



Original Article

The True Cost of Partial Fencing: Evaluating Strategies to Reduce Reptile Road Mortality

CHANTEL E. MARKLE,¹ *Department of Biology, McMaster University, 1280 Main Street W, Hamilton, ON L8S 4K1, Canada*

SCOTT D. GILLINGWATER, *Upper Thames River Conservation Authority, 1424 Clarke Road, London, ON N5V 5B9, Canada*

RICK LEVICK, *Long Point World Biosphere Reserve Foundation, P.O. Box 338, Port Rowan, ON N0E 1M0, Canada*

PATRICIA CHOW-FRASER, *Department of Biology, McMaster University, 1280 Main Street W, Hamilton, ON L8S 4K1, Canada*

ABSTRACT One of the deadliest roads in North America for species at risk fragments a marsh-lake ecosystem. To reduce road mortality, stakeholders installed >5 km of exclusion fencing along a southwestern Ontario, Canada, causeway in 2008–2009. Between 2012 and 2014, 7 culverts were installed to provide safe crossings. We evaluated the success of these mitigation strategies by 1) comparing results of road surveys conducted 5 years before and 5 years after fencing installation; and 2) monitoring use of culverts by turtles using motion-activated cameras at culvert openings and stationary antennas placed to detect movements of passive integrated transponder (PIT)-tagged turtles (68 Blanding's turtles [*Emydoidea blandingii*] and 30 spotted turtles [*Clemmys guttata*]). We also radio-tracked 30 Blanding's turtles to measure culvert use in relation to home ranges. Turtle and snake abundance was 89% and 53% lower, respectively, in completely fenced road sections than in unfenced sections; abundance was 6% and 10% higher, respectively, between partially fenced and unfenced sections. After mitigation, locations where we found reptiles on the road were associated with fence ends, underscoring the importance of fence integrity and ineffectiveness of partial fencing as a mitigation strategy. We confirmed use of culverts by Blanding's turtles, northern map turtles (*Graptemys geographica*), snapping turtles (*Chelydra serpentina*), and midland painted turtles (*Chrysemys picta*). Through radio-tracking, we determined that male and female Blanding's turtles home ranges overlapped with different segments of the causeway. We recommend that stakeholders emphasize ensuring fence integrity and continuity, limiting impact of edge effects, and conducting a comprehensive monitoring program. © 2017 The Wildlife Society.

KEY WORDS Blanding's turtle, *Clemmys guttata*, culverts, *Emydoidea blandingii*, exclusion fencing, reptiles, road mortality, spotted turtle.

Roads are a significant threat to biodiversity, often resulting in declines in sensitive species (e.g., Fahrig et al. 1995, Ashley and Robinson 1996, Steen and Gibbs 2004, Aresco 2005a, Fahrig and Rytwinski 2009). In addition to causing mortality, roads can have direct negative effects through habitat loss and fragmentation, and indirect effects through reduced landscape connectivity and gene flow (Jackson and Griffin 2000). Roads can also influence thermal and hydrological regimes, noise and light levels, and invasive species colonization beyond the road edge, generally known as the road-effect zone (Andrews et al. 2008, Beckmann et al. 2010). In southwestern Ontario, Canada, roads are of particular concern because of the high incidences of species-road interactions, given that the great majority of all land in this region is within 1.5 km of a road (Gunson 2010). For reptiles, these interactions may be more frequent because

they can be attracted to roads for thermoregulation (Sullivan 1981) and nesting (Andrews et al. 2008), or need to cross roads to access habitat for foraging, brumation, and mating. Road mortality can be detrimental for many species but especially for turtles, which have life-history traits (low juvenile recruitment and delayed sexual maturity) that increase their susceptibility to population declines following even small increases in adult mortality (Marchand and Litvaitis 2004, Steen and Gibbs 2004, Aresco 2005a), and snakes, which already face a number of threats in this region, including heavy persecution by humans (Ashley et al. 2007).

The conservation or establishment of corridors is often recommended to mitigate negative effects of habitat fragmentation by roads (Beier and Noss 1998). To properly implement this mitigation strategy, a 2-stage approach is required. The first step is to construct exclusion fencing to prevent animals from accessing the road and guide them to crossing structures. Exclusion fencing can be installed in a variety of ways such as along one side of the road, both sides of the road, or certain segments of the road (Gleeson and Gleeson 2012); however, installation of any exclusion fence could restrict movement of

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¹E-mail: marklece@mcmaster.ca

target species across the landscape (Jaeger and Fahrig 2004). Therefore, the second step is to build culverts to permit safe passage under the road that fragments the landscape, or build wildlife bridges to permit passage over the roadway. Implemented crossing structures (i.e., corridors or eco-passages) can include drainage pipes (Mata et al. 2008), concrete box culverts, open-grate underpasses (Jackson 2003), large aquatic underpasses (Kaye et al. 2006, Baxter-Gilbert et al. 2015), or large overpasses (Clevenger and Waltho 2000, Healy and Gunson 2014).

Installation of fencing and culverts is a costly mitigation strategy that often results in limited resources being allocated toward a rigorous monitoring program. Consequently, little is known about overall strategy effectiveness (Lesbarrères and Fahrig 2012). We 1) analyzed the effectiveness of fencing to reduce reptile abundance on the road, 2) determined whether target species used culverts and compared culvert monitoring techniques; and 3) established how Blanding's turtle (*Emydoidea blandingii*) ranges overlapped with a causeway. The causeway we investigated is one of the deadliest roads in North America for imperiled species (Aresco 2005b), especially semi-aquatic turtles such as the Blanding's turtle and spotted turtle (*Clemmys guttata*) that live in the area. First, we used data 5 years pre- and post-mitigation to analyze the effectiveness of fencing, making this one of the longest evaluation periods documented in available literature, considering that the average post-monitoring period is 1.7 years (van der Ree et al. 2007). We hypothesized that exclusion fencing would mitigate the negative effects of roads, and predicted that turtle and snake abundance would significantly decrease along sections of the road with exclusion fencing in the post-mitigation time period compared to the pre-mitigation time period. Furthermore, if our mitigation strategy eliminated negative effects of roads, then we would expect reptile locations to be randomly distributed in relation to fence ends and culverts. Secondly, we wanted to confirm use of recently installed culverts by our target species to travel between habitat fragments and compare strengths and weaknesses of available culvert monitoring techniques. Specifically, we used passive integrated transponder (PIT) tags and stationary antennas to monitor culvert use by Blanding's turtles and spotted turtles, and motion-activated cameras to monitor use by all species. Thirdly, we established if male and female Blanding's turtles require road mitigation efforts at different locations because of differences in home ranges and movements. Blanding's turtles are of particular interest in road mitigation studies because they are designated a Threatened species at the provincial level, and Endangered at the federal level (Committee on the Status of Endangered Wildlife in Canada [COSEWIC] 2005, Ontario Government 2007), and known to make long-distance movements throughout the active season (COSEWIC 2005).

STUDY AREA

Our study took place along a 2-lane paved causeway (3.6 km) in southwestern Ontario. On average, 2,780 vehicles per day use the causeway between April and October, quadrupling on summer weekends (Wilson and Craig 2009). Following

its initial construction in the 1920s, the causeway was raised by 1.5 m in the 1950s to reduce flooding across the road, which severed 3 of the natural marsh-lake connections. In 2012, one of these natural marsh-lake connections was restored, and efforts are underway to reconnect the remaining 2. To the east of the causeway was an open bay (referred to as bay or lake habitat herein) and to the west was a 1,200-ha wetland complex (Fig. 1). This wetland complex provided critical habitat for waterfowl, fish, and many Threatened and Endangered species (Environment Canada 2015). High levels of road mortality were documented in road-kill surveys conducted in 1979, 1980, 1992, and 1993 (Ashley and Robinson 1996). In response to growing concern over the high level of mortality, a project steering committee (consisting of government and non-government participants) was formed in 2006 to develop an action plan to mitigate against road mortality. This resulted in >5 km of exclusion fencing being installed along various portions of the causeway in 2008 and 2009 (Table S1, available online in Supporting Information). Silt fencing (1 m in height) was initially installed along the causeway, but later replaced with a woven geotextile (Hinspergers Poly Industries Ltd., Mississauga, Ontario, Canada). The 122-cm (48-inch) geotextile material was mounted on 5 × 10-cm (2 × 4-inch)

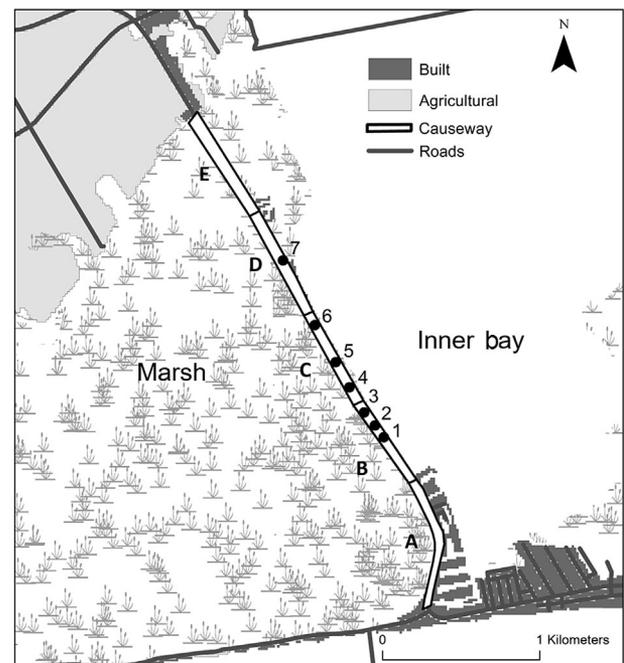


Figure 1. The 2-lane paved causeway in our study area is 3.6 km in length and separates the inner bay (east) from a wetland complex (marsh; west) in southwestern Ontario, Canada. We surveyed the causeway 5 years before (2003–2007) exclusion fencing installation in 2008–2009 and continued to survey 5 years after (2010–2014) beginning in April and concluding in October. On each survey occasion, we identified and counted all species that were alive or deceased in each of the 5 road sections. Sections A and D were partially fenced, sections B and C were completely fenced on both sides of the road, and section E had no fencing. Seven culverts (solid circles) were installed under the causeway during the post-mitigation period (2010–2014). Culverts 3–5 were constructed in the fall of 2012 and culverts 1, 2, 6, and 7 were constructed in the fall of 2014. Another road runs east-west along the southern end of the marsh. Built refers to any residential or commercial areas.

pressure-treated wooden posts using non-corrosive, large washers and deck screws. The bottom of the fencing was buried in a 20–25-cm (8–10-inch) excavated trench that was later backfilled. Following complications due to substrate and environmental conditions, some sections of the geotextile were replaced by small gauge (0.32 cm or 1/8 inch) polyvinyl chloride (PVC) mesh netting (obtained from a fishing company in Nova Scotia, Canada), with small gauge (0.32 cm or 1/8 inch) galvanized hardware cloth used in drier, upland areas. Because segments of the road bordered privately owned property that could not be fully fenced off, there were segments of the causeway with only partial fencing (incompletely fenced on one or both sides of the road; sections A and D). Sections B and C had complete fencing (both sides fully fenced) and section E had no fencing throughout the 10-year study. Between 2012 and 2014, one hydraulic concrete box culvert, 3 terrestrial open-grate culverts, and 3 concrete box culverts were installed to provide safe passage under the causeway (Table S2). An additional culvert was installed in 2014 but remained blocked off and inaccessible to species for the duration of our study.

METHODS

Field Methods

Road mortality surveys began 5 years before (2003–2007) exclusion fencing installation in 2008–2009 and continued 5 years after (2010–2014) beginning in April and concluding in October. Between 2003 and 2007, we surveyed both sides of the 2-lane causeway on foot, or by vehicle. In the 5 years surveyed in the pre-mitigation period, we conducted an average of 22 surveys each month and a total of 154 surveys. From 2010 to 2014, we conducted road surveys on foot every Monday, Wednesday, and Friday. In the 5 years surveyed in the post-fencing period, we conducted an average of 40 surveys each month and a total of 284 surveys. On each survey occasion, we identified and counted all reptiles that were alive or deceased in each of the 5 road sections (A to E in Fig. 1, Table S1). When we encountered a live individual, we moved it off the road in the direction it was headed. We considered both live and deceased individuals in our analyses because presence of either on the road represents a failure in exclusion fencing.

Culvert monitoring took place during 2014 and 2015 and involved the use of PIT tags with stationary antennas and motion-activated cameras. In spring of both years, we used baited hoop nets and dip nets to capture 68 adult Blanding's turtles (38 M, 30 F) and 30 adult spotted turtles (17 M, 13 F). In 2014, we affixed PIT tags (Biomark, Boise, ID, USA) to the carapace of 30 Blanding's turtles (15 M, 15 F); in 2015, we inserted PIT tags under the skin of 38 Blanding's turtles (23 M, 15 F) and 30 spotted turtles (17 M, 13 F). To detect the PIT tags as turtles traveled through the culverts, we mounted a stationary cord antenna attached to a custom-built PVC frame at the west entrance of 3 culverts each spring (Table S3). Both ends of the antenna were attached to an IS1001 portable enclosure (Biomark) powered by 2 deep-cycle batteries (12-volt, 75-amp hour, sealed lead acid; DCM0075, Interstate Batteries, Dallas, TX, USA). We housed the IS1001 portable

enclosure and batteries in a plastic container covered by tarp and placed it on a raised wooden platform to prevent water damage. Any time a PIT-tagged turtle passed through the antenna, PIT-tag readers logged the date, time, and identification of the turtle. Additionally, we monitored culverts using motion-activated, time lapse cameras (PC900 HyperFire Professional, Reconyx, Holmen, WI, USA; Table S3). Cameras were set to medium-high sensitivity and on a 30-min timer. Each time the motion-activation was set off, 3 consecutive photos were taken. Each week, we changed antenna and camera batteries, downloaded data, and tested all equipment for proper functioning.

In 2014, we fitted 30 Blanding's turtles (15 F, 15 M) with radio-transmitters and 5 individuals also carried global positioning system (GPS) devices (Lotek Wireless, Newmarket, ON, Canada, 10 g; Telemetry Solutions, Walnut Creek, CA, USA, 30 g). We weighed all turtles to ensure attachments did not exceed 5% of the turtle's total mass. We cleaned rear marginal scutes to allow for the best attachment of the AI-2F transmitter (Holohil Systems Ltd., Carp, ON, Canada, 19 g). We used a combination of speed set epoxy and putty epoxy to attach the transmitter. Transmitter and epoxy were colored black for camouflage. Once the epoxy was hard to the touch, we checked the transmitter for proper operation and immediately released the turtle at its capture location. Throughout the 2014 and 2015 active seasons, we radio-tracked each turtle at a minimum of once a week with a 3-element Yagi antenna (Wildlife Materials International, Murphysboro, IL, USA) and a Lotek Biotracker Receiver (Lotek Wireless, Newmarket, ON, Canada). Whenever a turtle was located, we recorded the date, time of day, GPS location (accuracy <3 m), and activity. We carried out all work out under approved animal use protocols from McMaster University (no. 11-02-05, no. 14-09-35) and site-specific permits (NWA-2014-02, WSCA 1076122, SARA-0R-2014-0260, ESA M-102-6326447130).

Statistical Analysis

We used a Poisson generalized linear mixed model in R 3.3.1 (R Core Team 2015) to determine whether number of adults on the road differed before and after exclusion fencing in road sections with complete fencing, partial fencing, or no fencing at all. We used a Poisson distribution to model our nonparametric count data and included amount of fencing (complete, partial, none) and period (before mitigation, after mitigation) as fixed effects. Because our data were repeated measures, we included year (2003–2014) and road section (A–E) as random effects to control for lack of independence. We conducted a larger survey effort post-mitigation; thus, to facilitate comparisons, we explicitly accounted for varying survey effort by including number of surveys as an offset term.

We used the SANET v4.1 (www.sanet.csis.u-tokyo.ac.jp, accessed 1 Sep 2015) extension toolbar in ArcGIS 10.2 (ESRI, Redlands, CA, USA) to test whether locations where we found reptiles on the road (after mitigation) were independently and identically distributed along the causeway (i.e., unaffected by culvert locations or fence ends). Because we considered reptile road locations to be on-network events, we used the cross K function method in SANET v4.1 to

analyze reptile road locations under the complete spatial randomness (CSR) hypothesis. We used locations of culverts or fence ends as our structural point inputs and reptile road locations as our temporal point inputs. We ran 1,000 iterations for Monte Carlo simulations and accepted significance at $\alpha = 0.05$.

We estimated Blanding's turtle population range, population core range, and male and female home ranges and core ranges using kernel density estimation (KDE) and the least-squares cross validation (LSCV) estimator. Kernel density estimation yields utilization distributions based on distribution of locations (Kernohan et al. 2001, Franklin 2010), providing an estimation of space use. We calculated all utilization distributions using at least 50 locations as suggested by Seaman et al. (1999). We used the resulting utilization distributions or kernel density surfaces to calculate 95% and 50% isopleths to estimate range boundaries (Worton 1989, Seaman et al. 1999, Powell 2000). We used the 95% isopleth to estimate home range and excluded 5% of the outermost locations, which we considered to be casual forays and thus not part of the home range (Burt 1943). We used the 50% isopleth to estimate the core range, excluding 50% of the outermost locations. In ArcGIS 10.2, we calculated overlap of home ranges and core ranges with habitat types beyond the management boundaries of the marsh our study was conducted in. These habitat types included privately managed land, additional roads, agricultural land, and local conservation authority land. We calculated overlap to identify areas of potential conservation concern and direct future management actions. We calculated home range to estimate average Blanding's turtle dispersal distance (square root of home range area; Bissonette and Adair 2008). The average dispersal distance provides a measure of how far a turtle may move to access a culvert or bypass the fence through a compromised area or move around a fence end (Baxter-Gilbert et al. 2015).

In the event that an individual Blanding's turtle used a culvert, we estimated the individual's range and used ArcGIS 10.2 to determine the percent of overlap with the causeway. To estimate individual ranges, we used the minimum convex polygon method (MCP) because we did not have more than 50 locations per turtle to use the KDE method (Seaman et al. 1999). The MCP method is commonly used because of its simplicity and calculated by creating the smallest polygon around a designated set of locations (Mohr 1947, Burgman and Fox 2003). We calculated all kernel density estimations and isopleths in GME 0.7.2.1 (Spatial Ecology LLC, Brisbane, Australia) and minimum convex polygons in ArcGIS 10.2.

RESULTS

Between 2003–2007 and 2010–2014, we recorded 1,153 deceased reptiles (498 were hatchlings or juveniles). Mortality was greatest in April, with >90% of deaths corresponding to turtles and snakes. Reptile mortality continued to be high in May and June, consisting of almost half of all recorded species. Of the 13 reptile species recorded during the 10-year survey period, 6 had sufficient abundance data to be used for in-depth analyses of the effectiveness of exclusion fencing as a mitigation strategy. These species were the Blanding's turtle, spotted

turtle, snapping turtle (*Chelydra serpentina*), eastern foxsnake (*Pantherophis vulpinus*), eastern ribbonsnake (*Thamnophis sauritus*), and eastern garter snake (*Thamnophis sirtalis sirtalis*).

For species at risk, such as the Blanding's turtle (Endangered; COSEWIC 2005), spotted turtle (Endangered; COSEWIC 2004), and ribbonsnake (Special Concern; COSEWIC 2002), the number of individuals found per survey, post-mitigation, declined by 79%, 88%, and 96%, respectively. After the installation of fencing, we observed an 18% increase in the number of snapping turtles (Special Concern; COSEWIC 2008a) found per survey and an 8% increase for foxsnakes (Endangered; COSEWIC 2008b).

The average abundance of turtles (i.e., Blanding's, spotted, and snapping turtles) found on the road declined by 16% after installation of exclusion fencing. Of the total number of turtles found, 91% were deceased. There was a significant interaction between mitigation period (pre-fencing vs. post-fencing) and amount of fencing (complete, partial, none) on turtle abundance (Fig. 2; $\chi^2 = 36$, $P < 0.001$). When the road was completely fenced, average turtle abundance declined by 89% between the pre-mitigation and post-mitigation periods, relative to the no fencing condition ($Z = -3.91$, $P < 0.001$). Although not significant, mean turtle abundance was 6% greater in sections with partial fencing compared to those with no fencing ($Z = 0.47$, $P = 0.64$).

The average abundance of snakes (i.e., eastern foxsnake, ribbonsnake, and eastern garter snake) found on the road declined by 13% after installation of exclusion fencing. Of the total number of snakes found, 93% were deceased. There was a significant interaction between mitigation (pre-fencing vs. post-fencing) and amount of fencing (complete, partial, none) on snake abundance (Fig. 3; $\chi^2 = 15.9$, $P < 0.001$). When the road was completely fenced, average snake abundance declined by 53% between the pre-mitigation and post-mitigation periods, relative to the no fencing condition ($Z = -1.7$, $P = 0.08$). Although not significant, mean snake abundance was 10% greater in sections after installation of

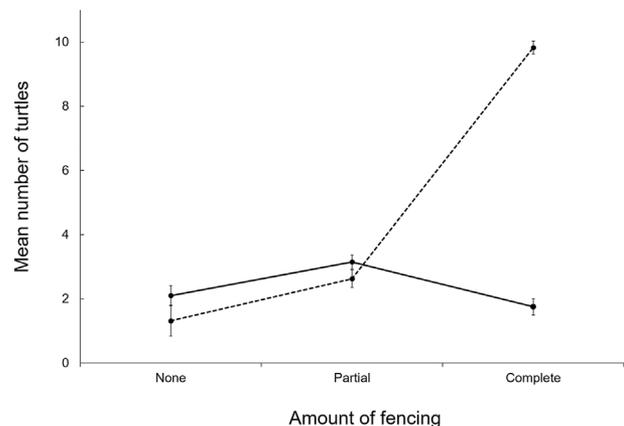


Figure 2. Results of a Poisson generalized linear mixed model displaying the mean number of turtles (\pm SE) found on a 3.6-km paved causeway in southwestern Ontario, Canada, during the pre-mitigation period (2003–2007, dashed line) and the post-mitigation period (2010–2014, solid line) in road sections with no fencing, partial fencing, or complete fencing.

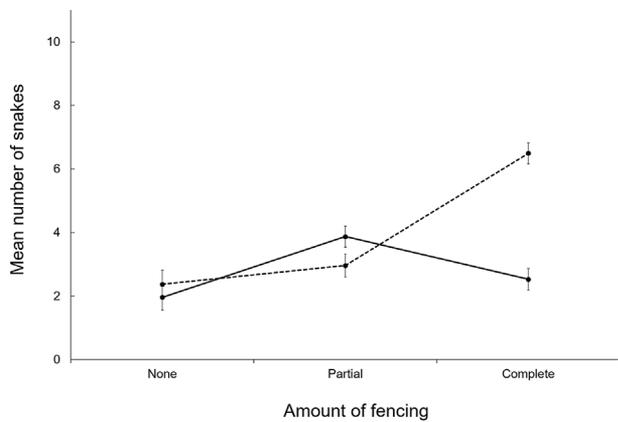


Figure 3. Results of a Poisson generalized linear mixed model displaying the mean number of snakes (\pm SE) found on a 3.6-km paved causeway in southwestern Ontario, Canada, during the pre-mitigation period (2003–2007, dashed line) and the post-mitigation period (2010–2014, solid line) in road sections with no fencing, partial fencing, or complete fencing.

partial fencing compared to those with no fencing ($Z = 0.69$, $P = 0.49$).

Culvert Use

Of the 30 PIT-tagged spotted turtles and 68 PIT-tagged Blanding’s turtles, we confirmed that 2 male Blanding’s turtles used the large aquatic culvert to safely cross under the road, one individual using the culvert in consecutive years (Table S4). Of the 15 male Blanding’s turtles we radio-tagged, these were the only 2 to use the inner bay. Both male Blanding’s turtles spent up to a month in the bay, traveled no farther than 65 m from the road and maintained a small active range of 20–25 ha (MCP) of which less than 15% overlapped with the causeway. In one instance, we tracked a turtle to the bay but had no corresponding record of culvert use. After examining the fencing, we ascertained that the turtle crossed over the road through a compromised area. We immediately repaired the fencing and the turtle used the culvert later in the summer to cross back to the marsh.

Based on motion-activated, time-lapse cameras, we confirmed that the terrestrial open-grate culverts were used by northern map turtles, midland painted turtles, and snapping turtles, and terrestrial concrete box culverts (flooded throughout season) were used by Blanding’s turtles (Table S4). Throughout the summer, our cameras also captured photos of snakes, but it was nearly impossible to identify these to species because of their small body size and most of them crossed during the night. We had a similar issue with species identification when culverts were flooded and most of the individual was submerged. In some instances, we could not determine culvert use because the camera did not capture a photo of the turtle on both sides of the culvert or inside the culvert. Instead, we interpreted a single photo of a turtle as an investigation of the culvert rather than as a confirmed usage.

Cross K Function

In our cluster analysis, we found that post-mitigation reptile road encounters were distributed randomly and independent of the location of culverts, indicated by the observed curve

falling within 95% confidence intervals. The observed curve of post-mitigation reptile road encounters indicated that events significantly clustered between 35 m to 75 m from the fence ends. We also found that reptile road encounters clustered significantly between 90 m and 800 m from the fence ends. At the remaining distances from fence ends, road abundance was distributed randomly.

Home Range Analysis

We estimated population range, core range, and home range using kernel density estimation (LSCV estimator) with locational data from the 30 Blanding’s turtles (15 F, 15 M) surveyed in 2014 and 2015 (Fig. 4a and b). We collected 349 locations for females and 433 locations for males over 2 years. As expected, female home range (828 ha) was larger than male home range (217 ha), given the longer distances females migrate to nest. But this was also true with respect to core range; female core range (159 ha) was larger than that for males (39 ha). Based on home range areas, average female dispersal distance was 2.9 km and average male dispersal distance was 1.5 km. In addition, home ranges indicated different areas of conservation concern. Female home range extended over unfenced section E and partially fenced section A, both of which lacked culverts. In contrast, male home range was highly concentrated over areas of the causeway that had culverts and complete fencing. Although male and female home ranges overlapped about 11–12% with the causeway, male core range overlapped 8% and female core range overlapped by <1%. Approximately 20% of the female home range stretched beyond the habitat managed by the federal government, and included a beach that was managed by the local conservation authority (2%), agricultural fields (5%), a public road (6%), and a privately managed marsh (6%).

With 790 Blanding’s turtle locations (radio-tracked data and opportunistic finds), the population range spanned 3.1 km with a length of 4 km. Area of the 95% population range was 526 ha (dispersal distance of 2.3 km) and the area of the 50% core population range was 83 ha. Even though the population range (13%) and core range (8%) overlapped with majority of the causeway (Fig. 4c), there were gaps in the existing mitigation strategy, notably in sections A and E, where there was limited fencing and no culverts. An estimated 11% of the population range extended over another road (south of the marsh; 3%), a private marsh (5%), section of protected beach (1%), and agricultural land towards the north (2%).

DISCUSSION

Similar to other studies, we identified seasonal patterns in road mortality (e.g., Ashley and Robinson 1996, MacKinnon et al. 2005). Spring is a particularly vulnerable time for reptiles after they emerge from their overwintering ground; therefore, fences damaged from the previous winter should be repaired as early in the season as possible. A challenge when conducting a decade-long study on road mortality is maintaining consistent funding for the many aspects of the project (e.g., materials, installation, personnel, monitoring). As a result, the number of surveys conducted post-mitigation was greater than pre-mitigation (40 surveys/per month vs. 22

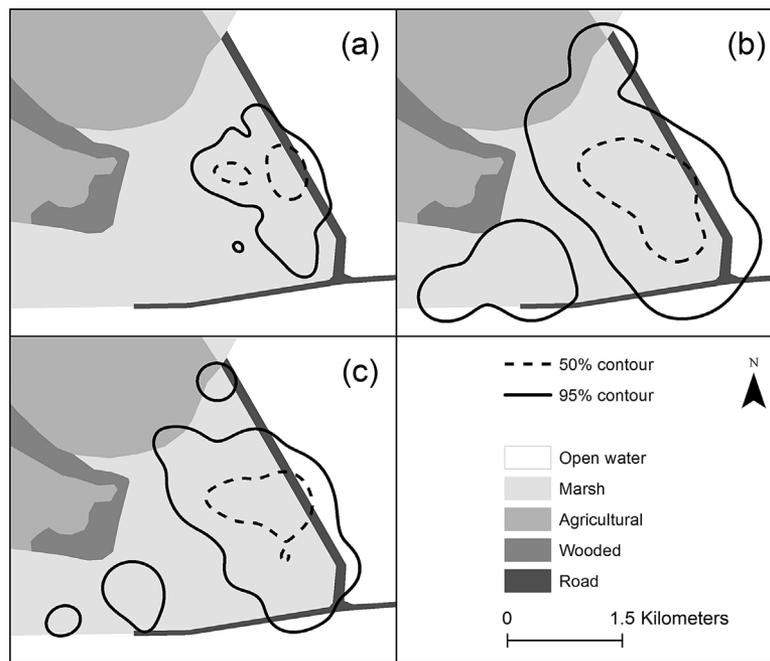


Figure 4. The 95% isopleth calculated from the kernel density estimation used to estimate home range (solid line) and the 50% isopleth used to estimate core range (dashed line) for 15 adult male Blanding's turtles radio-tracked in 2014 and 2015 (a) and 15 adult female Blanding's turtles radio-tracked in 2014 and 2015 (b) in a marsh complex in southwestern Ontario, Canada. We estimated the population range from the 95% isopleth and core range from the 50% isopleth using data from all adult Blanding's turtles radio-tracked in 2014 and 2015 (c). The population range and core range overlapped with marsh habitat, agricultural land, and surrounding roads (causeway and southern road).

surveys/per month, respectively). When more surveys are conducted, the expected bias would be toward a greater number of individuals recorded on the road; however, this was not the case in our data. When accounting for survey effort in our analysis, we found reduced reptile road abundance in areas with complete fencing. In all likelihood, we underestimated reptile road abundance pre-mitigation, and the reduction in number of reptiles on the road was even greater with complete fencing than what we reported.

For Blanding's turtles, spotted turtles, and ribbonsnakes, there was a decrease in number of individuals found per survey following mitigation by 79%, 88%, and 96%, respectively. There was a 16% reduction in abundance of turtles and 13% reduction in abundance of snakes on the road post-mitigation; however, abundance varied significantly across the different road sections. We found 89% fewer turtles and 53% fewer snakes in sections with complete fencing. For turtles, sections with partial fencing did not reduce road abundance and even resulted in 6% greater road abundances. Implications of partial fencing were even more severe for snakes, resulting in 10% greater abundances on the road compared to pre-mitigation conditions. In the case of foxsnakes, we found 8% more snakes per survey after mitigation, but where they were found along the causeway changed, with more occurrences in areas with partial fencing. This finding is not surprising for the eastern foxsnake because fencing currently in use has limited effectiveness for larger climbing snakes. For adult snapping turtles, abundance increased by 18% after mitigation efforts, occurring primarily in sections of roads that had only partial fencing.

Increased mortality had also been observed in central Ontario, where turtles were forced to make multiple crossings and increased their time on the road because of partial or compromised fencing (Baxter-Gilbert et al. 2015). In the central Ontario study, turtles accessed the road through an unfenced area, only to meet a fence on the other side of the road, and were then forced to cross back over the road again. Turtles are also known to retreat into their shell when cars pass by individuals on the road, a response that may increase time on the road (Andrews et al. 2015). Because the population of snapping turtles in our study area also appear to use the bay more frequently than do Blanding's and spotted turtles (S. D. Gillingwater, Upper Thames River Conservation Authority, unpublished data), we advocate for complete fencing on both sides of the road and the development and installation of escape hatches to exit the fenced portion of the road where complete fencing is not possible.

Fences are the key to a successful road mitigation strategy but require a well thought out design to be effective (van der Ree et al. 2015), and finding effective fencing is a common challenge (e.g., Langen 2011). Initially, we installed silt fencing (commonly used to control erosion at construction sites) along the causeway. Although inexpensive, the material suffered from ultraviolet and wind damage, and the metal staples that attached the fencing to the wooden stakes often rusted away. A year later, we replaced the silt fencing with a woven geotextile; however, after 2 years, the geotextile fencing began to fail in areas exposed to high winds. The high winds would eventually cause the geotextile to rip off

fence posts or sag between posts. We replaced some of these sections with mounted galvanized hardware cloth. Once again, these failed in some areas because the hardware cloth rusted away and ripped in damp marsh conditions. In marsh areas, we replaced the hardware cloth with a PVC mesh netting capable of withstanding both windy and wetter conditions, with a small enough mesh size to not entrap snakes. Since 2012, the main fencing system has consisted of geotextile material with smaller sections of PVC mesh netting in upland, windy areas. Even with this system, regular maintenance of fencing must be carried out to ensure tears, gaps, uprooted fence bottoms, flooded ditches, and vegetation do not compromise the effectiveness of the fencing. Going forward, Animex fencing (Animex International, Fareham, United Kingdom) will be installed and tested as a more durable, long-term solution. Based on our findings, it is important that the new fencing design consider features to prevent species from climbing over the fencing (e.g., foxsnakes). More permanent concrete or metal sheet piling barriers have been beyond the financial limitations of the project.

Based on our cluster analysis, we found the majority of reptiles that are still getting onto the road do so by traveling around fences. We detected significant clusters within 35–75 m and >90 m of fence ends. Because the causeway has 6 fence ends that are at least 400 m apart, clusters identified at large distances (90–800 m) are likely identifying neighboring fence end clusters. Based on the average dispersal distance of a female Blanding's turtle, an individual would travel up to 2.9 km (straight line distance) during a movement event. Given that the causeway is 3.6 km in length, a Blanding's turtle is likely to encounter one of the 7 culverts under the roadway. This makes it imperative to have intact fencing to direct turtles to the culvert openings, because they will gain access to the road if they encounter a compromised area in the fencing. Instead of fences ending abruptly, we suggest that fencing be angled away from the road to lead animals back to safety, as demonstrated in Florida (Aresco 2005b) and recommended in Ontario (Ontario Ministry of Natural Resources and Forestry 2013). Length of the angled fencing will likely depend on the habitat features, but based on our results, should reach 75 m if possible. If the entire 3.6 km roadway is fenced, featuring curved ends, even species that make large distance movements will be less likely to circumvent the fence. Furthermore, with the addition of new culverts at the end of 2016, the causeway will provide a total of 10 safe crossing opportunities, increasing the chance an individual will encounter a culvert before a fence end or compromised area.

We identified use of the large aquatic culvert (culvert 3) by 2 male Blanding's turtles using PIT tags and radio-tracking. Photos obtained with motion-activated cameras were unable to confirm Blanding's turtles traveling through a culvert; however, we did confirm a Blanding's turtle investigating a large terrestrial culvert (culvert 5). Neither Blanding's turtles nor spotted turtles were found to use or investigate the remaining culverts (culverts 1, 2, 4, 6, 7) during the period surveyed (May–Aug), although it is

possible some movements to or from overwintering sites had not been captured. Lack of confirmation for the spotted turtle may mean that this species needs more time to discover and use culverts because radio-tracking data from 2004 to 2007 indicate that spotted turtles do cross the road in this marsh complex (S. D. Gillingwater, unpublished data). We were able to confirm that northern map turtles, painted turtles, and snapping turtles used terrestrial open-grate culverts (1, 2, and 7), all of which were only recently installed in the fall of 2014 (and operational for the first time in 2015). This suggests that with correct placement within the landscape, culverts can reconnect habitat in the next active season following installation.

The 3 approaches we used to monitor culvert usage had different advantages and disadvantages (Table S5). Motion-activated cameras were the least costly (\$700 per camera) and required minimal field work (10 hr/week), and have been widely used (e.g., Dodd et al. 2004, Crosby 2014, Taylor et al. 2014, Baxter-Gilbert et al. 2015); however, in this study, they were difficult to use to confirm usage of culverts by animals, nor did they allow us to positively identify small-bodied species. Furthermore, they were ineffective for aquatic culverts because cameras could not capture images of animals below the water surface. The PIT tag and stationary antenna cost about \$4,000 per set up plus \$10 per PIT tag. This approach allowed us to confirm usage of culverts regardless of the animal being submerged. We were also able to easily identify species and sex of the animal within the culvert because they had been tagged at the beginning of the study. This method required about 30 min of processing per week to download data and change batteries, but was limited to the number of animals fitted with a PIT tag. The third approach was to combine the PIT tag and stationary antenna with radio-tracking. This option was the most costly (~\$13,000 for 30 turtles) and also required up to 40 hr per week of tracking, but it allowed us to determine how the target species made use of their fragmented habitats. Given these available options, we recommend that terrestrial culverts (ones which remain dry throughout the season) be monitored with cameras, whereas aquatic culverts (or those that flood for part or all of the season) be monitored with PIT tags and antennas. Most importantly, a radio-tracking program should be implemented before and after the mitigation strategy to determine whether or not culverts are actually reconnecting habitat fragments (Clevenger and McGuire 2001, Dodd et al. 2004, Lesbarrères and Fahrig 2012).

Population and home range estimates for Blanding's turtles were useful for identifying vulnerable sites within the landscape and to direct next steps. For example, despite the overlap with privately managed marsh and agricultural land, <2% of the population range occurred in agricultural fields; hence, conservation efforts on farm land should not receive top priority in this specific instance. Instead, mitigation should be focused on specific areas within the population home range where there are currently no culverts and that have limited or partial fencing. This study also revealed that 3% of the population and 6% of the female range included the unpaved road at the southern end of the study area, where Blanding's turtles crossed to access nesting habitat. Although

this represents a proportionately small amount of the adult range, the unpaved road likely results in significant mortality of hatchlings as they make their way from the beach to the marsh. Safe access to nesting habitat is key to long-term sustainability of the population. This beach habitat should be protected from further anthropogenic alterations. Our movement and habitat-use data were also consistent with our Blanding's turtle culvert crossing data, showing that only a relatively small proportion of the tracked turtles and associated home ranges included the causeway in 2014–2015. We expect the percentage of turtles using the culvert to fluctuate on a long-term basis depending on environmental conditions from year to year. Through mark-recapture studies carried out on Blanding's turtles and spotted turtles from 2003 to 2016 (S. D. Gillingwater, unpublished data), we have observed shifts in habitat use and behavior of turtles that reflect shifts in vegetation and water levels within the marsh.

MANAGEMENT IMPLICATIONS

One of the most important findings in our decade-long study was the failure of partial fencing in mitigating against road mortality. We found that partial fencing had no significant effect on reducing road abundance and was no better than road sections with no fencing. The second important lesson is the need to curve fence ends (up to 75 m) to redirect species toward interior habitat. Although culverts can help connect fragmented habitats, fencing is what keeps animals off the road and directs them to the culverts. Therefore, fence integrity is key to success and a vigilant inspection and maintenance program is essential. When selecting fence materials, it is important to consider your target species but also your site (van der Ree et al. 2015). A combination of upland, windy areas and wet, marsh conditions required 2 different types of fencing in our study area (PVC mesh and woven geotextile). Although this fencing system withstood site conditions, regular maintenance was still required to repair damages, and it did not prevent larger climbing snakes from accessing the road. We recognize that in some cases complete fencing cannot be installed; in our study area, gaps in the fencing were necessary because of private driveways and marinas. In these situations, improved designs that prevent access to the road must be considered (van der Ree et al. 2015). Research to determine effectiveness of culvert placement and type, length of lag time before use, and species-specific preferences will require a carefully designed long-term before-after-control-impact monitoring program. In developing mitigation strategies, future projects should consider incorporating movement and habitat-use data, in addition to road mortality data, to identify the most vulnerable road segments.

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LITERATURE CITED

- Andrews, K. M., J. W. Gibbons, D. M. Jochimsen, and J. Mitchell. 2008. Ecological effects of roads on amphibians and reptiles: a literature review. *Herpetological Conservation* 3:121–143.
- Andrews, K. M., P. Nanjappa, and S. P. D. Riley. 2015. Road and ecological infrastructure: concepts and applications for small animals. John Hopkins University Press, Baltimore, Maryland, USA.
- Aresco, M. J. 2005a. The effect of sex-specific terrestrial movements and roads on the sex ratio of freshwater turtles. *Biological Conservation* 123:37–44.
- Aresco, M. J. 2005b. Mitigation measures to reduce highway mortality of turtles and other herpetofauna at a north Florida lake. *Journal of Wildlife Management* 69:549–560.
- Ashley, E. P., A. Kosloski, and S. A. Petrie. 2007. Incidence of intentional vehicle-reptile collisions. *Human Dimensions of Wildlife* 12:137–143.
- Ashley, E. P., and J. T. Robinson. 1996. Road mortality of amphibians, reptiles and other wildlife on the Long Point Causeway, Lake Erie, Ontario. *Canadian Field Naturalist* 110:403–412.
- Baxter-Gilbert, J. H., J. L. Riley, D. Lesbarrères, and J. D. Litzgus. 2015. Mitigating reptile road mortality: fence failures compromise ecopassage effectiveness. *PLoS ONE* 10:e0120537.
- Beckmann, J. P., A. P. Clevenger, M. P. Huijser, and J. A. Hilty. 2010. Safe passages: highways, wildlife, and habitat connectivity. Island Press, Washington, D.C., USA.
- Beier, P., and R. F. Noss. 1998. Do habitat corridors provide connectivity? *Conservation Biology* 12:1241–1252.
- Bissonette, J. A., and W. Adair. 2008. Restoring habitat permeability to roaded landscapes with isometrically-scaled wildlife crossing. *Biological Conservation* 141:482–488.
- Burgman, M. A., and J. C. Fox. 2003. Bias in species range estimates from minimum convex polygons: implications for conservation and options for improved planning. *Animal Conservation* 6:19–28.
- Burt, W. H. 1943. Territoriality and home range concepts as applied to mammals. *Journal of Mammalogy* 24:346–352.
- Clevenger, A., and T. McGuire. 2001. Research and monitoring the effectiveness of Trans-Canada highway mitigation measures in Banff National Park, Alberta. Pages 1–16 in *Proceedings of Annual Conference and Exhibition of the Transportation Association of Canada*, Halifax, Nova Scotia, Canada.
- Clevenger, A. P., and N. Waltho. 2000. Factors influencing the effectiveness of wildlife underpasses in Banff National Park, Alberta, Canada. *Conservation Biology* 14:47–56.
- Committee on the Status of Endangered Wildlife in Canada [COSEWIC]. 2002. COSEWIC assessment and update status report on the eastern ribbonsnake *Thamnophis sauritus*. Committee on the Status of Endangered Wildlife in Canada, Ottawa, Canada.
- Committee on the Status of Endangered Wildlife in Canada [COSEWIC]. 2004. COSEWIC assessment and update status report on the spotted turtle *Clemmys guttata* in Canada. Committee on the Status of Endangered Wildlife in Canada, Ottawa, Canada.
- Committee on the Status of Endangered Wildlife in Canada [COSEWIC]. 2005. COSEWIC assessment and update status report on the Blanding's turtle *Emydoidea blandingii* in Canada. Committee on the Status of Endangered Wildlife in Canada, Ottawa, Canada.
- Committee on the Status of Endangered Wildlife in Canada [COSEWIC]. 2008a. COSEWIC assessment and update status report on the snapping

- turtle *Chelydra serpentina* in Canada. Committee on the Status of Endangered Wildlife in Canada, Ottawa, Canada.
- Committee on the Status of Endangered Wildlife in Canada [COSEWIC]. 2008b. COSEWIC assessment and update status report on the eastern foxsnake *Elaphe gloydi*, Carolinian population and Great Lakes/St. Lawrence population, in Canada. Committee on the Status of Endangered Wildlife in Canada, Ottawa, Canada.
- Crosby, J. 2014. Amphibian occurrence on south Okanagan roadways: investigating movement patterns, crossing hotspots, and roadkill mitigation structure use at the landscape scale. Thesis, University of Waterloo, Ontario, Canada.
- Dodd, C. K., W. J. Barichivich, and L. L. Smith. 2004. Effectiveness of a barrier wall and culverts in reducing wildlife mortality on a heavily traveled highway in Florida. *Biological Conservation* 118:619–631.
- Environment Canada. 2015. National Wildlife Areas. <https://ec.gc.ca/>. Accessed 2 Nov 2014.
- Fahrig, L., J. H. Pedlar, S. E. Pope, P. D. Taylor, and J. F. Wegner. 1995. Effect of road traffic on amphibian density. *Biological Conservation* 73:177–182.
- Fahrig, L., and T. Rytwinski. 2009. Effects of roads on animal abundance: an empirical review and synthesis. *Ecology and Society* 14:21. <http://www.ecologyandsociety.org/vol14/iss1/art21/>.
- Franklin, J. 2010. Mapping species distributions. Cambridge University Press, New York, New York, USA.
- Gleeson, J., and D. Gleeson. 2012. Reducing the impacts of development on wildlife. Commonwealth Scientific and Industrial Research Organization, Collingwood, Australia.
- Gunson, K. E. 2010. Green infrastructure design for municipal roads. *Municipal World* 120:9–10.
- Healy, A., and K. Gunson. 2014. Reducing wildlife collisions: what is working in northeastern Ontario. Pages 1–22 in *Transportation 2014: past, present, future*. 2014 Conference and Exhibition of the Transportation Association of Canada, Montreal, Quebec, Canada.
- Jackson, S. 2003. Proposed design and considerations for use of amphibian and reptile tunnels in New England. University of Massachusetts Report, Amherst, USA.
- Jackson, S. D., and C. R. Griffin. 2000. A strategy for mitigating highway impacts on wildlife. Pages 143–159 in T. A. Messmer and B. West, editors. *Wildlife and highways: seeking solutions to an ecological and socio-economic dilemma*. The Wildlife Society, Bethesda, Maryland, USA.
- Jaeger, J. A. G., and L. Fahrig. 2004. Effects of road fencing on population persistence. *Conservation Biology* 18:1651–1657.
- Kaye, D. R., K. M. Walsh, E. L. Rulison, and C. C. Ross. 2006. Spotted turtle use of a culvert under relocated Route 44 in Carver, Massachusetts. Pages 426–432 in *Proceedings of the 2005 International Conference on Ecology and Transportation*. Center for Transportation and the Environment, North Carolina State University, Raleigh, USA.
- Kernohan, B. J., R. A. Gitzen, and J. J. Millsaugh. 2001. Analysis of animal space use and movement. Pages 125–166 in J. J. Millsaugh and J. M. Marzluff, editors. *Radio tracking and animal populations*. Academic Press, New York, New York, USA.
- Langen, T. A. 2011. Design consideration and effectiveness of fencing for turtles: three case studies along northeastern New York state highways. Pages 521–532 in *Proceedings of the 2011 International Conference on Ecology and Transportation*. Center for Transportation and the Environment, North Carolina State University, Raleigh, USA.
- Lesbarrères, D., and L. Fahrig. 2012. Measures to reduce population fragmentation by roads: what has worked and how do we know? *Trends in Ecology & Evolution* 27:374–380.
- MacKinnon, C. A., L. A. Moore, and R. J. Brooks. 2005. Why did the reptile cross the road? Landscape factors associated with road mortality of snakes and turtles in the southeastern Georgian Bay area. Pages 153–166 in *Proceedings of the Parks Research Forum of Ontario and Carolinian Canada Coalition Annual General Meeting*. CASIOPA, Waterloo, Ontario, Canada.
- Marchand, M. N., and J. A. Litvaitis. 2004. Effects of habitat features and landscape composition on the population structure of a common aquatic turtle in a region undergoing rapid development. *Conservation Biology* 18:758–767.
- Mata, C., I. Hervás, J. Herranz, F. Suárez, and J. Malo. 2008. Are motorway wildlife passages worth building? Vertebrate use of road-crossing structures on a Spanish motorway. *Journal of Environmental Management* 88:407–415.
- Mohr, C. O. 1947. Table of equivalent populations of North American small mammals. *American Midland Naturalist* 37:223–249.
- Ontario Government. 2007. Endangered Species Act. <https://www.ontario.ca/laws/statute/07e06>. Accessed 11 Nov 2016.
- Ontario Ministry of Natural Resources and Forestry. 2013. Reptile and amphibian exclusion fencing: best practices. First edition. Species at Risk Branch Technical Note. Ontario Ministry of Natural Resources and Forestry, Peterborough, Ontario, Canada.
- Powell, A. R. 2000. Animal home ranges and territories and home range estimators. Pages 65–110 in L. Boitani and L. Fuller, editors. *Research techniques in animal ecology: controversies and consequences*. Columbia University Press, New York, New York, USA.
- R Core Team. 2015. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.
- Seaman, D. E., J. J. Millsaugh, B. J. Kernohan, G. C. Brundige, K. J. Raedeke, and R. A. Gitzen. 1999. Effects of sample size on kernel home range estimates. *Journal of Wildlife Management* 63:739–747.
- Steen, D. A., and J. P. Gibbs. 2004. Effects of roads on the structure of freshwater turtle populations. *Conservation Biology* 18:1143–1148.
- Sullivan, B. 1981. Observed differences in body temperature and associated behavior of four snake species. *Journal of Herpetology* 15:245–246.
- Taylor, S. R., N. Stow, C. Hasler, and K. Robinson. 2014. Lessons learned: Terry Fox drive wildlife guide system intended to reduce road kills and aid the conservation of Blanding's turtle (*Emydoidea blandingii*). Pages 1–21 in *Transportation 2014: past, present, future*. 2014 Conference and Exhibition of the Transportation Association of Canada, Montreal, Quebec, Canada.
- van der Ree, R., N. Gulle, K. Holland, E. van der Grift, C. Mata, and F. Suarez. 2007. Overcoming the barrier effect of roads—how effective are mitigation strategies? An international review of the use and effectiveness of underpasses and overpasses designed to increase the permeability of roads for wildlife. Pages 423–432 in C. L. Nelson and K. P. McDermott, editors. *Proceedings of the 2007 International Conference on Ecology and Transportation*. Center for Transportation and the Environment, North Carolina State University, Raleigh, USA.
- van der Ree, R., D. J. Smith, and C. Grilo. 2015. *Handbook of road ecology*. John Wiley & Sons, Ltd, West Sussex, United Kingdom.
- Wilson, A., and B. Craig. 2009. Road mortality on the Long Point causeway in 2008. Pages 87–95 in A. Liviņa, editor. *Proceedings of Sustainable Planning Instruments and Biodiversity Conservation*. University of Latvia Press, Riga, Latvia, Europe.
- Worton, B. J. 1989. Kernel methods for estimating the utilization distribution in home-range studies. *Ecology* 70:164–168.

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SUPPORTING INFORMATION

Additional supporting information may be found in the online version of this article at the publisher's website. Supporting information provides the dimensions of the culverts in our study, the monitoring periods and methods used to monitor each culvert in the study, and a table outlining culvert use by turtles in our study. Supporting information includes details on habitats found along the causeway and a comparison of culvert monitoring methods.