



Research Article

Survey and Analysis Design for Wood Turtle Population Monitoring

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ABSTRACT Population monitoring is a fundamental component of wildlife management, and is necessary to track site- and regional-level status and recovery of species of conservation concern. The wood turtle (*Glyptemys insculpta*) is a species of conservation concern for federal and state agencies because of population declines across the species' range. We developed and tested a survey and analysis design to assist agencies in the Upper Midwest, USA, with establishment of long-term monitoring programs for wood turtle populations. In spring of 2016, we conducted 8 replicate population surveys at 8 candidate long-term monitoring sites in northeastern Minnesota, USA. Using field survey data and simulation models, we assessed the influence of distance from river surveyed, number of survey replications, and number of sites on abundance estimates; we also delineated important survey covariates and compared demographic estimates based on distance from river surveyed. We estimated site-level abundances and compared survey designs using a multinomial N -mixture model that included a removal sampling observation process. Mean abundance estimates were similar when surveying 2 transects (i.e., the river-land interface to ~ 25 m inland) or 4 transects (i.e., the river-land interface to ~ 55 m inland), but decreasing the survey distance from river reduced the precision of estimates. Mean abundance estimates were similar with ≥ 6 replications. Air temperature was an important predictor of survey-specific detection probability, with maximum detectability at 19–23°C. Sex ratio and mean carapace length did not differ based on whether we surveyed 2 or 4 transects, and percentage of individuals by size class was nearly identical between the sampling designs. Simulations indicated that 75% of mean abundance estimates were within $\pm 8\%$ of true abundance when ≥ 15 sites were surveyed. The wood turtle survey and analysis design we developed and tested was effective for estimating abundance of wood turtle populations in northeastern Minnesota, and we encourage its use as a template for wood turtle monitoring programs in the Upper Midwest. © 2017 The Wildlife Society.

KEY WORDS abundance, *Glyptemys insculpta*, Midwest, Minnesota, N -mixture model, removal sampling, visual encounter survey.

Population monitoring programs are a fundamental component of wildlife management. These programs allow managers to track changes in distribution and abundance, improve our understanding of species-habitat relationships, and provide quantitative information on population responses to management actions (Gibbs et al. 1999, Campbell et al. 2002, Sauer and Knutson 2008). Optimally, monitoring programs should use standardized protocols that result in reliable site-level estimates of population parameters, while also being amenable to assessments of broad-scale trajectories in occupancy and abundance. Large monitoring programs in the United States with standardized

protocols include the North American Breeding Bird Survey (1966–present), North American Amphibian Monitoring Program (1997–2015), and North American Bat Monitoring Program (2015–present). These programs have resulted in important assessments of regional species and community trends (Adams et al. 2013, Bled et al. 2013, Weir et al. 2014).

Accurate and precise population estimates are particularly important for species of conservation concern, such as those listed in the United States as federally or state threatened or endangered. Thus, research has been devoted to development and improvement of monitoring programs for focal species (Flint and Harris 2005, Probst et al. 2005, Brown et al. 2013, Erb et al. 2015). However, many species of concern do not have standardized monitoring protocols because of limited funding for wildlife research, or the protocols are not amenable to contemporary analyses because of rapid advancements in statistical approaches for analyzing

Received: 8 December 2016; Accepted: 24 January 2017

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monitoring data, which have inherent sampling design requirements.

The wood turtle (*Glyptemys insculpta*) is a semi-aquatic freshwater turtle species endemic to northeastern North America that is of conservation concern for federal and state agencies because of population declines across the species' range (Garber and Burger 1995, Daigle and Jutras 2005, Willoughby et al. 2013). Wood turtles are currently considered endangered by the International Union for Conservation of Nature and Natural Resources (IUCN; van Dijk and Harding 2011), categorized as threatened by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC 2007), and are currently under review for listing under the United States Endangered Species Act (U.S. Fish and Wildlife Service 2016). In the United States, all but 2 states (Maryland and Pennsylvania) within the wood turtle's distribution list the species as endangered, threatened, or a species of concern (Jones et al. 2015).

The wood turtle is a riverine species, and individuals typically remain near flowing water throughout the year (Arvais et al. 2002, Brown et al. 2016). However, wood turtles are largely terrestrial from late spring to early fall (Kaufmann 1992, Niederberger and Seidel 1999, Compton et al. 2002). Therefore, unlike most freshwater turtle species, terrestrial-based survey designs could be effective for wood turtles. Previous researchers have used several techniques to survey wood turtle populations, including active searches by foot of upland, riparian, and stream habitat (Brooks et al. 1992, Daigle 1997, Greaves and Litzgus 2009), active searches by boat of shorelines and transparent streams (Buech et al. 1997, Daigle 1997, Saumure and Bider 1998), and passive aquatic traps (Ratner and Anderson 1978, Akre 2002).

Foscarini and Brooks (1997) proposed that standard survey, measurement, and analysis protocols be developed for wood turtles but provided few specific suggestions. To our knowledge, a study conducted in the eastern United States by Jones et al. (2015) represents the only work on development and evaluation of a standardized monitoring protocol for wood turtles. The principle characteristics of their survey protocol consist of active searches of an approximately 1-km stream segment by 1–4 individuals (1 individual considered the lead surveyor), with searches restricted to the water and ≤ 10 m from the stream edge, searches restricted to 1 hour (not including turtle processing time), and 6 replications completed annually (during spring or fall). Jones et al. (2015) estimated abundance at 196 sites in the northeastern United States using binomial N -mixture models with a zero-inflated Poisson distribution, and at 17 and 27 sites using open and closed population capture-mark-recapture (CMR) models, respectively. They concluded that the N -mixture model underestimated total abundance among the surveyed sites because of overestimated detection probabilities, and that CMR models were generally unable to estimate abundance unless ≥ 9 survey replications were completed. Jones et al. (2015) also concluded that detection probabilities in their study were influenced by season, weather, stream width, survey duration, and number of observers.

State agencies in the Upper Midwest (i.e., Minnesota, Wisconsin, Michigan, and Iowa) are currently interested in

establishing long-term monitoring programs for wood turtle populations to track population trends and responses to recent management actions (e.g., nest site creation and restoration). Although Jones et al. (2015) provided valuable information for assisting with development of a standardized monitoring protocol in the Upper Midwest, there is potentially room for improvement. Specifically, managers need a survey and analysis design that produces reliable abundance estimates across a wide range of population sizes, while maintaining reasonable survey effort and flexibility to accommodate changes in survey effort across years due to funding or personnel constraints. We developed and tested a survey and analysis design to assist agencies in the Upper Midwest with creation of wood turtle abundance monitoring programs. We assessed the importance of distance from river surveyed, number of survey replications, and number of sites on abundance estimates, and delineated important survey covariates. In addition, we estimated the number of survey replications necessary to obtain reliable abundance estimates for distribution monitoring.

STUDY AREA

We conducted this study at 8 sites in northeastern Minnesota that were located along a 17-km stretch of a river occupied by wood turtles (specific locations withheld in compliance with state of Minnesota data practices law). We identified the sites as candidate long-term monitoring sites because of their locations relative to recent management actions implemented by the Minnesota Department of Natural Resources (i.e., constructed road barriers, created and restored nest sites, predator exclusion from nest sites). Three sites either contained or were proximal to managed areas, and 5 served as control monitoring sites, to allow for tracking of long-term population-level effects of management actions. In addition, previous wood turtle surveys in the study area indicated that we would encounter a wide range of population sizes (Buech 1995), making this a useful study area to assess our survey and analysis design.

Each site consisted of a 380–560-m stretch of river ($\bar{x} = 486$ m) and adjacent riparian and upland habitat. We chose a target river survey length of 0.5 km based on a pilot study and previous research on individual movement patterns in the study area (Brown et al. 2016), which indicated that approximately 0.5 km provided a good balance between amount of time required to complete surveys and number of potential captures and recaptures. Using the 0.5-km stretch of river as a basis, we delineated the actual survey length at each site based on accessibility, location of managed areas, and a qualitative assessment of suitability for use by wood turtles.

More than 90% of the surrounding land was forested, with the remainder in non-forest and aquatic habitat classes. Approximately 75% of the area was in public ownership. The study area was dominated by mesic forest types, with common tree species including aspen (*Populus* spp.), balsam fir (*Abies balsamea*), and paper birch (*Betula papyrifera*). Pine (*Pinus* spp.) forest types occurred within sandy soils adjacent to some nest sites at river cutbanks. Hydric forest types were

also present, with common species including black spruce (*Picea mariana*), balsam fir, northern white cedar (*Thuja occidentalis*), and tamarack (*Larix laricina*). Nonforest cover types consisted of lowland alder (*Alnus* spp.), grass-forb openings, oxbow lakes, and other non-flowing water features. Additional habitat information in the study area can be found in Brown et al. (2016).

METHODS

Population Sampling

In 2015, we developed a preliminary population survey protocol based on a literature review and discussions with regional wood turtle biologists, and performed a pilot study (i.e., 1 survey completed at each site) to test the survey design. In 2016, we revised and implemented the survey protocol, completing 8 replicate surveys at each of the 8 long-term monitoring sites between 30 April and 5 June. This survey window corresponded to early spring in northern Minnesota, and allowed us to complete all surveys prior to full leaf-out. Time between replications ranged from 1–10 days ($\bar{x}=4$ days).

Our survey protocol consisted of visual encounter surveys by foot. We surveyed sites by walking 4 transects on each side of the river and searching for wood turtles, with transects spaced at 15-m intervals beginning with the river-land interface (Fig. 1). We surveyed both sides of the river simultaneously, with 2 people on each side of the river, and 2 transect lines walked per person. We typically surveyed the 0-m and 15-m transects first, followed by the 30-m and 45-m transects. Surveyors at the river-land interface wore polarized glasses to maximize detectability of wood turtles in the water near the river's edge (Jones et al. 2015). Surveyors had prior experience with wood turtle identification and searching terrestrial habitat for individuals through participation in additional wood turtle research. However, we would classify surveyors in this study as novices because they had ≤ 1 year of wood turtle field experience prior to the study.

We pre-loaded transect lines in handheld global positioning system (GPS) units (~ 3 m accuracy), which served as the center line of the approximately 20-m survey bands. Thus, there was 5 m of survey band overlap between transect lines to maximize detection of wood turtles near the edge of survey bands. Based on the pilot study, we determined that placing transects at 15 m was reasonable for maximizing distance from river surveyed while maintaining a high probability of detecting individuals within the survey band. When areas surveyed contained dense vegetation it was often not possible to stay near the center of the survey band, but the transect line allowed surveyors to navigate back to the center. Further, surveyors were free to move from the center to search cover objects but otherwise remained near the center to maximize visibility across the survey band. We did not place a time constraint on the survey.

For each new wood turtle detected, we recorded the survey band where it was detected and time of detection, obtained standard measurements and photographs, marked the individual using carapace notches (Cagle 1939), and released

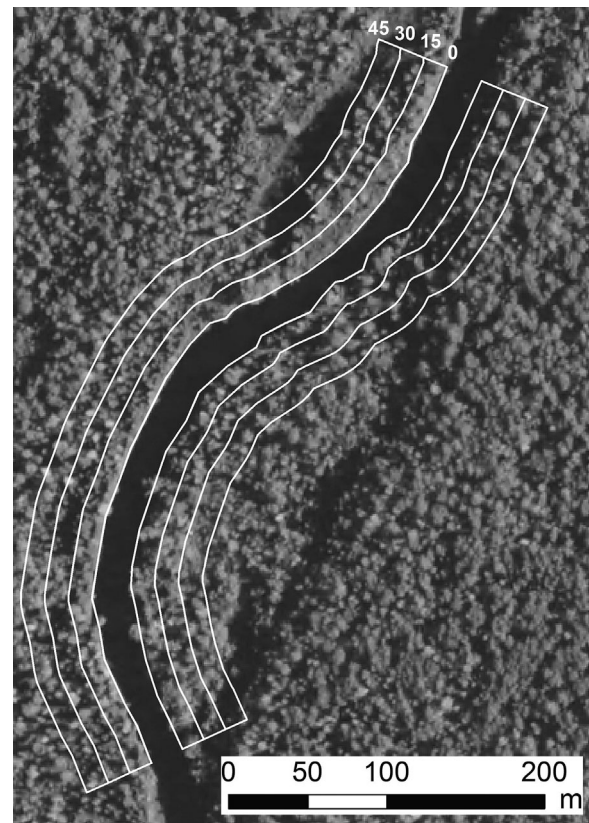


Figure 1. Transect-based sampling design for surveying wood turtle populations in the Upper Midwest, USA. We actively searched for wood turtles along transects placed at 0 m, 15 m, 30 m, and 45 m from the river's edge. We instructed surveyors to visually search for wood turtles within 10 m of transect lines, allowing for 5 m of search area overlap at the edges of survey bands. The example survey site depicted in the figure is not located in the study area.

the individual where it was detected. We processed within-year recaptures using the same protocol, with the exception that we did not re-measure or re-mark individuals. For each survey replicate, we recorded date of survey, air temperature at the beginning and end of the survey period, and time at the beginning and end of the survey period. We also recorded whether surveys were conducted before or during initial leaf-out for understory vegetation (pre-leaf-out and early-leaf-out, respectively), which could affect detectability. Sampling and handling methods were approved by the University of Minnesota Institutional Animal Care and Use Committee (Protocol No. 1504-32514A), and permitted by the Minnesota Department of Natural Resources.

Statistical Analyses

We chose to use the N -mixture class of models, rather than capture-recapture models, for wood turtle abundance estimation. N -mixture models have less stringent data requirements and can accommodate sites with very small populations, and sites with few or no recaptures (Kéry and Royle 2016). Thus, N -mixture models could be ideal for analyzing regional or range-wide wood turtle monitoring data sets. For this study, we took advantage of having marked captures by using a multinomial N -mixture model that

included a removal sampling observation process (removal model; Royle 2004a). We note that a standard binomial N -mixture model can be considered if individuals are not marked (Royle 2004b), and marking could become a monitoring limitation if the species is federally listed under the Endangered Species Act.

We defined a series of candidate model structures *a priori* to determine important survey covariates for detection probability (p ; Burnham et al. 2011). The covariates we tested included day of year (linear and quadratic relationship), survey start time, temperature (i.e., \bar{x} air temperature based on a reading at the start and end of the survey; linear and quadratic relationship), leaf-out (i.e., whether the survey was conducted during pre-leaf-out or early-leaf-out), and visibility during the survey (i.e., a categorical covariate representing sunny-partly cloudy or overcast-rainy). For the latent abundance distribution, we included a categorical covariate that represented the 8 sites, thus allowing the abundance intercept to differ among sites (Kéry and Royle 2016). Preliminary analyses found inclusion of this covariate substantially improved model fit. We assessed model goodness-of-fit using a 1,000-replication parametric bootstrap of the Pearson chi-square statistic (Mazerolle 2016), which indicated some overdispersion ($\hat{c} = 1.52$). We ranked candidate model structures using Quasi-Akaike's Information Criterion corrected for small sample size (QAIC_c), which is a modification of AIC_c to account for overdispersion (Symonds and Moussalli 2011), and selected the model structure with the highest model weight as the top model. We also accounted for this overdispersion in our site-level abundance estimates by inflating the 95% confidence intervals based on the \hat{c} values (Kéry and Royle 2016). We used the top model structure for additional analyses.

To assess the influence of distance from river surveyed (i.e., no. transects), we estimated abundance based on individuals detected from surveying transect 1, 1–2, 1–3, and 1–4, where transect 1 represents the river-land interface. To assess the influence of number of survey replications, we estimated site-level abundance based on surveying the 4 transects with 3–8 replications. We performed this assessment using the field survey data in the order surveys were conducted, and also with a simulation model (described below). We compared mean abundance estimates using reduced survey effort to mean abundance estimates using the full data set, and chose an approximately 10% difference as a reasonable difference for consideration of adopting reduced survey effort for a monitoring program. We also computed 95% confidence intervals to compare precision of abundance estimates among sampling designs.

We used simulation models to investigate the influence of number of surveys and sites on model accuracy. Using template code provided in Fiske et al. (2015) and Veech et al. (2016), we created a simulation model that generated true abundance and hypothetical count data using a removal sampling design, estimated abundance based on the hypothetical count data, and calculated the proportional difference between estimated and true abundance (i.e., \hat{N}/N). For each simulation iteration, we allowed true mean

abundance to vary randomly from 8 to 45 individuals, representing the 25–75th percentile of estimated abundances among our field sites. We generated true site-level abundances using random draws from a Poisson distribution based on the mean abundance. For each survey replication at each site, we allowed probability of detection for new individuals to vary randomly from 0.10 to 0.20. In our field study, the mean proportion of new individuals detected in each survey relative to estimated new individuals available ranged from 0.06 to 0.17, but we expect this proportion to increase in the future assuming more survey replications are completed during optimal conditions. There was no clear temporal trend in proportion detected, and thus we drew values from a single uniform distribution for all survey replications. We performed the survey replication simulations using 100 sites and 3–8 survey replications. We performed the site replication simulations using 8 surveys and 5, 10, 15, 20, and 40 sites. We completed 1,000 replications for each simulation and calculated the median, minimum, maximum, and 25–75th percentile values.

To estimate the number of surveys required to reliably infer site absence, we used binomial probability distribution simulations based on sites with low (0.25), moderate (0.5), and high (0.75) probability of species detection per survey. We chose this range of probabilities based on our field data results from surveying 2 transects (i.e., we detected the species during $\geq 25\%$ of surveys among sites). For each probability, we performed 1,000 simulation replications for each of 1–12 survey replications, and calculated the proportion of trials where the species was detected during ≥ 1 survey.

Finally, we determined whether demographic estimates differed depending on distance from river surveyed. For each site, we used unique individuals detected based on surveying transects 1–2 and 1–4 to calculate the mean sex ratio (i.e., proportion of adults and sub-adults that were M) and mean size (i.e., straight-line carapace length). We used paired randomization tests with 10,000 iterations to determine whether these metrics differed between the sampling designs. Specifically, we paired estimates from each site; for each iteration, we randomized the site data and computed the difference between survey designs. The P -values represented the proportion of trials resulting in a mean difference in demographic parameters between sampling designs greater than the one obtained in our study (Sokal and Rohlf 1995). We conducted statistical analyses using program R (version 3.2.4, www.r-project.org, accessed 17 Jan 2017). For abundance analyses, we used the software packages unmarked (version 0.11-0; Fiske and Chandler 2011) and AICcmodavg (version 2.0-4). We specified the removal models using the function multinomPois in unmarked.

RESULTS

We detected 313 wood turtles, including 174 unique individuals, during the 64 surveys. Number of detections per site ranged from 4–95 ($\bar{x} = 39$), and unique individuals detected ranged from 3–54 ($\bar{x} = 22$). We obtained 35.7%, 33.9%, 18.9%, and 11.5% of detections from transects 1, 2, 3,

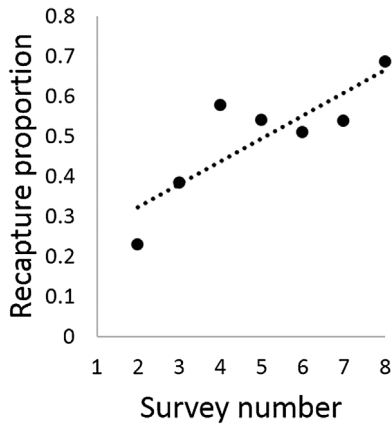


Figure 2. Proportion of total captures that were recaptures during 8 wood turtle visual encounter surveys at 8 candidate long-term monitoring sites in northeastern Minnesota, USA, in spring 2016. The dotted line represents a least squares line of best fit.

and 4, respectively. Among surveys, day of year ranged from 121–157 ($\bar{x} = 142$), survey start time ranged from 0845 to 1700 ($\bar{x} = 1203$), and survey air temperature ranged from 10.3–31.8°C ($\bar{x} = 20.9^\circ\text{C}$). We conducted 41 surveys during sunny–partly cloudy conditions, and 23 during overcast–rainy conditions. We conducted 4 surveys during pre-leaf-out and 4 during early-leaf-out at each site.

The proportion of detected individuals that were recaptures increased as survey replications increased, with 68% of captures being recaptures by the eighth replication (Fig. 2). Thus, our survey method appears effective for detecting a substantial proportion of the population. The top-ranked model structure included air temperature (quadratic) for p (Table 1). The model indicated a strong relationship between

Table 1. Model selection results to determine which covariates strongly influenced detection probability (p) of wood turtles in northeastern Minnesota, USA, in spring 2016 based on Quasi-Akaike’s Information Criterion corrected for small sample size (QAIC_c) to account for overdispersion ($\hat{c} = 1.52$). The covariates we tested included day of year (linear [DayL] and quadratic [DayQ]), survey start time (Start), temperature (i.e., mean air temperature based on a reading at the start and end of the survey; linear [TempL] and quadratic [TempQ]), leaf-out (i.e., whether the survey was conducted during pre-leaf-out or early-leaf-out; LeafOut), and visibility during the survey (i.e., a categorical covariate representing sunny–partly cloudy or overcast–rainy; Visibility). The null model is shown as $p(\cdot)$. We also present model weights (w_i). For these analyses we used multinomial N -mixture models that included a removal sampling observation process. All models included a categorical site covariate for abundance.

Structure	Parameters	QAIC _c	ΔQAIC _c	w_i
$p(\text{TempQ})$	12	107.44	0.00	0.52
$p(\cdot)$	10	108.92	1.49	0.25
$p(\text{TempQ}+\text{LeafOut})$	13	109.23	1.80	0.21
$p(\text{LeafOut})$	11	116.69	9.25	0.01
$p(\text{TempL})$	11	117.19	9.75	0.00
$p(\text{DayL})$	11	118.00	10.56	0.00
$p(\text{Start})$	11	118.08	10.64	0.00
$p(\text{TempL}+\text{LeafOut})$	12	120.78	13.35	0.00
$p(\text{DayQ})$	12	121.02	13.58	0.00
$p(\text{Visibility})$	11	122.91	15.48	0.00

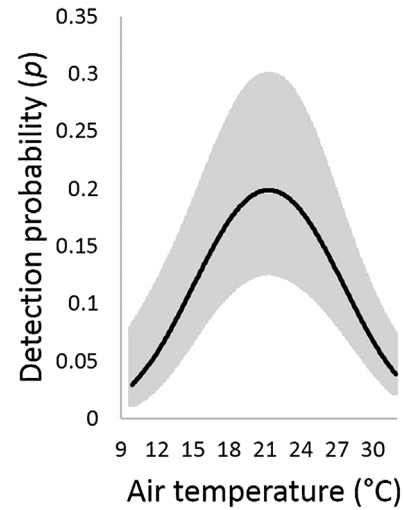


Figure 3. Estimated relationship between detection probability (p) and air temperature for wood turtles in northeastern Minnesota, USA, in spring 2016 based on visual encounter surveys at 8 candidate long-term monitoring sites. The gray band represents the 95% confidence interval.

air temperature and p , with maximum p occurring at 19–23°C (Fig. 3). Using 8 sites and 8 replications, mean estimated abundance was 247, with mean estimated abundance per site ranging from 5 to 77 (Table 2).

For our assessment of the influence of distance from river surveyed based on our field study, mean estimated abundance was 81, 227, 232, and 247 when including 1, 1–2, 1–3, and 1–4 transect(s), respectively. Thus, surveying only the first 2 transects resulted in a mean abundance estimate within 10% of the 4-transect data set, but with a substantial loss in precision (Fig. 4A). For our assessment of the influence of number of survey replications based on our field study, mean estimated abundance was 185, 137, 169, 275, 271, and 247 when including 3, 4, 5, 6, 7, and 8 replications, respectively. Thus, completing 6 replications resulted in a mean abundance estimate within 11% of 8 replications, but again with substantial loss in precision (Fig. 4B).

For our assessment of the influence of number of survey replications based on simulations using 100 sites, the 25–75th percentiles for \hat{N}/N ranged from 0.93–1.10

Table 2. Total unique individuals detected and estimated abundance of wood turtles at 8 sites in northeastern Minnesota, USA, in spring 2016 based on 8 survey replications and use of a multinomial N -mixture model that included a removal sampling observation process. To account for overdispersion (\hat{c}), we inflated the 95% confidence intervals by $\hat{c} = 1.52$.

Site code	Unique individuals	\hat{N}	95% CI
BO	3	4.57	1.09–19.13
CUT	5	7.65	2.49–23.53
GLN	5	6.61	2.17–20.10
IL	54	76.72	49.72–118.38
LG	25	36.36	20.66–63.98
NLG	7	9.95	3.82–25.97
SP	32	43.36	26.19–71.79
TR	45	62.15	39.55–97.68

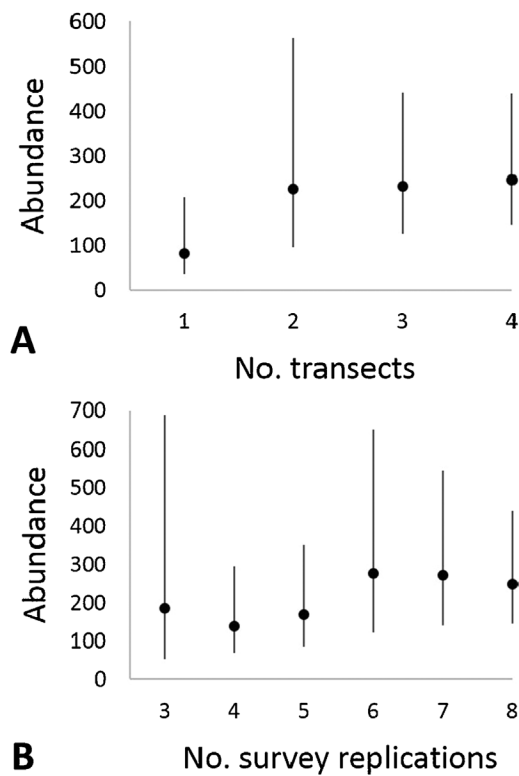


Figure 4. Estimated total wood turtle abundance at 8 candidate long-term monitoring sites in northeastern Minnesota, USA, in spring 2016 based on number of transects (A), and number of survey replications (B). We modeled abundance using a multinomial N -mixture model with a removal sampling observation process. We conducted surveys along transects placed at 0 m, 15 m, 30 m, and 45 m from the river's edge, and estimated abundance based on individuals detected from surveying transects 1, 1–2 (2), 1–3 (3), and 1–4 (4). We conducted 8 survey replications at each site, and estimated abundance based on completing the first 3–8 surveys. Circles show the sum of mean abundance estimates at each site (i.e., estimated total abundance), and lines contain the sum of lower and upper 95% confidence intervals at each site (i.e., estimated precision).

with 3 surveys, to 0.99–1.01 with 8 surveys (Fig. 5A). The 25–75th percentile range was similar when ≥ 6 survey replications were completed. For our assessment of the influence of number of sites based on simulations using 8 surveys, the 25–75th percentiles for \hat{N}/N ranged from 0.91 to 1.14 with 5 sites, to 0.95–1.07 with 40 sites (Fig. 5B). The 25–75th percentile range was similar when ≥ 15 sites were surveyed.

The simulations to estimate number of survey replications required to reliably infer species absence indicated 9 and 11 surveys were required to restrict false absences to $<10\%$ and $<5\%$ of trials, respectively, for low detection sites (Fig. 6). Moderate detection sites required 5 and 10 surveys to restrict false absences to $<5\%$ and 0% of trials, respectively. High detection sites required 3 and 6 surveys to restrict false absences to $<5\%$ and 0% of trials, respectively. For our assessment of whether distance from river surveyed affected demographic estimates, estimated proportion of males ($P=0.681$) and mean straight-line carapace length ($P=0.542$) did not differ based on whether 2 or 4 transects were surveyed. Further, percentage of individuals by size

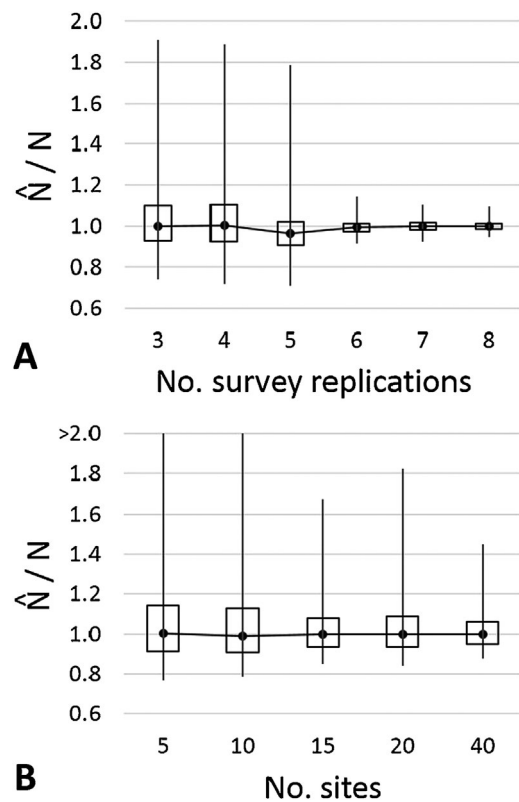


Figure 5. Proportional difference between mean estimated and true abundance of wood turtles based on 100 sites with a variable number of surveys (A), and 8 surveys with a variable number of sites (B). We performed these simulations using a multinomial N -mixture model with a removal sampling observation process. We allowed true mean abundance to vary randomly from 8–45 individuals, and generated true site-level abundances using random draws from a Poisson distribution based on the mean abundance. We allowed probability of detection of new individuals during each survey replication to vary randomly from 0.10–0.20. Circles show the median value, rectangles contain the 25–75th percentile of simulation replications, and lines contain the minimum and maximum values from 1,000 simulation replications.

class was nearly identical between the sampling designs (Fig. 7).

DISCUSSION

Our visual encounter survey design coupled with abundance estimation using a removal model resulted in estimates that appear reasonably accurate and precise for wood turtle populations in northeastern Minnesota. In comparison with the survey design of Jones et al. (2015), we found that for our populations, increasing the survey distance from river from 10 m to 25 m (i.e., having a survey transect at the riverland interface and 15 m inland) was necessary for obtaining abundance estimates that were similar to the full data set. However, considering this modification to their survey design would require relaxing the 1-hour search time constraint. Our monitoring sites were smaller (380–560 m vs. ~ 1 km of river), but our 4 transect surveys took between 3.0 and 18.7 person survey hours ($\bar{x}=6.7$ hr), including time spent processing turtles. However, we used 4 people for surveys, and thus our actual survey time took between

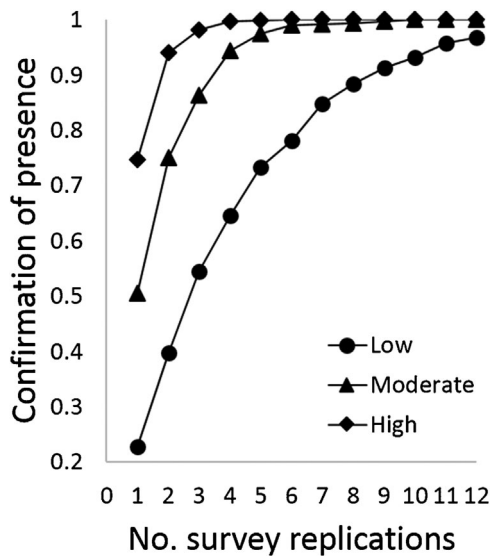


Figure 6. Proportion of 1,000 simulations resulting in confirmation of presence at occupied sites based on 1–12 survey replications and low (0.25), moderate (0.50), and high (0.75) survey detection probabilities (p). We chose survey detection probabilities based on results of wood turtle surveys in northeastern Minnesota, USA, in spring 2016.

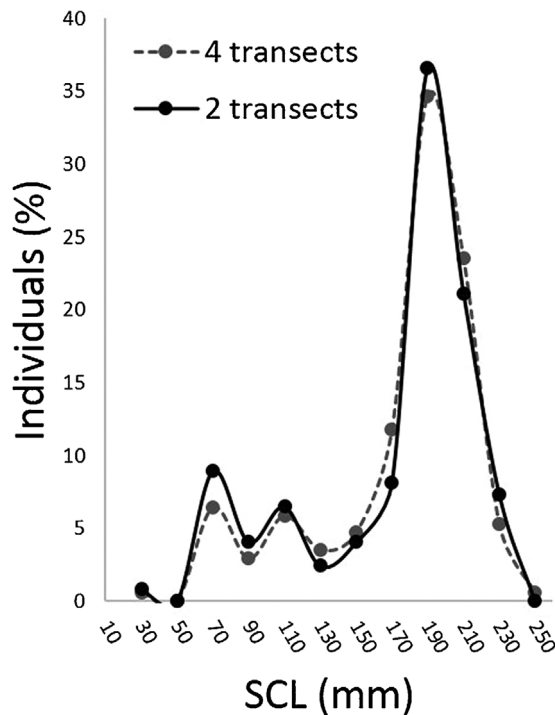


Figure 7. Percentage of unique wood turtles detected by straight-line carapace length (SCL) size class at 8 candidate long-term monitoring sites in northeastern Minnesota, USA, in spring 2016 using visual encounter surveys. We conducted surveys along transects placed at 0 m, 15 m, 30 m, and 45 m from the river's edge, and calculated size class percentages based on individuals detected from surveying transects 1–2 (2 transects; $n = 123$ [unmeasured captures removed]), and 1–4 (4 transects; $n = 170$ [unmeasured captures removed]).

0.8 and 4.7 group survey hours ($\bar{x} = 1.7$ hr). Processing wood turtles typically took 5–8 minutes for first captures, and 2–4 minutes for recaptures.

Time constraints could be more appropriate for survey protocols when the site does not require much observer movement (e.g., bird point count stations), or when the planned response variable is captures per unit effort. For N -mixture and occupancy models, the survey site is the sampling unit (MacKenzie et al. 2006), and thus the area surveyed should be consistent among survey replicates. However, Jones et al. (2015) reported that time spent searching was an influential covariate for detectability. Thus, we recommend that surveyors record their search time in the future for use as a p covariate, and to assist with defining a target search time for a long-term monitoring protocol. In addition, we recommend that surveyor identifiers be recorded for potential use as a p covariate. This information would allow surveyor teams to be ranked based on years of wood turtle monitoring experience, and this variable could be modeled as either a year-specific or survey-specific site covariate.

The removal model appeared to perform well for abundance estimation, even though our study area was not closed to immigration and emigration. Specifically, there was a river in the middle of each site, which represented a largely unobservable portion of the site (i.e., the river was relatively deep, wide, and tannic). In addition, during the active season wood turtles move among river, riparian, and upland habitats, and travel farther inland than the boundaries of our survey zone (but typically remain within 100 m of the river in our study area [Brown et al. 2016]), which affects availability during survey replications (Kaufmann 1995, Arvisais et al. 2002, Compton et al. 2002). Thus, both availability of an individual to be detected and our ability to detect an individual influenced estimated p . However, by spreading out survey replications over several weeks, we maximized our opportunity to eventually detect each individual that was using the site. Many river systems in the northeastern United States are clear and shallow, allowing for high detectability in the river (M. T. Jones, Massachusetts Division of Fisheries and Wildlife, personal communication). Under these circumstances, our survey protocol could be modified to take advantage of this additional site availability by adding a within-river transect.

There is interest in the potential to restrict surveys at some monitoring sites to one side of the river (A. F. Badje, Wisconsin Department of Natural Resources, personal communication). This survey modification would be useful when survey area availability is constrained by land ownership, or when it is logistically or physically difficult to access both sides of the river. An assessment of this survey modification indicated abundance would usually be underestimated (Table S1, available online in Supporting Information). This was likely due to individuals showing preference for one side of the river, resulting in non-random heterogeneity in p among individuals (Veech et al. 2016). Many factors could influence riparian and upland habitat quality, resulting in preference for one side of the river over the other (e.g., structural characteristics that affect thermo-

regulation [Dubois et al. 2009]). Thus, we do not recommend using this survey modification unless the other side of the river does not contain potentially suitable habitat, or it is acceptable to have an estimate of minimum abundance for the site. Modifying our survey design to include a double-observer approach might help ameliorate this problem by allowing availability and p to be estimated separately (Chandler et al. 2011). However, if many individuals at the site are never available for detection on the side of the river that is surveyed, the superpopulation size will likely be underestimated. We encourage further research on this topic.

We found that air temperature was a strong predictor of p , which we would expect based on previous studies that linked wood turtle aquatic and terrestrial habitat selection to temperature (Arvisais et al. 2004, Dubois et al. 2009, Tamplin 2009). Our analyses indicated the relationship was quadratic, and we speculate this is due to wood turtle habitat use patterns. As temperature increases, wood turtles are more likely to be terrestrial, and thus have higher availability for detection. However, if temperatures are high enough, wood turtles may seek cooler environments such as the river or upland burrows (Ernst 1986), thus reducing p at high temperatures. Conceptually, leaf-out could influence this apparent relationship, but mean and median survey temperatures were nearly identical between the pre-leaf-out and early-leaf-out periods in our study, and thus leaf-out probably had a minimal effect on the estimated relationship. Future research on fine resolution diel activity patterns in Upper Midwest populations would improve our understanding of wood turtle space use-temperature relationships.

We would expect that individuals are more difficult to detect after leaf-out, and we restricted our surveys to spring during the pre-leaf-out and early-leaf-out period to maximize detections. Although not the top model, a model that included temperature and leaf-out had some support (Table 1), indicating leaf-out had some explanatory power for p . Although we are unable to provide quantitative estimates of how p changes throughout the active season based on leaf cover and wood turtle activity patterns, we recommend that surveys be conducted in spring to maximize p , which is also supported by Jones et al. (2015). We found the other covariates we tested had little effect on p . Of particular importance, the time of day when surveys are conducted does not appear to be inherently important, allowing for flexibility in timing of surveys (assuming temperature is suitable).

With respect to survey design, we found the 15-m transect to be nearly as productive as the river-land interface transect for wood turtle detections, and including this transect in analyses improved accuracy of abundance estimates (i.e., estimated abundance at every site based on surveying only the river-land interface transect was lower than the no. of unique individuals detected at the site). The difference in estimated abundance and precision between 3 and 4 transects was minimal, but it makes little sense to survey 3 and not 4 transects, given the surveyor will likely need to re-traverse the area to return to their vehicle or boat. Based on our assessments of changes in accuracy and precision from survey design modifications, the optimal monitoring design would include ≥ 3 transects, ≥ 6

survey replications, and ≥ 15 sites. However, given time and personnel limitations, we recommend that, at minimum, sites are surveyed using 2 transects.

We emphasize that the chosen number of transects should be used for all within-year survey replications, otherwise p will inherently differ between replications. Survey-specific p can be modeled, but it would require a large data set and the model may not converge. In addition, all survey replications should be completed within the same calendar year to satisfy the assumption of within-year demographic closure. However, N -mixture models can accommodate sites without full survey replication effort (Kéry and Royle 2016), and thus discrepancies in survey replication among sites and years can be accommodated in data analyses. To balance temporal and spatial data resolution for a monitoring program operating under funding constraints, we recommend that annual site surveys be conducted on a rotating basis. Given the likelihood of slow population turnover for this long-lived species, and assuming poaching is not a major concern for a given population, it would probably be sufficient to resurvey each site every 5 years. Using this strategy, 25 sites could be monitored with a survey commitment of only 5 sites/year.

We found that having transect tracks in GPS units was helpful for keeping surveyors near the center of the survey band. Creating survey transects for GPS units was a simple process of first delineating the edges of the river, then creating 15-m interval bands using the Multiple Ring Buffer tool in ArcGIS (Environmental Systems Research Institute, Redlands, CA, USA), then exporting the band perimeters as tracks to GPS units using the software DNRGPS (version 6.1.0.6, www.dnr.state.mn.us/mis/gis/DNRGPS/DNRGPS.html, accessed 17 Jan 2017). Importantly, the width of rivers can change during the course of the survey period (e.g., because of snowmelt runoff). We addressed this issue in our study area by creating a series of transect groups that differed in the location of the river-land interface, and for a given survey, we selected the transect group that most closely matched the interface location.

Our survey and analysis design provides a flexible platform for handling variation in the size of monitoring sites and changes in survey effort across survey years. Because our model included site-specific abundance intercepts, it was not necessary for sites to be the same size. However, sites should be large enough that a substantial proportion of the population can be sampled (e.g., river distance of 400–600 m). Further, if the categorical site variable is replaced by quantitative predictors (e.g., to create predictive models for non-surveyed habitat), total survey area should be included as a covariate for the latent abundance parameter to account for variation in area among sites.

Our principle goal for this study was to assist the state of Minnesota and other agencies in the Upper Midwest with creation of long-term abundance monitoring programs for wood turtles. We encourage the states to consider a collaborative program that uses the same monitoring protocol to facilitate regional assessments. From an analysis perspective, this approach has the non-trivial benefit of increasing spatial

replication, which will enhance model performance and flexibility (Kéry et al. 2009) and promote tracking of abundance trends across multiple scales.

MANAGEMENT IMPLICATIONS

The wood turtle survey and analysis design we developed and tested was effective for estimating abundance of wood turtle populations in northeastern Minnesota, and we encourage its use as a template for wood turtle monitoring programs in the Upper Midwest. The cost of implementing our survey design is primarily for travel and personnel, and the design allows for flexibility in number of surveyors and amount of survey effort. Thus, our study will likely be a valuable contribution toward creating a standardized monitoring design for wood turtles in the Upper Midwest.

ACKNOWLEDGMENTS

Any use of trade, product, or firm names is for descriptive purposes only and does not imply endorsement by the U.S. Government. We thank A. F. Badje, M. D. Nelson, R. R. Buech, M. Schrage, D. Ryan, J. Hines, C. N. Lapin, M. A. Hamady, Y.-M. Lee, and C. D. Hall for discussions and suggestions related to survey design. We thank K. Huston, C. Reno, A. Holleran, K. Goebel, J. Flory, M. Swingen, and N. Ose for assistance with population surveys. We thank M. T. Jones and 2 anonymous reviewers for providing suggestions that improved the quality of this manuscript. This study was funded by the Minnesota Department of Natural Resources through a Competitive State Wildlife Grant (#F14AP00028), with additional financial support provided to M. Cochrane through a University of Minnesota Integrated Biosciences Graduate Program summer fellowship.

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Associate Editor: Matt Goode.

SUPPORTING INFORMATION

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SUPPORTING INFORMATION

Survey and Analysis Design for Wood Turtle Population Monitoring

There is interest in the potential to restrict wood turtle (*Glyptemys insculpta*) surveys at some monitoring sites to one side of the river. This survey modification would be useful when survey area availability is constrained by land ownership, or when it is difficult to access both sides of the river. To assess the effect of only surveying one side of the river, we estimated abundances using our field data set as described in the main text, but treated each side of the river as a separate site (hereafter sub-site). We removed individuals at each sub-site once detected (regardless of whether or not the individual had been detected previously on the other side of the river), which mimics the expected survey results had only one side of the river been surveyed. We then compared estimated abundances to estimates when both sides of the river were treated as a single site (hereafter site).

The comparability of sub-site and site abundance estimates was dependent on the side of the river surveyed (Table S1). Specifically, the sub-site with more individual detections resulted in abundance estimates closer to the site estimates. Even though sub-site and site estimates were similar in some cases, sub-site estimates were lower in 12 of 16 cases, indicating single-side surveys will usually under-predict abundance. This is likely due to individuals showing preference for one side of the river, resulting in non-random heterogeneity in detection probability (p) among individuals. Of 79 individuals recaptured at least once, 44 (55.6%) were always recaptured on the same side of the river.

Table S1. Mean estimated abundance of wood turtles (*Glyptemys insculpta*) at 8 sites in northeastern Minnesota, USA, based on 8 survey replications and use of a multinomial N-mixture model that included a removal sampling observation process. Site refers to estimates from surveying both sides of the river. Sub-site refers to estimates from surveying only one side of the river, with columns separating the lower (low) and higher (high) sub-site abundance estimates. The number of unique individuals detected at each sub-site is represented by n .

Site Code	Site	Sub-site (low)	n (low)	Sub-site (high)	n (high)
BO	4.6	2.0	1	5.9	3
CUT	7.7	5.9	3	7.9	4
GLN	6.6	0.0	0	8.4	5
IL	76.7	40.6	22	73.8	40
LG	36.4	26.4	14	30.2	16
NLG	9.9	3.6	2	9.1	5
SP	43.4	27.7	16	43.4	25
TR	62.2	40.8	23	62.1	35
<i>Sum</i>	247.5	147.0		240.8	