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Survival outcomes of rehabilitated riverine turtles following a freshwater diluted bitumen oil spill☆,☆☆

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ABSTRACT

Rehabilitation is often used to mitigate adverse effects of oil spills on wildlife. With an increase in production of alternatives to conventional crude oil such as diluted bitumen (dilbit), emergency spill responders and wildlife rehabilitators need information regarding the health and survival of free-ranging vertebrates exposed to dilbit under natural conditions. In 2010, one of the largest freshwater oil spills in the United States occurred in the Kalamazoo River in Michigan, when over 3.2 million liters of spilled dilbit impacted 56 km of riverine habitat. During 2010 and 2011 cleanup efforts, thousands of northern map turtles (*Graptemys geographica*) were captured from oiled stretches of the river, cleaned, rehabilitated, and released. We conducted extensive mark-recapture surveys in 2010, 2011, and 2018–2021, and used this dataset to evaluate the monthly survival probability of turtles 1–14 months post-spill and 8–11 years post-spill based on whether turtles were temporarily rehabilitated and released, overwintered in captivity and then released, or were released without rehabilitation. We found that rehabilitated or overwintered turtles had a higher probability of survival 1–14 months post-spill than non-rehabilitated turtles; however, 8–11 years post-spill the among-group differences in monthly survival probability had become negligible. Additionally, following the oil spill in 2010, nearly 6% of northern map turtles were recovered dead, died during rehabilitation, or suffered injuries that precluded release back into the wild. Our results demonstrate that exposure to dilbit in free ranging turtles causes direct mortality, while effort spent on the capture and rehabilitation of oiled freshwater turtles is important as it increases monthly survival 1–14 months post-spill.

1. Introduction

The adverse effects of oil spills on wildlife populations are highly visible and well documented, from the oiling of large numbers of individuals to direct oil exposure mortalities (Dunnet, 1982; Barron et al., 2020; King et al., 2020). Emergency response to oil spills generally includes rescue of oiled wildlife in the first days to weeks following a spill, rehabilitation of oil-exposed animals, or collection of individuals that died (Jessup, 1998). Studies documenting the effects of oil spills on wildlife generally focus on acute (i.e., short-term; typically, a result of initial oiling) rather than chronic effects (i.e., long-term; persisting after the initial oiling or resulting from persistent environmental pollution; Helm et al., 2015). Despite being less well-studied, chronic effects can extend for months or years after the spill and cleanup, and often exceed the magnitude of acute effects and mortalities (Iverson and Esler, 2010; Monson et al., 2011).

While rescue and rehabilitation of oiled wildlife has become routine over the past 50 years (Newman et al., 2003; Wolfaardt et al., 2008; De La Cruz et al., 2013), there is on-going debate about the effectiveness and conservation value of rehabilitating oiled wildlife. Questions remain

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regarding whether effort and financial resources should be spent on rehabilitating individual animals, particularly if there is uncertainty over their survival after release (Moore et al., 2007; Baker et al., 2015; Henkel and Ziccardi, 2018). Critics argue that funds spent on rehabilitation would be better spent on other conservation efforts such as restoring and conserving natural habitat (Henkel and Ziccardi, 2018). There is also no clear consensus on how to evaluate the success of a rehabilitation effort: that is, should rehabilitation success be measured as the survival rate of oiled animals during the rehabilitation process itself (Mignucci-Giannoni, 1999; Newman et al., 2003; Stacy, 2015), or is it instead necessary to assess post-release survival rates of rehabilitated animals (Seivwright et al., 2019), and if so, for how long? Relatively few post-release monitoring studies have been conducted to quantify survival of rehabilitated animals, despite their importance for assessing the effectiveness and conservation value of rehabilitation efforts. Furthermore, with the exception of one freshwater turtle study (Saba and Spotila, 2003), most research on rehabilitation of oiled vertebrates has focused on birds and marine animals and has generally found lower post-release survival rates of rehabilitated individuals compared to control groups (Seivwright et al., 2019). While freshwater oil spills are usually smaller in scale than spills in marine systems, freshwater spills may have a greater relative impact on oiled wildlife because the oil cannot be diluted and degraded by large volumes of water, as can occur with marine oil spills (Lee et al., 2015).

Environmental catastrophes like the *Exxon Valdez* (1989) and the Deepwater Horizon (2010) oil spills have led to a large body of research on the toxicity of conventional crude oil on wildlife. The toxic effects of such spills are mainly attributed to polycyclic aromatic hydrocarbons (PAHs; Peterson et al., 2003; Barron, 2012; Esler et al., 2018; Barron et al., 2020). Exposure to PAHs, whether acute or chronic, can lead to cardiotoxicity, behavioral changes, immunotoxicity, and decreases in reproductive success in a variety of aquatic invertebrates, fish, seabirds, and marine mammals (e.g., Reynaud and Deschaux, 2006; Barron, 2012; Wilkin et al., 2017; Honda and Suzuki, 2020). Due to recent, increased demand for crude oil and oil-related products, the use of alternatives to conventional crude oil have increased concomitantly. One such alternative is bitumen oil, production of which in the Canadian Oil Sands nearly quadrupled from 2000 to 2017 (Heyes et al., 2018). Pure bitumen oil is too viscous to be transported via pipelines directly, so it is mixed with natural gas condensates for ease of transport, which creates a product known as diluted bitumen (dilbit; Dew et al., 2015).

Although many of the chemical compounds in dilbit are also found in other crude oils, their relative proportions may differ. As a result, dilbit has higher density, viscosity, and adhesion than conventional crudes. It is also known to weather more rapidly than conventional crudes, such that its low-molecular-weight components will evaporate quickly upon exposure to wind and wave action. Weathering of dilbit leaves a mixture of high-molecular-weight compounds that may become denser than water, especially freshwater, and sinks through the water column to settle on the sediment (Dew et al., 2015; Hua et al., 2018). Laboratory studies suggest that dilbit can have similar morphological and physiological effects on wildlife as conventional crude (Dew et al., 2015; Madison et al., 2015; Philibert et al., 2021); specifically, toxicity of dilbit to fish (Alderman et al., 2016; Robidoux et al., 2018; Timlick et al., 2020; Philibert et al., 2021), invertebrates (Robidoux et al., 2018; Barron et al., 2018; Barron et al., 2021) and birds (Ruberg et al., 2022) is similar to that of conventional crude. Importantly, to our knowledge, there are no published studies on the effects of dilbit on reptiles, marine mammals, or any other free-ranging animals.

We currently know very little regarding the fate of dilbit in natural ecosystems and its effects on aquatic species. To date, most data describing the effects of dilbit on free-ranging freshwater organisms were collected in relation to the Kalamazoo River oil spill, which occurred near Marshall, Michigan, USA. On 25–26 July 2010, 3.2 million L (834,444 gallons) of material were reportedly released after a pipeline carrying dilbit ruptured (National Transportation Safety Board, 2012). Importantly, the U.S. Environmental Protection Agency (EPA) estimated that 4.5 million L (1,181,599 gallons) of dilbit were actually recovered, which would make the Kalamazoo River oil spill one of the largest inland oil spills in U.S. history, and the largest dilbit spill to date (Environmental Protection Agency, 2016). The dilbit initially pooled in a marshy area near the ruptured pipeline before flowing 213 m into Talmadge Creek, and then into the Kalamazoo River where it impacted nearly 56 km of river channel (Environmental Protection Agency, 2016, Fig. 1). The presence of submerged and sunken oil deposits reported by responders within days following the spill suggests that weathering may have occurred quickly as dilbit flowed from the pipeline rupture into the creek and river, which were in flood stage and presumably carrying large amounts of suspended solids. Ultimately, 10–20% of recovered dilbit was found to be mixed with sediment (Crosby et al., 2013). The volume of weathered and unweathered dilbit removed from the Kalamazoo River and river sediment provided an opportunity to study the potential acute and chronic effects of dilbit in a natural environment on survival of freshwater turtles, the most commonly captured animal during the Kalamazoo River oil spill cleanup.

Seven species of aquatic turtles were known to have been oiled during the Kalamazoo River oil spill, with northern map turtles (*Graptemys geographica*) being the most commonly observed oiled turtle (Environmental Protection Agency, 2016). In 2010 and 2011, >2100 northern map turtles with varying degrees of oiling were captured, cleaned, rehabilitated, and released in the Kalamazoo River. Here, we estimate monthly survival rates of northern map turtles exposed to this freshwater spill of dilbit, 1–14 months post-spill and then again for 8–11 years post-spill. We also modeled whether rehabilitation affected survival probability of male and female northern map turtles.

2. Materials and methods

Study Site – Our Study Site was ~20.2 km of channel from Talmadge Creek to E. Dickman Road in Calhoun County, MI, as this was where the majority of wildlife survey work occurred in 2010 (Fig. 1). Within the Study Site, the Kalamazoo River ranges from 9.0 to 40.0 m wide and 0.2–3.5 m deep.

Study Species – Northern map turtles exhibit pronounced sexual dimorphism, with adult females growing nearly twice the length of males (18.0–27.3 cm straight carapace length [SCL] vs 9.0–15.9 cm SCL, respectively). Males reach sexual maturity at 3–5 years of age (Iverson, 1988), while females mature after at least 10 years (Lindeman, 2013). Sex can typically be identified between 1 and 2 years of age using secondary sex characteristics (e.g., longer thicker tail, cloacal placement; Lindeman, 2013). It is estimated that wild *Graptemys* species can live between 30 and 50 years (Ernst and Lovich, 2009), and individuals of both sexes at the Study Site have been estimated to be 40+ years of age. Annual survival rates are slightly higher in females (87–94%) compared to males (81–83%; Bulté and Blouin-Demers, 2009; Bulté et al., 2009).

Both sexes are primarily aquatic but leave the water to bask daily on deadfall, rocks, or banks, making them susceptible to oiling. Turtles at the Study Site are typically active from April to October and enter a state of brumation when air and water temperatures decrease, during which they are entirely aquatic, either buried in sediment, wedged between rocks and branches, or under banks.

Turtle Capture and Rehabilitation – Immediately following the 2010 oil spill, turtle rescue and rehabilitation began on July 29, 2010, was conducted by numerous volunteers and paid contractors, including J.O., and was overseen by L.W. and the U.S. Fish and Wildlife Service (USFWS; Environmental Protection Agency, 2016). In 2010, turtle rescue efforts concluded on 24 October due to changes in weather conditions that made it difficult to capture additional turtles as they entered brumation. Level of effort differed daily, with one to five boats surveying the Study Site each day. A survey day constituted a day in which at least one boat actively captured turtles within the Study Site, and we used boats per day (boat-day) to calculate survey effort. One

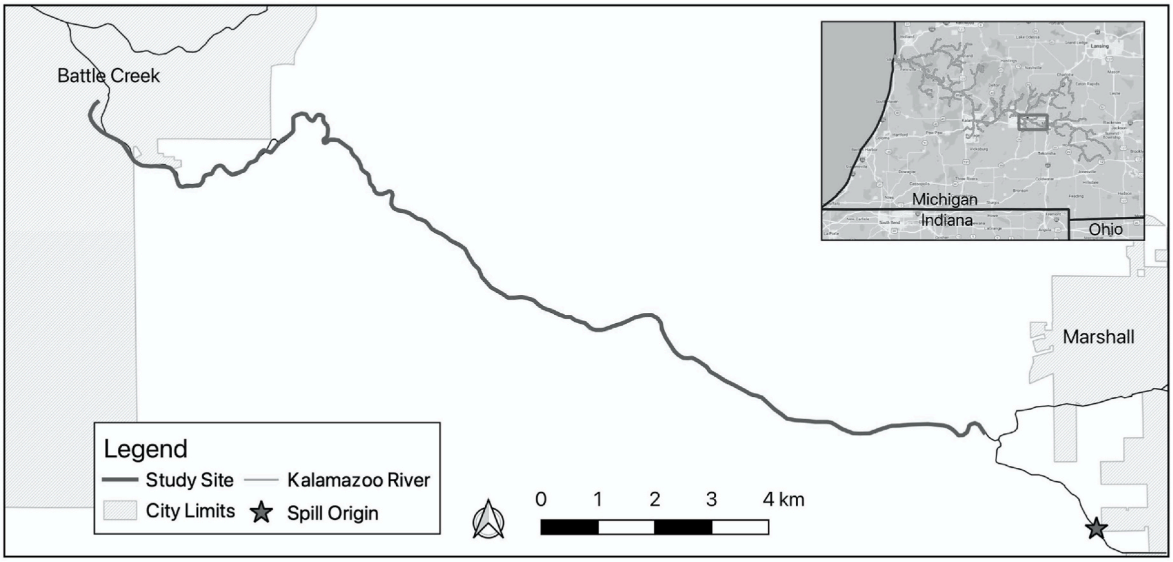

Fig. 1. Study Site surveyed to compare monthly survival rates based on rehabilitation type for northern map turtles (*Graptemys geographica*) following the 2010 Kalamazoo River oil spill. Monthly survival rates were calculated 1–14 months post-spill (2010–20110) and 8–11 years post-spill (2018–2021). The Study Site was 20.2 km of the Kalamazoo River in Calhoun County, MI, USA.

boat actively capturing turtles was considered one boat-day regardless of the number of people on the boat, rounded to the nearest ½ day. For example, if five boats surveyed the Study Site on a particular day, this would be considered five boat-days.

Turtles were captured using dipnets from a boat (Lager, 1943), baited hoop traps, and basking traps. Field crews recorded capture location of each turtle with a handheld GPS unit (Garmin International Inc.; <3m accuracy), identified sex when possible, measured shell length (i.e., straight carapace length, SCL) along the midline to the nearest 0.1 cm, mass to the nearest 0.1 g, and marked turtles >100 g with passive implanted transponder (PIT) tags (Avid Identification Systems, Inc.). Individuals <100 g were marked with a unique set of notches filed into the marginal scutes when possible (Cagle, 1939).

Upon capture, turtles exhibiting any visible oiling were retained for rehabilitation. Turtles that were not visibly oiled were processed as described above, and then released at the point of capture. The USFWS and Michigan Department of Natural Resources (MDNR) worked with Enbridge Inc., the operator of the ruptured pipeline, and their contractors (primarily Focus Wildlife and Stantec) to establish a temporary oil decontamination and wildlife rehabilitation facility in Marshall, MI (Environmental Protection Agency, 2016) where oiled turtles were photographed, physically examined by licensed veterinarians (veterinary staff was overseen by Dr. Chris Tabaka, DVM), and stabilized in individual housing until healthy enough to be cleaned (see below). For detailed description on the rehabilitation process see Supplementary Materials.

Turtle Release and Translocation – Release of rehabilitated turtles was complicated by the conflicting goals of releasing animals back to their capture location as soon as they were cleared by veterinarians, while also endeavoring to protect them from additional oiling and ongoing disturbance from cleanup operations at their original capture locations. Initially, the USFWS and MDNR coordinated translocation and release of rehabilitated turtles to other areas within the Kalamazoo River watershed (e.g., upstream or downstream of the spill, or within tributaries) as long as remaining oil precluded release of turtles at their original capture locations. On September 22, 2010, the EPA cleared impacted stretches of the Kalamazoo River for turtle release, following which turtles were released as near to their initial capture location as possible. Releases ceased on October 6, 2010, when air and water temperatures dropped to levels that stimulated winter brumation. Turtles captured after 6 October, or those still requiring cleaning and medical assistance, were overwintered and released at their point of capture between 26 April and May 19, 2011.

Surveys – We surveyed the Study Site in 2011 to both continue capturing oiled turtles, and to recapture previously rehabilitated and released turtles to assess survival. In 2018–2021, researchers from the University of Toledo (led by J.O.) attempted to recapture turtles that had been captured, rehabilitated, and released in 2010–2011. Surveys and turtle capture efforts in subsequent years used the same methods as described above for 2010. Level of effort and number of survey days varied by month and year, with the majority of effort in April–September. All captured turtles were checked for PIT tags or shell notches and were measured as described above. Unmarked turtles were individually marked with a unique combination of notches along marginal scutes (Cagle, 1939). We recorded all capture locations and turtle morphology data as described above, and all turtles were released at the point of capture within 24 h.

Data Analysis – We used northern map turtle survey data collected over six years (2010–2011 and 2018–2021) to calculate: 1) total survey effort and total number of turtle captures each year, 2) mortality rates of turtles captured in 2010 following the oil spill, 3) the monthly survival and recapture probabilities for overwintered, rehabilitated, and non-rehabilitated turtles *1*–14 months *post-spill*, and 4) monthly survival and recapture probabilities for overwintered, rehabilitated, and non-rehabilitated turtles *8–11 years post-spill*. Estimates of mortality rates, monthly survival, and recapture probabilities included only turtles that were identifiable to sex.

2.1. Turtle Capture and Survey Effort

For each survey year, we used all captures of northern map turtles, including individuals that were either released unmarked or were too young to identify to sex, to determine the total number of northern map turtles captured, and the average number captured per day. To calculate catch per unit effort (CPUE), we divided the total number of captures by the total number of boat-days.

2.2. Mortality Rates Following the Oil Spill

When determining the mortality rates of turtles in the months following the oil spill, we included only turtles captured in 2010. We defined mortality as any individual that was collected dead, died during rehabilitation or overwintering, or sustained injuries that did not allow for release back into the wild (e.g., turtles that could no longer fully submerge and therefore were transferred permanently to wildlife rehabilitators). We used a chi-square proportion test to compare the proportion of mortalities between the sexes.

2.3. Monthly survival and recapture probabilities

In our calculations of monthly survival and probability of recapture, we included only turtles that were captured, marked, and released within 1 km of their original 2010 capture location; that is, we excluded any turtles that were originally captured within the Study Site but were translocated and released elsewhere during cleanup operations. All turtles included in this analysis were categorized as "rehabilitated," "overwintered," or "non-rehabilitated." We considered any turtle that spent at least one night in captivity but was released in 2010 as "rehabilitated." Turtles that were overwintered in the rehabilitation facility during the winter of 2010–2011 and released in spring 2011 were categorized as "overwintered." Finally, turtles captured in 2010 or 2011 that did not go through any rehabilitation or overwintering were categorized as "non-rehabilitated." These were individuals with either no visible oiling, or light spotty oiling covering <5% of their body which could be easily removed with a brush. These individuals were cleaned immediately in the field and released at their point of capture.

To estimate monthly survival, and recapture probabilities of rehabilitated, overwintered, and non-rehabilitated turtles, we constructed the capture history of each individual based on its capture or non-capture during a particular sampling event. The two time periods (i.e., 2010–2011 and 2018–2021) were analyzed separately. To calculate 1–14 month post-spill monthly survival and recapture probability, we used seven sampling events: September and October 2010 combined; and monthly from May to September 2011. To calculate 8–11 year post-spill monthly survival and recapture probability, we used 19 sampling events: 3 in 2018 (May to July), 6 in 2019 (April to September), 6 in 2020 (April to September), and 4 in 2021 (April to June, and August). Only the first capture of each individual during each sampling event was included in models. Survey effort was calculated for each sampling event by totaling the number of boat-days.

We used the Cormack-Jolly-Seber (CJS) mark-recapture method (Cormack, 1964; Jolly, 1965; Seber, 1965) in program MARK (R software; White and Burnham, 1999) to estimate survival and recapture probabilities. Recapture probability is the probability that a marked individuals is recaptured, given that it is alive. Survival probability is the probability that an individual survives between sampling events. Program MARK provides parameter estimates from capture histories of marked animals when they are recaptured at later sampling events. The estimates of model parameters are computed via numerical maximum likelihood techniques (White and Burnham, 1999). To explain the mark-recapture data for each time period with respect to survival and recapture probabilities, we constructed 16 biologically plausible candidate models. Models estimating survival included all combinations of sex and rehabilitation category (i.e., rehabilitated, overwintered, or non-rehabilitated in 2010–11). Models of recapture probability also included the effect of time between surveys (t) and number of boat-days (survey effort; Table 1). We also evaluated a model with constant survival and recapture probabilities.

We conducted a goodness-of-fit (GOF) test prior to model selection to verify whether data met the assumptions of the CJS model that every animal present in the population at time *t* has the same recapture probability, and that every animal in the population immediately after time *t* has the same survival to time *t + 1* (Arnason-Schwarz Model, Pradel et al., 2003). We performed the GOF test using the R2ucare package in R (Choquet et al., 2009). We tested for overdispersion of the global model (Survival $_{sex * rehab\ type}$ Recapture $_{effort * t}$) using the median c-hat method in MARK (R software; White and Burnham, 1999), which assumes that a c-hat estimate near 1 indicates the model has reasonable fit to the data, whereas c-hat estimates >3 indicate structural deficiencies in the global model (Gonzalez-Tokman et al., 2012). Because our models were slightly over-dispersed, $QAIC_c$ (Quasi Akaike's Information Criterion corrected for bias and overdispersion) was used to compare the 16 models for survival both 1–14 months post-spill, and 8–11 years post-spill. If the $QAIC_c$ was <2, we assumed there was no difference between alternative models.

Table 1
Summary of capture efforts and results for northern map turtles (*Graptemys geographica*) in the Kalamazoo River, MI during 2010–2011 and 2018–2021 survival surveys. The total and mean number of captures were based on all individuals captured, regardless of recapture status, size, or sex. The number of individuals and number of times individually marked turtles were captured in subsequent years of survey (2018–2021) are divided by sex and rehabilitation category, i.e., whether an individual spent at least one night in the rehabilitation facility but was released in 2010 (rehab), was overwintered during the winter of 2010–2011 (overwintered), or was captured and either had no oil or was field cleaned (non-rehab).

		2010–2011	2018–2021	Total
Number of Survey Days		133	192	**325**
Number of Boat-Days[a]		247	236	**483**
Total Turtle Captures		3114	3976	**7090**
Avg. Turtle Captures/Day		23.4	20.7	**21.8**
CPUE[b]		12.6	16.8	**14.7**
Total Sexable Captures		2623	2645	**5268**
Total Captures of Marked (2010)/2011 Turtles[c]		2414	784	**3198**
Females	Non-Rehab	412	128	-
	Rehab	128	32	-
	Overwintered	164	58	-
Males	Non-Rehab	147	15	-
	Rehab	194	22	-
	Overwintered	121	32	-

[a] Number of boats actively surveying the Study Site.
[b] Total number of captures divided by the number of boat-days.
[c] Only individuals identifiable to sex marked in 2010 or 2011.

3. Results

Turtle Capture and Survey Effort – A similar number of boat-days occurred in 2010–2011 compared to 2018–2021, with an average of 1.86 boat-days per survey day compared to 1.23, respectively. The overall CPUE for 2010–2011 was 12.6 northern map turtles per day compared to 16.8 turtles per day during 2018–2021; however, the CPUE per day among years varied from 6.2 in 2018 to 32.3 in 2020, likely due to researchers' increased experience during this time period. During 2010–2011, we made 3114 total captures of 2015 individual northern map turtles over 133 survey days, while in 2018–2021, we made 3976 captures of 1845 individuals over 192 survey days (Table 1).

Mortality Rates Following the Oil Spill – We observed more mortalities of female northern map turtles immediately following the spill (up to November 1, 2010) compared to males (7.6% vs 4.1%; $x^2 = 5.83$ $p = 0.02$). Two individuals were found dead during surveys (1 female and 1 male), 50 turtles died in captivity during rehabilitation (31 females and 19 males), and 15 were deemed unfit for release and were transferred into permanent captivity (8 females and 7 males). On average, turtles that died during rehabilitation did so 57.6 (61.4 SD) days after capture.

Monthly Survival and Recapture Probability – From 2010 to 2011, we made 2414 captures of 1166 unique individuals (704 females and 462 males; Table 1), with individuals recaptured 2–12 times. A total of 322 rehabilitated (128 females and 194 males), 285 overwintered (164 females and 121 males), and 559 non-rehabilitated turtles (412 females and 147 males) that were originally captured in 2010 or 2011 were included in monthly survival analyses (Table 1). Nearly 25% of these individuals were recaptured at least once between 2018 and 2021 (228 of 1166), with the highest proportion of recaptures being turtles that were overwintered (31.6%; 90 of 285).

3.1. 1–14 Months post-spill monthly survival (2010–2011)

The GOF test of the global 1–14 months post-spill monthly survival model indicated that the model was slightly over-dispersed but still had a reasonable fit to the data (c-hat = 1.63). Of the candidate models considered, the best-supported model (i.e., lowest $QAIC_c$) was the model including rehabilitation type for survival, and time between surveys for recapture probability (φ (Rehab) ρ (t); Table 2). Under this model, 1–14 months post-spill survival probability was affected by rehabilitation type (i.e., non-rehabilitation, rehabilitation, or overwintered) that occurred in 2010, but was otherwise unaffected by sex of the turtle, in contrast to the sex difference in mortality that was observed only immediately following the spill.

For both sexes, the estimated 1–14 months post-spill monthly survival probability of turtles that had been overwintered (females n = 164, 0.983 ± 0.006 [SE; 95% CI = 0.964–0.992]; males n = 121, 0.988 ± 0.005 [SE; 95% CI = 0.975–0.994]) was significantly higher than that of turtles that had been rehabilitated but not overwintered (females n = 128, 0.910 ± 0.012 [SE; 95% CI = 0.883–0.931]; males n = 194, 0.909 ± 0.010 [SE; 95% CI = 0.888–0.926]). Female turtles that were neither rehabilitated nor overwintered had the lowest 1–14 months post-spill monthly survival probability (n = 412, 0.799 ± 0.037 [SE; 95% CI = 0.716–0.862]), while the 1–14 months post-spill monthly survival probability of males that were neither rehabilitated nor overwintered was similar to that of males that had undergone rehabilitation but not overwintering (n = 147, 0.916 ± 0.036 [SE; 95% CI = 0.812–0.965] (Fig. 2). Post hoc analysis indicated the 1–14 months post-spill monthly survival rates of rehabilitated, overwintered, and non-rehabilitated turtles to be similar for both sexes.

Under the ρ (t) model, recapture probabilities during this time period differed among survey periods, ranging from 0.036 ± 0.009 (95% CI = 0.022–0.059) in October 2010 to 0.414 ± 0.027 (95% CI = 0.362–0.468) in Sept 2011, with a mean of 0.251 ± 0.022 (95% CI = 0.211–0.295).

3.2. 8–11 Years post-spill monthly survival (2018–2021)

The GOF test of the global 8–11 years post-spill monthly survival model indicated that the model was slightly over-dispersed but still had a reasonable fit to the data (c-hat = 1.25). Of the candidate models considered, the best-supported model (lowest $QAIC_c$) was the model including sex for survival, and time between surveys for recapture probability (φ (Sex) ρ (t); Table 2). Under this model, the monthly survival probability for turtles that were alive 8–11 years post-spill was affected by sex but was otherwise unaffected by rehabilitation type. The estimated 8–11 years post-spill monthly survival probabilities of females that had been rehabilitated or overwintered were nearly identical to non-rehabilitated females (Fig. 2; rehabilitated n = 32, 0.998 ± 0.002 [SE; 95% CI = 0.987–0.999]; overwintered n = 58, 0.996 ± 0.002 [SE; 95% CI = 0.961–0.999]; non-rehabilitated n = 128, 0.991 ± 0.004 [SE; 95% CI = 0.977–0.996]). While females had significantly higher 8–11 years post-spill monthly survival rates than males, males that had been rehabilitated (n = 22, 0.977 ± 0.014 [SE; 95% CI = 0.928–0.993]) or overwintered (n = 32, 0.971 ± 0.014 [SE; 95% CI = 0.926–0.989])) were similar to those of males that were non-rehabilitated (Fig. 2; n = 15, 0.912 ± 0.035 [SE; 95% CI = 0.816–0.960]).

Under the ρ (t) model, recapture probabilities differed between among survey periods 8–11 years post-spill, ranging from 0.029 ± 0.012 (95% CI = 0.013–0.064) in April 2021 to 0.303 ± 0.040 (95% CI = 0.231–0.386) in June 2019, with a mean of 0.130 ± 0.028 (95% CI = 0.084–0.197).

4. Discussion

Determining the broad population- and community-level consequences of oil spills is necessary to establish the ecological impacts of pollution (Hinton et al., 2005). Here we provide an evaluation of the effectiveness of rehabilitation efforts on survival of a freshwater turtle population following exposure to a dilbit oil spill in the Kalamazoo River, Michigan, USA. We compared 1–14 months post-spill and 8–11 years post-spill monthly survival probabilities of oiled turtles that were either rehabilitated and released in 2010 or overwintered and released in 2011 following the 2010 oil spill, to those that had not been rehabilitated or overwintered. We found that both rehabilitated and overwintered turtles had a higher probability of survival 1–14 months post-spill than non-rehabilitated turtles; however, for those turtles surviving to 8–11 years post-spill, the among-group differences in survival probability by that time had become negligible. To our knowledge, this is the first study on impacts of a dilbit oil spill on long-term survival in a vertebrate species.

Despite the effort spent capturing and rehabilitating northern map turtles during 2010 cleanup activities, only 52 mortalities were directly observed. This effort included daily surveys from wildlife biologists specifically for turtles as well as oil spill cleanup workers inadvertently capturing turtles, often collected while removing oiled vegetation or soils. An additional 15 individuals were too severely injured for release and were instead transferred to permanent captivity; these turtles should be considered functional mortalities from a demographic perspective. Overall mortality (including un-releasable turtles) was 67 of 1181 (5.7%) northern map turtles recovered after the oil spill, 66 of which had

Table 2
Top five models describing 1–14 months post-spill and 8–11 years post-spill monthly φ (survival) and ρ (recapture) probabilities of northern map turtles (*Graptemys geographica*) in the Kalamazoo River, MI following the 2010 oil spill. Only individuals identifiable to sex were included in analysis. The null model (.) for both time periods is also included which has constant probabilities for both φ and ρ.

Type	Rank	Model[a]	K[b]	QAICc	Δ QAICc	*w*[c]	QDeviance
1–14 months post-spill (2010–2011)	1	φ (Rehab), ρ (t)	9	279.59	0.00	0.50	426.68
	2	Φ (Sex * Rehab), ρ (t)	12	281.84	2.25	0.50	420.57
	3	φ (Rehab), ρ (effort * t)	15	291.59	12.00	0.00	426.68
	4	φ (Sex * Rehab), ρ (effort * t)	18	293.84	14.25	0.00	420.57
	5	φ (Rehab) ρ (effort)	5	331.50	51.91	0.00	524.41
	15	φ (.), ρ (.)	2	486.20	206.61	0.00	786.53
8–11 years post-spill (2018–2021)	1	φ (Sex), ρ (t)	20	1136.05	0.00	0.79	1371.15
	2	φ (Sex * Rehab), ρ (t)	24	1139.37	3.32	0.20	1365.31
	3	φ (.), ρ (t)	19	1142.96	6.91	0.01	1382.31
	4	φ (Rehab), ρ (t)	21	1146.50	10.45	0.01	1371.15
	5	φ (Sex), ρ (effort * t)	38	1172.05	36.00	0.00	1365.31
	15	φ (.), ρ (.)	2	1276.77	140.72	0.00	1592.24

[a] Predictor variables for top 5 and null (.) models including sex, rehabilitation category (rehab; i.e., “rehabilitation”, “overwinter”, or “no rehabilitation”), time (t), level of trapping effort (effort), and interactions (*).
[b] Number of parameters.
[c] Akaike weight.

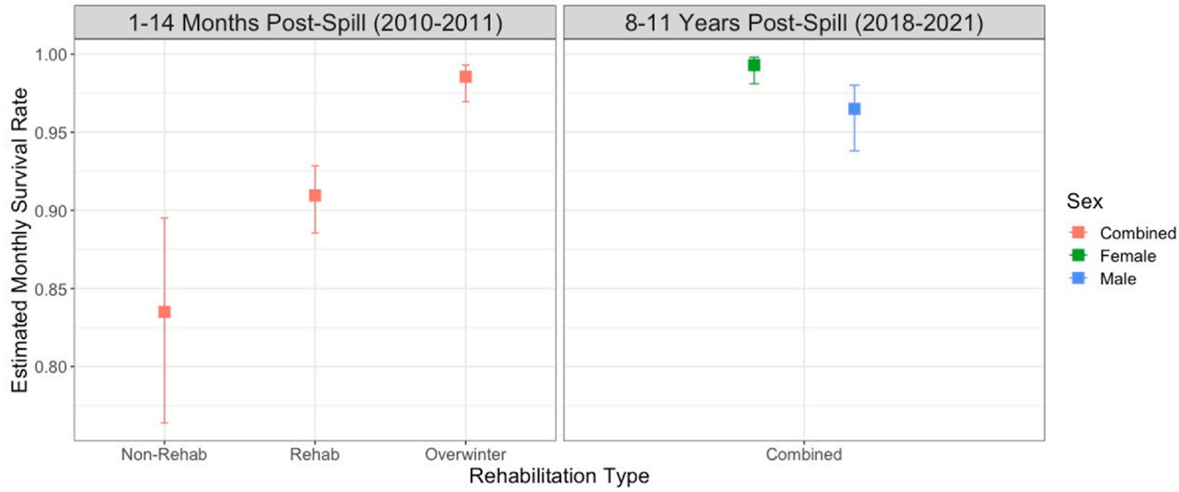

Fig. 2. Estimated monthly survival probability for northern map turtles (*Graptemys geographica*) captured following the 2010 Kalamazoo River oil spill, 1–14 months post-spill (left) and 8–11 years post-spill (right). Turtles that spent at least one night in the rehabilitation facility and were released in 2010 were categorized as "Rehab," those that spent the winter of 2010–2011 in the facility were categorized as "Overwinter," and those that were neither rehabilitated nor overwintered were categorized as "Non-Rehab." Monthly survival estimates 1–14 months post-spill were calculated from September 2010 to October 2011, while estimates 8–11 years post-spill were calculated from May 2018 to Aug 2021.

external oiling and 1 that was injured by a boat or other equipment. This apparent mortality rate was very similar to that of the only other freshwater crude oil spill that included turtle rehabilitation and reported rehabilitation mortality rates, wherein 5.3% of 19 oiled individuals died during rehabilitation (Saba and Spotila, 2003). Our observed mortality rate was nearly three times that reported during offshore sea turtle recovery in the Gulf of Mexico during the Deepwater Horizon spill of 2010, during which 328 sea turtles were rehabilitated, 7 of which later died (2.1%; Stacy, 2015; Stacy et al., 2017). Our observed mortality rate from the 2010 spill is likely an underestimate because detectability and recovery of oiled turtle carcasses was complicated by the difficulty of visually detecting smaller carcasses in the heavily oiled river and floodplain; the probability that heavily oiled carcasses were inadvertently removed along with oil, oiled vegetation, debris, and sediment; the possibility that some carcasses could have been scavenged; and safety constraints on timing and coverage of searches over a large geographic area. While this study quantified only observed mortality through oiled or injured individuals, a larger demographics and population study estimated the change in population 10 years after the spill, including estimates of mortality that may have been a direct or indirect result of the spill (Otten, 2022).

The similarity of our estimated mortality rate compared to those from other crude oil spills suggests that dilbit is similarly toxic to freshwater organisms as conventional crude oil. Importantly, the morphological and physiological effects of dilbit on freshwater organisms are still poorly understood. Toxicity of dilbit in some species of fish may be caused by PAHs binding to and activating aryl hydrocarbon (AhR) receptors (Hodson, 2017; Madison et al., 2017; Alsaadi et al., 2018), but Everitt et al. (2021) suggested both AhR-dependent and -independent mechanisms as causes of toxicities of weathered dilbit in zebrafish. Although toxicity of dilbit in turtles has not been studied, turtles are known to accumulate heavy metals (Yu et al., 2011; Hopkins et al., 2013), coal fly ash (Nagle et al., 2001; Steen et al., 2015), and PAHs (Camacho et al., 2012; Ylitalo et al., 2017) in their tissues. Dilbit weathers and degrades faster than conventional crude oil (King et al., 2014); however, acute toxicity of unweathered and weathered dilbit is similar in fish and invertebrates (Barron et al., 2018; Robidoux et al., 2018). In particular, concentrations as low as 3.5 μg/L can induce a liver biomarker of PAH exposure, while concentrations of 16.4 μg/L can induce a PAH biomarker in the heart (Alderman et al., 2016). Because our study was conducted opportunistically following an unexpected oil spill in wild habitat, data on dilbit concentrations, degree of weathering, or the duration of individual turtles' exposure to dilbit were not recorded. Moreover, to our knowledge, no toxicological post-mortem necropsies were conducted that would have provided such data. We did, however, find that most observed turtle mortalities occurred during rehabilitation, after removal of surficial oil and while turtles were under the daily care of veterinarians. These mortalities occurred an average of 57.6 days after capture, suggesting that latent deleterious physiological effects may have occurred, and which may have taken weeks or months to develop. Determining the precise effects of dilbit on the health of exposed wildlife should be a research priority, particularly considering the trend to increase transport of dilbit as an alternative to traditional crude oil.

While rehabilitation efforts similar to those used here typically occur with emergency wildlife rescue efforts following oil spills, surprisingly little is known about the long-term effectiveness of the rehabilitation process on individuals after release, or on population demographics following the spill event (Murphy et al., 2016). Studies comparing individual survival rates after rehabilitation found lower survival in rehabilitated sea otters (*Enhydra lutris*) and sea birds compared to control animals (Hartung, 1995; Rebar et al., 1995; Seivwright et al., 2019). In contrast, our results show that regardless of sex, rehabilitated oiled turtles released back into the wild had higher monthly survival probabilities 1–14 months after the 2010 Kalamazoo River oil spill compared to turtles that had not been rehabilitated. Although non-rehabilitated turtles were not a true control population and experienced the same environmental conditions as rehabilitated turtles following their release, non-rehabilitated turtles had either no or very minor surficial oil that was easily cleaned in the field. Moreover, rehabilitation efforts that included overwintering oiled turtles in captivity further increased these survival rates in both sexes: turtles that were overwintered in captivity had a monthly survival probability almost 8% higher than turtles that had been rehabilitated but not overwintered in 2010, and 13% higher than non-rehabilitated turtles. This difference in survival could equate to nearly 50% fewer individuals of both sexes in the population at the end of 2011 if no turtles had been overwintered in captivity as part of rehabilitation efforts, which could have severe consequences for population demographics. Our results suggest that rehabilitation efforts should target all individuals regardless of sex, as both sexes' survival rates increased similarly based on rehabilitation type. In late-maturing, long-lived species such as turtles, even a slight decrease in adult survival could result in a substantial decrease in recruitment and population growth rates, a trend which could take many years to reverse (Congdon et al., 2003) and which would be particularly detrimental in threatened or endangered species.

We found that both temporarily housing and rehabilitating oiled turtles and overwintering them in captivity until spring increased their probability of survival compared to turtles that had not been housed in captivity. On average, rehabilitated turtles were kept 6.2 days in captivity, while overwintered turtles were kept 210.2 days. Importantly, any time spent in captivity served not only to clean and rehabilitate individuals after exposure to oil, but also decreased their contact with residual oil in the environment and with human disturbance during subsequent cleanup operations. The increase in survival probability for overwintered turtles may have been a result of constant veterinarian supervision and feeding during a 6-month period in which they are usually dormant, which allowed turtles to gain additional mass and energetic resources necessary for survival. Cleanup operations observed that dilbit settled in the sediment in low-flow backwater areas of the river (Environmental Protection Agency, 2016), which are sometimes used for brumation by freshwater turtles. Sediment contaminated with dilbit would have led to additional exposure to weathered dilbit for several months during turtles' brumation period. In addition, lower flow depositional areas of the river were disturbed by oil recovery efforts including sediment agitation and dredging in 2010 and 2011.

While we found non-rehabilitated turtles had higher mortality than rehabilitated or overwintered turtles during the first year following the spill, 8+ years later the differences in monthly survival probabilities among survivors in the different rehabilitation categories were indistinguishable. In the 8–11 years post-spill, we found the differences in survival rates was best explained by sex, with females having higher monthly survival rates compared to males. This trend is similar in other populations of northern map turtles not impacted by oil (Bulté and Blouin-Demers, 2009; Bulté et al., 2009). Our monthly survival estimates (which included juveniles and subadults) were higher than the annual adult survival rates from a six-year study of an intact reference population of northern map turtles in Canada (94% for females and 81% for males; Bulté et al., 2009). Although the survival rates in our study population cannot be directly compared to those of the Bulté et al. (2009) study due to differences in the age classes included in the estimates, our results suggest that the map turtle population at our Study Site has returned to a "natural" mortality rate ~10 years after the 2010 oil spill. In comparison to long-lived species such as the northern map turtle, taxa with shorter generation times can likely recover more quickly following environmental disasters such as oil spills. For example, invertebrates in oil-impacted areas of the Kalamazoo River decreased in density and species diversity during 2010 and 2011 but appear to have recovered and stabilized within five years of the spill (Matousek, 2018).

Our study 11 years after the 2010 Kalamazoo River oil spill suggests that dilbit exposure, combined with other stressors from spill response and habitat restoration actions, may cause mortality to freshwater turtle species similar to that resulting from spills of conventional crude oil. Rehabilitation of oil-exposed northern map turtles significantly increased survival within 14 months of the spill, which emphasizes the importance and effectiveness of rehabilitation efforts for species such as freshwater turtles. While the same increase in survival probability was no longer apparent 8–11 years post-spill, nearly 25% of rehabilitated turtles were nonetheless recaptured during this time period, which is an impressive survival rate in a population that was severely impacted by a massive oil spill.

5. Conclusions

With the predicted increase in dilbit production and transport in the near future, research should concentrate on determining specific pathways of dilbit toxicology in turtles and other wildlife, its residence time in tissues and potential for biomagnification at higher trophic levels, and the effects of long-term exposure to individuals and populations. It is also important to determine the potential impacts of physical emergency response and habitat restoration actions such as sediment agitation on habitat quality and long-term population recovery. Finally, determining the specific rehabilitation activities that are most effective at increasing survival of oiled animals is the next logical step. Our results demonstrated that overwintering turtles in captivity resulted in increased survival rates; therefore, future research should endeavor to compare the efficacy of different overwintering strategies, such as keeping turtles fed, warm, and awake throughout the winter vs. inducing them to hibernate in captivity. Empirically testing the effectiveness of specific wildlife rehabilitation strategies, emergency spill responses, and habitat restoration protocols is critical for developing best management practices in order to ensure the survival of long-lived wildlife species, such as turtles, following large-scale spill events.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2022.119968.

Refrences

Alderman, S.L., Lin, F., Farrell, A.P., Kennedy, C.J., Gillis, T.E., 2016. Effects of diluted bitumen exposure on juvenile sockeye salmon: from cells to performance. Environ. Toxicol. Chem. 36, 354–360.

Alsaadi, F.M., Madison, B.N., Brown, R.S., Hodson, P.V., Langlois, V.S., 2018. Morphological and molecular effects of two diluted bitumens on developing fathead minnow (*Pimephales promelas*). Aquat. Toxicol. 204, 107–116.

Baker, L., Edwards, W., Pike, D.A., 2015. Sea turtle rehabilitation success increases with body size and differs among species. Endanger. Species Res. 29, 13–21.

Barron, M.G., 2012. Ecological impacts of the Deepwater Horizon oil spill: implications for immunotoxicity. Toxicol. Pathol. 40, 315–320.

Barron, M.G., Conmy, R.N., Holder, E.L., Meyer, P., Wilson, G.J., Principe, V.E., Willming, M.M., 2018. Toxicity of cold lake blend and wester Canadian select dilbits to standard aquatic test species. Chemosphere 191, 1–6.

Barron, M.G., Vivian, D.N., Heintz, R.A., Yim, U.H., 2020. Long-term ecological impacts from oil spills: comparison of Exxon Valdez, hebei spirit, and deepwater Horizon. Environ. Sci. Technol. 54, 6456–6467.

Barron, M.G., Moso, E.M., Conmy, R., Meyer, P., Sundaravadivelu, D., 2021. Toxicity of sediment oiled with diluted bitumens to freshwater and estuarine amphipods. Mar. Pollut. Bull. 163, 111941.

Bulté, G., Blouin-Demers, G., 2009. Does sexual bimaturation affect the cost of growth and the operational sex ratio in an extremely size-dimorphic reptile? Ecoscience 16, 175–182.

Bulté, G., Carriere, M.-A., Blouin-Demers, G., 2009. Impact of recreational power boating on two populations of northern map turtles (*Graptemys geographica*). Aquat. Conserv. 20, 31–38.

Cagle, F.R., 1939. A system of marking turtles for future identification. Copeia 1939 170–173.

Congdon, J.D., Nagle, R.D., Kinney, O.M., van Loven Sels, R.C., Quinter, T., Tinkle, D.W., 2003. Testing hypotheses of aging in long-lived painted turtles (*Chrysemys picta*). Exp. Gerontol. 38, 765–772.

Cormack, R.M., 1964. Estimates of survival from the sighting of marked animals. Biometrika 51, 429–438.

Camacho, M., Boada, L.D., Orós, J., Calabuig, P., Zumbado, M., Luzardo, O.P., 2012. Comparative study of polycyclic aromatic hydrocarbons (PAHs) in plasma of Eastern Atlantic juvenile and adult nesting loggerhead sea turtles (*Caretta caretta*). Mar. Pollut. Bull. 64, 1974. –1980.

Choquet, R., Lebreton, J.D., Gimenez, O., Reboulet, A.M., Pradel, R., 2009. U-CARE: utilities for performing goodness of fit tests and manipulation Capture-Recapture data. Ecography 32, 1071–1074.

Crosby, S., Fay, R., Groark, C., Kani, A., Smith, J.R., Sullivan, T., Pavia, R., 2013. Transporting Alberta oil sands products: defining the issues and assessing the risks. In: US Department of Commerce. NOAA Technical Memorandum: Seattle. WA.

De La Cruz, S.E., Takekawa, J.Y., Kyle, A., Spragens, A., Yee, J., Golightly, R.T., Massey, G., Henkel, L.A., Larsen, R.S., Ziccardi, M., 2013. Post-release survival of surf scoters following an oil spill: an experimental approach to evaluating rehabilitation success. Mar. Pollut. Bull. 67, 100–106.

Dew, W.A., Hontela, A., Rood, S.B., Pyle, G.G., 2015. Biological effects and toxicity of diluted bitumen and its constituents in freshwater systems. J. Appl. Toxicol. 35, 1219.

Dunnet, G.M., 1982. Oil Pollution and Seabird Populations, vol. 297. Philosophical Transaction of the Royal Society B, pp. 413–427.

Environmental Protection Agency (EPA), 2016. FOSC Desk Report for the Enbridge Line 6b Oil Spill Marshall, p. 241. Michigan. April 2016.

Ernst, C.H., Lovich, J.E., 2009. Turtles of the United States and Canada, second ed. John Hopkins University Press, Baltimore, Maryland, USA.

Esler, D., Ballachey, B.E., Matkin, C., Cushing, D., Kaler, R., Bodkin, J., Monson, D., Esslinger, G., Kloecker, K., 2018. Timelines and mechanisms of wildlife population recovery following the Exxon Valdez oil spill. Deep Sea Res., Part II 147, 36–42.

Everitt, S., Fujita, K.K., MacPherson, S., Brinkmann, M., Pyle, G.G., Wiseman, S., 2021. Toxicity of weather sediment-bound dilbit to early life stages of zebrafish (*Danio rerio*). Environ. Sci. Technol. 55, 1721–1729.

Gonzalez-Tokman, D.M., Munguia-Steyer, R.E., Gonzalez-Santoyo, I., Baena-Diaz, F.S., Cordoba-Aguilar, A., 2012. Support for the immunocompetence handicap hypothesis in the wild: hormonal manipulation decreases survival in sick damselflies. Evolution 66, 3294–3301.

Hartung, R., 1995. Assessment of the potential for long-term toxicological effects of the Exxon Valdez oil spill on wildlife. Exxon Valdez Oil Spill: Fate and Effects in Alaskan Waters. American Society for Testing Materials, Philadelphia, pp. 693–725.

Helm, R.C., Carter, H.R., Ford, R.G., Fry, D.M., Moreno, R.L., Sanpera, C., Tseng, F.S., 2015. Overview of efforts to document and reduce impacts of oil spills on seabirds. In: Fingas, M. (Ed.), Handbook of Oil Spill Science and Technology. John Wiley & Sons, Inc, Hoboken, New Jersey, pp. 431–453.

Henkel, L.A., Ziccardi, M.H., 2018. Life and death: how should we respond to oiled wildlife? J. Fish and Wildlife Manag. 9, 296–301.

Heyes, A., Leach, A., Mason, C.F., 2018. The economics of Canadian oil sand. Rev. Environ. Econ. Pol. 12, 242–263.

Hinton, D.E., Kullman, S.W., Hardman, R.C., Volz, D.C., Chen, P.J., Carney, M., Bencic, D.C., 2005. Resolving mechanisms of toxicity while pursuing ecotoxicological relevance? Mar. Pollut. Bull. 51, 635–648.

Hodson, P.V., 2017. The toxicity to fish embryos of PAH in crude and refined oils. Arch. Environ. Contam. Toxicol. 73, 12–18.

Honda, M., Suzuki, N., 2020. Toxicities of polycyclic aromatic hydrocarbons for aquatic animals. Int. J. Environ. Res. Publ. Health 17, 1363.

Hopkins, B.C., Willson, J.D., Hopkins, W.A., 2013. Mercury exposure is associated with negative effects on turtle reproduction. Environ. Sci. Technol. 47, 2416–2422.

Hua, Y., Mirnaghai, F.S., Yang, Z., Hollebone, B.P., Brown, C.E., 2018. Effect of evaporative weather and oil-sediment interactions on the fare and behavior of diluted bitumen in marine environments. Part 1. Spill-related properties, oil buoyancy, and oil-particulate aggregates characterization. Chemosphere 191, 1038–1047.

Iverson, J.B., 1988. Growth in the common map turtle, *Graptemys geographica*. Trans. Kans. Acad. Sci. 91, 153–157.

Iverson, S.A., Esler, D., 2010. Harlequin duck population injury and recovery dynamics following the 1989 Exxon Valdez oil spill. Ecol. Appl. 20, 1993. –2006.

Jessup, D.A., 1998. Rehabilitation of oiled wildlife. Conserv. Biol. 12, 1153–1155.

Jolly, G.M., 1965. Explicit estimates from capture-recapture data with both death and immigration - stochastic model. Biometrika 52, 225–247.

King, M.D., Elliot, J.E., Williams, T.D., 2020. Effects of petroleum exposure on birds: a review. Sci. Total Environ. 755, 1–31.

King, T.L., Robinson, B., Boufadel, M., Lee, K., 2014. Flume tank studies to elucidate the fate and behavior of diluted bitumen spilled at sea. Mar. Pollut. Bull. 83, 32–37.

Lager, K.F., 1943. Food habits and economic relations of the turtles of Michigan with special reference to fish management. Am. Midl. Nat. 29, 257–312.

Lindeman, P.V., 2013. The Map Turtle and Sawback Atlas: Ecology, Evolution, Distribution, and Conservation. University of Oklahoma Press, Norman, OK, USA.

Madison, B.N., Hodson, P.V., Langlois, V.S., 2015. Diluted bitumen causes deformities and molecular responses indicative of oxidative stress in Japanese medaka embryos. Aquat. Toxicol. 165, 222.

Madison, B.N., Hodson, P.V., Langlois, V.S., 2017. Cold Lake Blend diluted bitumen toxicity to the early development of Japanese medaka. Environ. Pollut. 225, 579–586.

Matousek, J.J., 2018. A Biological Survey of Sites on Talmadge Creek Near the Enbridge Oil Spill in Marshall, Calhoun County, Michigan. Michigan Department of Environmental Quality, Lansing, MI. March 2018 (MI/DEQ/WRD-18/006).

Mignucci-Giannoni, A.A., 1999. Assessment and rehabilitation of wildlife affected by an oil spill in Puerto Rico. Environ. Pollut. 104, 323–333.

Monson, D.H., Doak, D.F., Ballachey, B.E., Bodkin, J.L., 2011. Could residual oil from the Exxon Valdez spill create a long-term population "sink" for sea otters in Alaska? Ecol. Appl. 21, 2917–2932.

Moore, M., Early, G., Touhey, K., Barco, S., 2007. Rehabilitation and release of marine mammals in the United States: risks and benefits. Mar. Mamm. Sci. 23, 731–750.

Murphy, D., Gemmell, B., Vaccari, L., Li, C., Bacosa, H., Evans, M., Gemmell, C., Harvey, T., Jalali, M., Niepa, T.H.R., 2016. An in-depth survey of the oil spill literature since 1968: long term trends and changes since Deepwater Horizon. Mar. Pollut. Bull. 113, 371–379.

Nagle, R.D., Rowe, C.W., Congdon, J.D., 2001. Accumulation and selective maternal transfer of contaminants in the turtle *Trachemys scripta* associated with coal ash deposition. Arch. Environ. Contam. Toxicol. 40, 531–536.

National Transportation Safety Board (NTSB), 2012. Enbridge Incorporated, Hazardous Liquid Pipeline Rupture and Release. Marshall, Michigan, p. 164. July 25, 2010.

Newman, S.H., Ziccardi, M.H., Berkner, A.B., Holcomb, J., Clumpner, C., Mazet, J.A.K., 2003. A historical account of oiled wildlife care in California. Mar. Ornithol. 31, 59–64.

Otten, J.G., 2022. Long-term Impacts of a Freshwater Oil Spill on an Aquatic Turtle Species. PhD Dissertation, University of Toledo, Toledo, OH.

Peterson, C.H., Rice, S.D., Short, J.W., Esler, D., Bodkin, J.L., Ballachey, B.E., Irons, D.B., 2003. Long-term ecosystem response to the Exxon Valdez oil spill. Science 302, 2082–2086.

Philibert, D.A., Philibert, C.P., Lewis, C., Tierney, K.B., 2021. Comparison of diluted bitumen (dilbit) and conventional crude oil toxicity to developing zebrafish. Environ. Sci. Technol. 506091–506098.

Pradel, R., Wintrebert, C.M.A., Gimenez, O., 2003. A proposal for a goodness-of-fit test to the Arnason-Schwarz multisite capture-recapture model. Biometrics 59, 43–53.

Rebar, A.H., Libscomb, T.P., Harris, R.K., 1995. Clinical and clinical laboratory correlates in sea otters dying unexpectedly in rehabilitation centers following the Exxon Valdez oil spill. Vet. Pathol. 32, 346–350.

Reynaud, S., Deschaux, P., 2006. The effects of polycyclic aromatic hydrocarbons on the immune system of fish: a review. Aquat. Toxicol. 77, 229–238.

Robidoux, P.Y., Virginie, B., Judith, L., Marc, D., 2018. Assessment of acute and chronic toxicity of unweather and weathered diluted bitumen to freshwater fish and invertebrates. Ecotoxicol. Environ. Saf. 164, 331–343.

Ruberg, E.J., King, M.D., Elliot, J.E., Tomy, G.T., Idowu, I., Vermette, M.L., Williams, T. D., 2022. Effects of diluted bitumen exposure on the survival, physiology, and behaviour of zebra finches (*Taeniopygia guttata*). Ecotoxicol. Environ. Saf. 229, 113071.

Saba, V.S., Spotila, J.R., 2003. Survival and behavior of freshwater turtles after rehabilitation from an oil spill. Environ. Pollut. 126, 213–223.

Seber, G.A.F., 1965. A note on the multiple recapture census. Biometrika 52, 249–259.

Seivwright, K.A., Battley, P.F., McConnell, H., Armstrong, D.P., Morgan, K.J., 2019. Post-release breeding success of oil-rehabilitated and non-rehabilitated little blue penguins, *Eudyptula minor*, following the M/V Rena oil spill, New Zealand. Mar. Pollut. Bull. 146, 317–325.

Stacy, B.A., 2015. Summary of Findings for Sea Turtles Documented by Directed Captures, Stranding Response, and Incidental Captures under Response Operations during the BP Deepwater Horizon (Mississippi Canyon 252) Oil Spill. With 2015 Update: Summary of Stranded Sea Turtles Reported as Oiled within the Original BP Deepwater Horizon Response Area from October 21, 2010 through 2015. Administrative Record for the Deepwater Horizon Oil Spill, 5 Preassessment/ Assessment, 5.7 Sea Turtle Injury, 5.7.2 Technical Reports. United States Department of Commerce, NOAA, NMFS, Silver Spring, Maryland, USA.

Stacy, N.I., Field, C., Staggs, L., Maclean, R.A., 2017. Clinicopathological findings in sea turtles assessed during the BP Deepwater Horizon oil spill response. Endanger. Species Res. 33, 25–37.

Steen, D.A., Van Dyke, J.U., Jackson, B.P., Hopkins, W.A., 2015. Reproduction and hatchling performance in freshwater turtles associated with a remediated coal fly-ash spill. Environ. Res. 138, 38–48.

Timlick, L., Peters, L.E., Wallace, S., Dettman, H., Brown, R.S., Mason, J., Langlois, V., Palace, V., 2020. Effects of environmentally relevant residual levels of diluted bitumen on wild fathead minnows (*Pimephales promelas*). Bull. Environ. Contam. Toxicol. 105, 3–704.

White, G.C., Burnham, K.P., 1999. Program MARK: survival estimation from populations of marked animals. Bird Studies 46, 120–139.

Wilkin, S.A., Rowles, T.K., Stratton, E., Adimey, N., Field, C.L., Wissmann, S., Shigenaka, G., Fourgeres, E., Mase, B., 2017. Southeast region stranding network, Ziccardi, M.H. Marine mammal response operation during Deepwater Horizon oil spill. Endangered Species Research 33, 107–118.

Wolfaardt, A.C., Underhill, L.G., Altwegg, R., Visagie, J., 2008. Restoration of oiled African penguins (*Spheniscus demersus)* a decade after the Apollo Sea spill. Afr. J. Mar. Sci. 30, 421–436.

Ylitalo, G.M., Collier, T.K., Anulacion, B.F., Juaire, K., Boyer, R.H., da Silva, D.A.M., Keene, J.L., Stacy, B.A., 2017. Determining oil and dispersant exposure in sea turtles from the northern Gulf of Mexico resulting from the Deepwater Horizon oil spill. Endanger. Species Res. 33, 9–24.

Yu, S., Halbrook, R.S., Sparling, D.W., Colombo, R., 2011. Metal accumulation and evaluation of effects in a freshwater turtle. Ecotoxicology 20, 1801–1812.