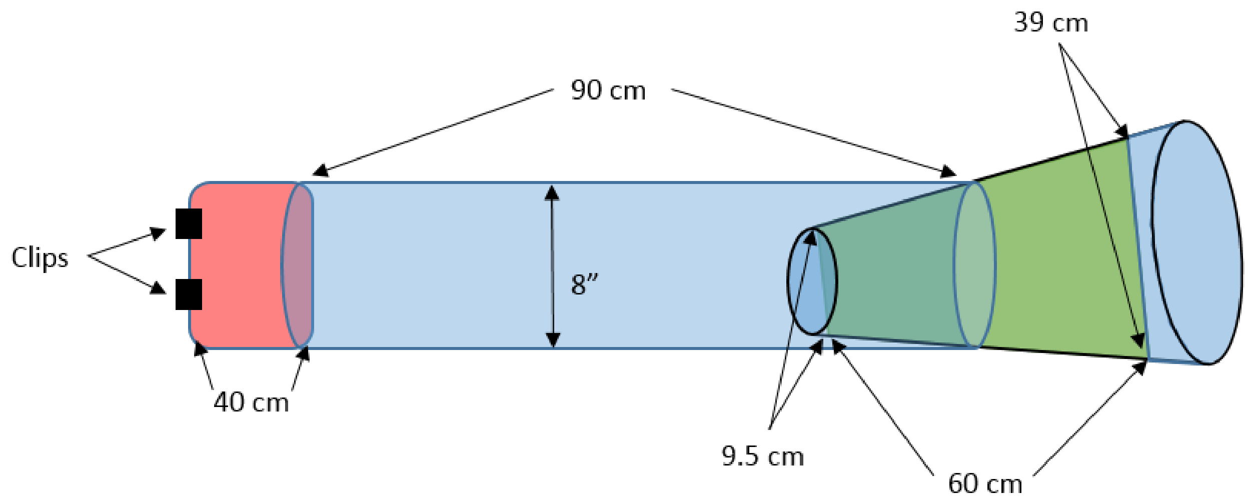


Figure 2: Schematic of funnel trap design. The main cylinder was made from two 90 cm long rectangles of aluminum fence material cut so that they would make an 8" diameter tube. Each tube had an interior made from a fine hardware cloth (1/4 inch by ¼ inch aluminum mesh) that would keep animals from getting stuck and an exterior made from a sturdier, coarser aluminum wire mesh material to provide structure. The main cylinder was fashioned together with zip ties. The green trapezoid depicts a wooden board 5.71 mm thick that is in the middle of the funnel. This guides the animals from the drift fence into either side of the funnel. The funnel was made from 75 x 90 cm sheet of aluminum fence material rolled into a funnel shape held together by zip ties and stapled to the wooden board. The entire funnel piece was then zip tied to the cylinder part of the trap. The red section on the left was made from hardware cloth that was 40 cm long and cut so that it would be 8" in diameter. It was made into a tube and attached to the main cylinder with zip ties. The left end was left opened. To close (i.e., "set") the trap you roll up the open end and secure it shut with binder clips. Spray foam was utilized to fill in any gaps in the funnel trap after construction.

Discussion:

The results of our study showcase the potential use of supplemental survey methods (e.g., drift fences with funnel traps) as standard protocol for eastern massasauga rattlesnake monitoring and illustrates the efficiency of passive and active traps for sampling snakes. Capture methods appear to target different sexes. Funnel traps were more efficient at capturing male rattlesnakes and coverboards were more efficient at capturing female rattlesnakes. However, carpet squares were ineffective as a survey method. All survey methods were more efficient at capturing a greater species diversity than visual surveys. Supplemental survey methods provide valuable, quick, and easy to obtain data when compared to visual surveys.  Our results do not discredit the use of visual surveys. Visual survey methods are cheaper and yield far more data than various methods but supplemental techniques used in tandem with visual surveys provide for the most useful data sets.

Drift fences likely passively sample a larger area than boards or carpets, so any snakes actively moving through an area with a drift fence will be captured. Male eastern massasauga rattlesnakes have a greater daily distance moved than females from May to August due to intensive mate searching behavior (Gillingham 1987). This phenomenon of male behavior could explain the high capture rates observed for males with funnel traps. Mating behavior in this species typically occurs from July to September peaking in late August (Jellen et al. 2007). The first male EMRs we captured occurred in mid-July and continued for the rest of the survey period which would support this hypothesis. By the same reasoning, we expect that this is why we did not find females often with this technique. With funnel traps we found approximately half the number of male snakes as visual surveys while only spending about 1% of the time we spent performing visual surveys (Table 1). Overall capture rates for funnel traps are significantly higher than visual surveys although male captures seem to be driving this result. We recommend the use of drift fences and funnel traps if male captures, or even sex ratios, are being sought.

The different artificial cover objects used in this study, carpets and coverboards, had different results when compared to visual surveys. Coverboards had significantly higher capture rates than visual surveys while carpets did not. All females snakes found with artificial cover objects were gravid, andmost of these females were found on top of the cover object basking. Cover objects were rarely used as refugia for rattlesnakes. Gravid females have an affinity for open basking sites that promote embryological development (Graves and Duvall 1993). Our data suggest that coverboards, rather than providing refugia, actually provide basking sites for female EMRs, and that coverboards should be preferred over carpets when specifically targeting eastern massasaugas.

Our results agree with other studies aimed at determining the effectiveness of various survey methods (Crosswhite et al. 1999, Fogarty and Jones 2003). Crosswhite et al. (1999) found that traps associated with drift fences (i.e. funnel traps) were most efficient at capturing snakes. Both studies agreed that using comprehensive survey methods that include visual and trapping techniques is the best way to sample a herpetofaunal community. Trapping techniques are far more efficient in terms of person hours than visual surveys but techniques differed in the species that were able to be captured. For example, we found eastern hognose snakes (*H. platirhinos*) using visual surveys but not cover boards; however, coverboards were effective in capturing eastern milksnakes (*L. triangulum triangulum*) while visual surveys were not. Other research has found this to be true also (Ribeiro-Júnior et al. 2008). Varied defensive and foraging behavior between snakes observed in this study could explain these results. For instance, eastern massasauga rattlesnakes are ambush predators that tend to rely on crypsis and envenomation for foraging and defense while other species (e.g., blue racers, *C. constrictor foxii*) tend to be active foragers that may evade surveyors when approached (Prior and Weatherhead 1994). This results in rattlesnakes being relatively easy to capture via visual surveys compared to more active species. Also other snakes were observed using artificial cover objects as refugia while eastern massasaugas tended to use them as basking sites  Because of these reasons we suggest the use of all methods (visual, carpets, coverboards, and funnels) when assessing the diversity of a site.

Our study highlights the benefits of supplemental survey methods and how they can augment monitoring and mark-recapture data sets. Drift fences with funnel traps are efficient at capturing males while coverboards are efficient at capturing females. We suggest using these supplemental survey methods in addition to any eastern massasauga rattlesnake visual mark-recapture studies. Placement of artificial cover objects and funnel traps are at the land manager's discretion. Specific trapping arrays used in this study were for experimental design and comparison purposes. It is important to note that visual surveys are the only method that do not require extensive habitat modifications (e.g. digging trenches for fences). One factor that we did not take into account with our analyses is the construction and setup time associated with methods other than visual surveys. In total, assembly of sampling arrays took 4 people 19.6 hours to complete, which mostly consisted of digging in drift fences and making funnel traps. This was not accounted for in the effort for each trap type. However, the construction of trapping arrays does not need to occur every season. Materials do tend to break down over multiple years, yet drift fences, for example, can be left over winter and patched at the beginning of the next season and funnel traps can be re-used. We suggest if using these methods to plan for a survey season that is at least similar to the length of this study i.e. 12 weeks. Another point to note is that all members of the survey team in this study are very experienced at surveying herpetofuana. This has a significant effect on the effectiveness of visual surveys (Heyer et al. 1994, Bailey et al. 2004a). Because of this effect, inexperienced surveyors could benefit from using passive and active trapping techniques because experience does not determine the effectiveness of these methods (Ribeiro-Júnior et al. 2008).

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