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Legacy effects of land cover and land use on painted turtle (Chrysemys picta) populations over a 20-year period in southeastern New Hampshire

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ABSTRACT

More than half of all freshwater turtles and tortoises are threatened with extinction. Human land use practices and land cover conversion often decrease adult turtle survivorship and dampen the reproductive potential of turtle populations. The ability to make informed conservation and management decisions depends on understanding the relative impacts of different landscape contexts on turtle populations, and the spatial extent over which they have an impact. We surveyed painted turtle (*Chrysemys picta*) populations along an urbanization gradient in southeastern New Hampshire, USA to assess the relative impact of land cover, land use, and chemical contaminants on population density and sex ratio. We compared the findings from our comprehensive field surveys (2019-2021; “modern”) to earlier surveys (2001-2001; “historic”) conducted at identical and nearby ponds. In both time periods, population density increased with proportion of wetland and decreased with forest cover. Modern turtle density was best explained by historic land cover suggesting a lag between land cover change and a detectable impact on the population. Modern road length negatively influenced modern population density whereas historic road length did not have a measurable impact on density at that time. In both time periods we found increased forest cover associated with male-skewed populations. We tested 30 painted turtle blood sampled for 63 chemical contaminants. Organochlorine pesticides and synthetic pyrethroids were pervasive in turtle blood but not detected in water samples suggesting these chemicals are bioaccumulating in turtles despite low environmental concentrations. Our findings illustrate that effective management of turtle populations requires consideration of legacy effects of land use and land cover conversion as well as direct sources of mortality.

INTRODUCTION

Human activity disrupts ecosystems through habitat loss and degradation, leading to rates of species loss not seen since the last mass extinction (Barnosky et al. 2011; Pimm et al. 2014; Watson et al. 2016). Land-cover conversion disproportionately impacts habitat specialists with low reproductive potential (DeStefano & DeGraff 2003; Eskew et al. 2010). One such group of species are freshwater turtles, which require a mosaic of aquatic and terrestrial habitats to complete their life cycle and have low reproductive potential due to the life history traits associated with long-lived organisms. Long-lived organisms have low juvenile survivorship and delayed sexual maturity that require high adult survival rates to maintain a stable population (Congdon et al. 1993; Heppell 1998). Conversion of natural land cover, exposure to chemical contamination, and the expansion of the road networks have the potential to decrease adult turtle survivorship and further dampen reproductive potential, placing freshwater turtle populations at increased risk.

Changes to terrestrial vegetation cover surrounding a pond can alter microhabitat conditions of nesting grounds, which can impact freshwater turtle embryo development and result in skewed sex ratios. Most freshwater turtles exhibit Type 1a temperature dependent sex determination, which results in more female hatchlings at higher incubation temperatures (Bull & Vogt 1979). The decrease in vegetative cover common throughout human-dominated landscapes can increase incubation temperatures which has resulted in a positive association between the proportion of female turtles and urbanization (Bowne et al. 2018). To complicate matters further, turtles may be exposed to chemicals from agricultural and residential sources that accumulate in soil and surface water. Many chemical pollutants have endocrine disrupting properties and thus the potential to skew population sex ratios through the disruption of sex hormones (Willingham 2005). Endocrine-disruption can also decrease the reproductive potential of the population through increased proportions of unhatched eggs (Jain et al. 2012) and the feminization of male gonads (Hayes et al. 2011).

Turtle population size and structure are also greatly impacted by vehicle collisions. Road mortality is of particular concern for slow-moving species like turtles (González-Suárez et al. 2018), and female turtles are especially vulnerable to road mortality due to their annual nesting migrations when they leave the pond in search of nesting areas on land (Haxton 2000b). The disproportionate loss of reproductive females may result in male-biased populations with reduced annual recruitment rates (Garber & Burger 1995). An analysis of freshwater turtle population sex-ratios across the United States found that the proportion of males in a population increased linearly with the expansion of the road network from 1930 to 2003 (Gibbs & Steen 2005). Urbanization brings additional threats as migrating turtles are susceptible to predation by red foxes (*Vulpes vulpes*) (Spencer 2002; Spencer & Thompson 2005; Dawson et al. 2016). Red foxes often achieve their highest densities in close association with humans (Lombardi et al. 2017; Campbell et al. 2020) and represent another threat to turtles migrating across an urbanized landscape. Red foxes will also depredate turtle nests (Burger 1977; Macdonald et al. 1994; Riley & Litzgus 2014), along with other generalist predators such as raccoons (*Procyon lotor*) (Burger 1977; Congdon et al. 1983, 1987; Christens & Bider 1987), and striped skunks (*Mephitis mephitis*) (Snow 1982; Clarke & Gruenig 2003). Together, additive mortality from vehicle collisions and increased predator presence poses a significant threat to turtle populations.

The ability to make informed conservation and management decisions depends on an understanding of the relative impacts of different land cover and land use types and the spatial scale at which they have an impact (Noss 1992; Willis & Whittaker 2002; Wiens 2009). In New Hampshire, USA, four of the seven native turtle species are classified as 'species in greatest need of conservation', with habitat degradation listed as the primary threat (New Hampshire Fish and Game 2015; Dijk & Rhodin 2019). Here, we use a survey-resurvey approach to assess the impact of land cover and land use on painted turtle (*Chrysemys picta*) population dynamics over a 20-year period in southeastern New Hampshire. Specifically, our objectives were to:

1. Assess the effect of land cover and land use on historical and modern painted turtle density, and the spatial extent over which they have an impact.
2. Assess the effect of land cover and land use on historical and modern painted turtle sex ratio.
3. Investigate the presence of time lags between a disturbance and a detectable response in painted turtle population density and sex ratio.
4. Compare results between the two time periods to understand how painted turtle populations and the factors that drive them have changed over 20 years.

We focused on the painted turtle since it is found in high enough densities to facilitate robust comparisons and shares many of the same habitat requirements of the rarer and more threatened turtle species. Moreover, their relatively fast generation time compared to co-occurring turtle species makes them a good early indicator of the response of freshwater turtles to environmental change. Our findings provide insight into the factors that shape freshwater turtle populations over time, specifically the relative influences of historic and modern landscape context, and can inform conservation and management of freshwater turtles in the region.

METHODS

Study System & Data Collection

Over the past 60 years, southeastern New Hampshire, USA, has undergone rapid development and population growth. With the completion of a new interstate highway system in the 1960's the urban sprawl of Boston, began to transform New Hampshire from a historically slow-growing region into the fastest growing state in the Northeast (Wallace 2007). The Greater Boston metropolitan area is the tenth largest urbanized area in the country with more than 4.5 million residents (US Census Bureau 2012). More than 266,000 people immigrated to New Hampshire during the 1970's, 1980's, and 1990's, accounting for 53% of the population growth during that period (Johnson 2012). More than 3,200 lane miles of new roads were constructed in New Hampshire between 1980 and 2007 (Federal Highway Administration 2014). The housing developments, businesses, and farms built to accommodate this growing population removed valuable wildlife habitat and left what remains fragmented by roads.

We used survey data on turtle population density and sex ratio collected during two time periods separated by a 20-year interval ("historic": 2000-2001, "modern": 2019-2021). The historic data come from a survey of 32 ponds along an urbanization gradient in southeastern New Hampshire to determine the impacts of landscape composition on painted turtle populations (Marchand & Litvaitis 2004). We received landowner permission to resurvey 24 of the ponds from the original survey. An additional seven ponds were selected to ensure sufficient sample sizes of ponds with varying land cover compositions. All ponds ranged in size from 0.28 hectares to 4.89 hectares and were considered independent of each other as they were separated by at least 0.52 kilometers (Fig 1).

Following Marchand and Litvaitis (2004), we sampled each pond in the modern time period four times between May and September of a single year. Each sampling period used five hoop traps and lasted two days, with traps checked 24 and 48 hours after being set. We spaced traps approximately 20-meters apart along the shallow margins of the pond and baited traps with canned cat food. We gave each newly captured turtle a unique identifier by notching several marginal scutes (Nagle et al. 2017). We also placed each turtle into a stage class (adult or sub-adult) and determined the sex using foreclaw and pre-cloacal measurements. Stage class was determined by carapace length; turtles with a carapace length greater than or equal to 90mm were considered adults (Marchand & Litvaitis 2004). We released all turtles at the capture site immediately following data collection. Trapping procedures were approved by the University of New Hampshire Animal Care and Use Committee (190406, 200503) (Appendix A).

To assess exposure and susceptibility to common chemical contaminants, we collected a one-liter composite water sample from every pond and blood samples from 30 painted turtles across 12 ponds that varied in their land use and land cover classification. The ponds selected included the highest proportion of forest, development, agriculture, and the highest human population density of all the study ponds. We collected all blood and water samples in 2021 with all water samples collected during July. The composite water sample comprised three subsamples that we collected at varying depths (next to the substrate, in the middle of the water column, and just below the surface). We collected subsamples on the same day and away from water sources (Decker & Simmons 2013). We collected blood samples (ca. 2ml) from the dorsal coccygeal vein in the tail of adult turtles using a sterile 24-g needle following the protocols laid out by the Northeast Wood Turtle Working Group (2018). We froze all blood and water samples on the same day as collection.

All blood and water samples were tested for 65 different chemical contaminants spanning three major groups: synthetic pyrethroids, organophosphate pesticides, and organochlorine pesticides (Table 1). Testing was conducted by Michigan State University Veterinary Diagnostics Laboratory (MSU VDL) using gas chromatography-mass spectrometry with specific assays for each chemical compound that allowed for quantification of the chemical concentration of each contaminant. This testing method has a detection limit of 10 ppb for organochlorine pesticides and 1000 ppb for synthetic pyrethroids and organophosphate pesticides.

At each pond we quantified land cover and land use covariates within 2000-meters from the pond perimeter using QGIS (QGIS Geographic Information System, v2.34.3, QGIS Association). We further discretized the 2,000-meter buffers into concentric 50-meter wide, non-overlapping rings to allow for a distance-weighted smoothing analysis (Aue et al. 2012; Chandler & Hepinstall-Cymerman 2016; Miguet et al. 2016, 2017; Moll et al. 2020). We isolated pond shapefiles using the National Wetlands Inventory dataset (U.S. Fish & Wildlife Service 2020) and verified using Google satellite imagery (Google 2022). We quantified the land cover variables for three different time periods, 1970-1980, 2001, and 2019. Covariates included the proportion of eight cover classes (forest, wetlands, agriculture, open water, open/disturbed, shrubland, low-intensity development, and high-intensity development) derived from the National Land Cover Database (NLCD) by grouping the 20 cover types into physiognomic categories (U.S. Geological Survey 2003; Dewitz 2021). The 2001 and 2019 land cover covariates were extracted directly from the NLCD. The 1970-1980 data were compiled from the

Enhanced Historical Land Cover and Land Use Dataset which used historical USGS and NASA imagery to recreate the NLCD land cover categories for that time period (Price et al. 2006). For the 2001 and 2019 time periods we also quantified three land use covariates within the same 2,000-meter buffer: road length (kilometers), traffic volume (vehicles per day), and human population density (people/km^2). Road length and traffic volume data for 2001 and 2019 were available through the NH Department of Transportation (NH Department of Transportation 2001, 2019). Human population density at 100m^2 resolution for 2001 and 2019 came from WorldPop (WorldPop 2020a, 2020b). These land use data were not available for the 1970-1980 time period.

Data Analysis

To analyze sex ratio, we compared the proportion of males across time periods and used a binomial significance test to determine if any of the historic or modern turtle populations had a male or female bias. The binomial significance test calculates the probability that the observed sex ratio came from a population with a 50/50 male to female distribution (Hardy 2002). We considered p-values of less than 0.05 to indicate sufficient evidence to reject the null hypothesis and conclude that the population sex ratio is skewed. This test is appropriate for small sample sizes because the model considers both the proportion of males and the sample size. However, small sample sizes will not have the statistical power to detect small deviations from the expected sex ratio.

To estimate abundance, we used a closed-population capture-mark-recapture model with time dependent detection probability. We fit the model using a Bayesian approach with parameter-expanded data augmentation (Kéry 2010) using JAGS (Plummer 2003) in R (version

4.1.2) via R2jags (Yu-Sung & Yajima 2012). We ran the models using three chains and chain convergence was assessed using Rhat values, where an Rhat value less than 1.05 indicates good convergence (Kery 2010). We ran the models for 2,500 iterations with a burn in period of 500 iterations and a thinning rate of two. We visually assessed traceplots to confirm convergence and to confirm that the burn in value was sufficient to eliminate the effect of initial values. All priors were diffuse and uninformative to avoid introducing bias into the results (Appendix C.1). We considered four other capture-mark-recapture models: a closed-population null model, a closed-population random individual effects model, a closed population time and random individual effects model, and a Jolly-Seber open-population model. Model selection using DIC suggested that the closed-population model with time dependent detection probability had the most support (Table 2). We estimated historic turtle abundance using the capture data from 2000-2001 (Marchand & Litvaitis 2004) and modern turtle abundance using the capture data from 2019-2021. To standardize abundance across ponds of varying size, we converted both historic and modern turtle abundance to density (turtles/hectare) for use in our landscape models.

Following Moll et al (2020), we used a distance-weighted smoothing model to assess the effects of land cover and land use on turtle density (Aue et al. 2012; Chandler & Hepinstall-Cymerman 2016; Miguet et al. 2017). We developed separate models for the historic and modern time periods. This hierarchical model estimates the effect of a given covariate as well as its maximal scale of effect (i.e., the spatial extent at which that covariate is having the greatest effect on turtle density). The model uses a distance-weighted smoothing function to down-weight the effect of a covariate as distance from the pond perimeter increases.

We calculated a distance-weighted representation of each covariate at each pond by first discretizing the 2,000-meter buffer surrounding each pond into 50-meter-wide non-overlapping concentric rings and quantifying each of the eight land cover and the three land use covariates within each ring. The weight of each ring is then determined by a Gaussian smoothing kernel:

$$W_m = \frac{exp\left(-\frac{d_{mj}^2}{2\sigma^2}\right) * A_m}{\sum_m exp\left(-\frac{d_{mj}^2}{2\sigma^2}\right) * A_m}$$

Where W_m is the weight of the ring, d_{mj} is the distance between pond j and ring m, A_m is the area of the ring, and σ is the scaling parameter. The scaling parameter is estimated by the model and determines how quickly the effect of a covariate will diminish with distance. A smaller scaling parameter results in a sharp decrease in effect as distance from the pond increases and is typically associated with smaller bodied and less mobile species (Thornton and Fletcher 2014; Concepción and Díaz 2011).

We then calculated the distance-weighted representation for each covariate at each location by multiplying the value of the covariate in a ring by the weight of that ring, and then summing all the rings across a pond. Using the proportion of wetlands (WET) as an example:

$$WETw_{(j)} = \sum_m W_m * WET_{m(j)}$$

Where $WETw_{(j)}$ is the distance-weighted representation of the proportion wetlands at pond j. Wm is the weight of ring m. $WETm_{(j)}$ is the proportion of wetlands in ring m at pond j.

We then used the distance-weighted representation of each covariate in a log normal turtle density model. Continuing with the proportion of wetlands as an example, this model took the form:

$$N_i \sim Normal(\mu_i, \tau)$$

$$\mu_i = \beta 0 + \beta 1 * WETw_i$$

Where, N_i represents the turtle density (turtles/hectare) at pond i, as estimated by the closed-population mark-recapture model. N_i is normally distributed with a mean of μ_i, and a variance of τ. The mean at pond i (μ_i) is equal to the intercept ($\beta 0$) plus the distance-weighted representation of the proportion wetlands multiplied by the effect of wetlands ($\beta 1$).

Given the sample size of ponds (historic n = 32, modern n = 31), we used a stepwise procedure with univariate models to arrive at a final model for inference, whereby we retained all covariates that had an effect. We considered a covariate to have an effect if the 90% credible interval did not overlap zero. We tested all combinations of retained covariates while limiting the number of covariates in a single model to three due to modest sample sizes. We used a Pearson correlation to assess collinearity between covariates and only included non-correlated covariates in any single model. We ran separate models for the historic and the modern turtle density. For historic turtle density we tested contemporary land cover (2001) and land cover from 1970-1980. For modern turtle density, we considered current land cover covariates (2019) and tested the effect of the historic (2001) land cover covariates. We did this because previous research on freshwater turtles suggests a lag time between the introduction of a disturbance and a measurable effect on abundance due to their long lifespans (Garber & Burger 1995). In both the modern and historic density models we only considered the contemporary land use covariates since they were selected to represent sources of direct mortality. With direct mortality, we expected a more immediate population response than from land cover change which primarily influences the reproductive potential of the population.

Similar to the abundance models, we fit all landscape models to the data using the Bayesian approach described above. All priors were diffuse and uninformative (Appendix C.2). We ran the models for 5,000 iterations with a burn in of 500 iterations and a thinning rate of one. We assessed the model's goodness of fit using a Bayesian P-value. The Bayesian P-value is a summed squares of residuals goodness of fit test where the deviation about the mean from the observed data is compared to the deviation about the mean of expected data generated by the model. A Bayesian-P value of 0.5 indicates perfect fit. We performed model selection using DIC. When paired with diffuse and uninformative priors, Bayesian DIC performs similarly to AIC under frequentist analysis (Ellison 2004). DIC estimates the effective number of parameters in the model which makes it especially useful for complex models such as those with random effects or a hierarchical structure (Ward 2008; Wilberg & Bence 2008). We averaged parameter estimates across all models within two DIC units of the top model by calculating the mean and 95% confidence intervals across all iterations for each parameter (Hooten and Hobbs 2015). The resulting averaged parameter estimates are slightly less precise than any single model but better capture the uncertainty inherent in the model selection process (Ellison 2004).

For sex ratio, we used a generalized linear model to assess the effects of land cover and land use on the proportion of males in the population. We log transformed the proportion data in order to constrain model predictions to biologically possible outcomes (Hardy 2002). We constrained land cover and land use covariates to 500-meters, the maximum scale at which any given covariate was estimated to have an impact on the population under the distance-weighted smoothing density model. We used the same stepwise univariate approach here as above to arrive at a final model. We excluded ponds with less than ten unique captures because estimation of sex ratio based on so few captures would be unreliable. AICc was used to select between models. We interpreted the strongest model as that having the greatest AICc weight, recognizing that unnested models with delta AICc less than two are often considered equivalent (Aho et al. 2014).

For chemicals that were detected in turtle blood, we used generalized linear models to assess the relationship between the chemical concentrations and land cover and land use. We used the same 500-meter constrained covariates as the sex ratio analysis and used a univariate approach. We used R^2 to assess the strength of the relationship and p-values to assess statistical significance. Since there is no historic chemical contaminant data, we performed this analysis for the modern time period only using modern land cover and land use covariates.

RESULTS

Population Demographics

Between 2019 and 2021, we captured 1,694 turtles, comprising 1,285 unique captures and 409 recaptures across five species (Table 3). Painted turtles were the most common species by far (84% of all captures). Snapping turtles (*Chelydra serpentina*) were the second most abundant followed by the rarer species, Blanding's turtles (*Emydoidea blandingii*), musk turtles (*Sternotherus odoratus*), and spotted turtles (*Clemmys guttata*). The average recapture rate was 31.8% across all species and 33.9% for painted turtles.

We modeled historic (2000-2001) turtle abundance estimates for each of the 32 ponds from the original survey (Marchand and Litvaitis 2004) (Table 4). Abundance estimates ranged from five to 189 turtles with an average population size of 53 turtles. Average detection probability ranged from 0.18 to 0.25 across all 4 historic sampling occasions. The abundance estimates for the 31 ponds sampled in the modern period (2019-2021) ranged from five to 123 turtles with an average population size of 38 turtles (Table 5). Average detection probability ranged from 0.16 to .28 across the modern sampling occasions.

We compared the abundance estimates for the 24 ponds that were sampled in both time periods (Table 6). We considered the population size to have changed if the 95% credible interval for the abundance estimate in each time period did not overlap with one another. We found that nine ponds experienced a decrease in population size, while two ponds experienced an increase, and 13 ponds had no change in population size (Fig. 6).

Average sex ratio in the historic period was 0.63 and ranged from 0.3 to 1. The modern sex ratio ranged from 0.33 to 0.86 with an average of 0.56. The binomial significance tests indicated there were six historic ponds with a skewed sex ratio and five modern ponds with a skewed sex ratio (Table 7).

Land Cover and Land Use Effects

We quantified land cover at each pond and found that forest cover varied the most across ponds in both the historic (Fig. 2) and modern (Fig. 3) time periods. The historic proportion of forest ranged from 7.6% to 87.6% with an average of 58%. The modern proportion of forest ranged from 5.4% to 87.5% with an average of 54%. Agriculture, wetland, open/disturbed, low-intensity development, and high-intensity development varied between 0 and 40% of the buffer area across all ponds in both time periods. There was little variation in the proportion of open water or shrubland between ponds or across time periods, with none of the ponds having more than 10% of the buffer area in either land cover class. For the land use covariates, average historic human population density was 1.03 people/km^2 and average modern historic human population density was 0.84 people/km^2. Historic human population density ranged from 0.06 to 8.98 people/km^2 (Fig. 4) and modern human population density ranged from 0.02 to 6.37 people/km^2 (Fig. 5). Historic road length ranged from 1171 m to 11,120 m with an average of 3225.9 m. Modern road length ranged from 578.57 m to 11,202.2 m with an average of 2948.5 m. Historic average daily traffic ranged from 0 to 25,091.9 cars/day with an average of 2,104.9 cars/day. Modern average daily traffic ranged from 0 to 15,542 cars/day with an average of 2,118.9 cars/day.

For historic turtle density, the best supported model (lowest DIC) included the proportion of forest and the proportion of wetlands. Five other models received substantial support (<2 ΔDIC), and mostly consisted of combinations of proportion forest, proportion wetlands, and road length (Table 8). Averaged across the top 6 historic painted turtle density models, the proportion of forest cover was the only covariate that had an effect on turtle density, with forest cover negatively impacting painted turtle density (β_{Forest} = -0.72, 95% CI = -1.29 – -0.72). However, the effect of wetlands on population density could not be dismissed entirely as the 95% confidence interval narrowly crossed zero (Fig. 7) and wetlands did have a positive effect on turtle density under a 90% confidence interval ($\beta_{Wetland}$ = 0.51, 90% CI = 0.02 – 1.02). After model averaging, road length did not have an effect on historic turtle density ($\beta_{Road\ Length}$ = 0.3, 95% CI = -0.38 – 0.97). The maximal scale of effect for proportion forest and proportion wetlands was 260.4 m (95% CI = 22.9 – 485.8 m) and 196 m (95% CI = 31 – 483 m) respectively. We tested for the influence of the 1970-1980 land cover covariates on historic turtle density, but they performed significantly worse than the 2001 land cover covariates in all instances (Appendix B2).

Modern painted turtle density was best explained by historic proportion of forest and wetlands, and modern road length. There were six other models with substantial support (<2 ΔDIC). Similar to the historic models, most comprised combinations of proportion forest, proportion wetlands, and road length (Table 9). In each instance, the models with historic land cover covariates (2001) outperformed those with modern land cover covariates (2019), supporting the hypothesis that there is a lag time between a landscape disturbance and a measurable effect on turtle density.

Averaged over the top seven modern painted turtle density models, proportion wetland and road length had an effect on turtle density (Fig. 7). The proportion of wetlands had a positive impact on turtle density ($\beta_{Wetland}$ = 0.79, 95% CI = 0.19 – 1.36), while road length had a negative impact ($\beta_{Road\ Length}$ = -0.79, 95% CI = -1.59 – -0.02). Similar to the effect of wetlands on historic density, the 95% confidence interval for proportion forest narrowly crossed zero and forest cover did have an negative effect on turtle density under a 90% confidence interval (β_{Forest} = -0.64, 95% CI = -1.42 – -0.03). The maximal scale of effect for wetlands, road length, and forest were 255.5 m (95% CI = 84 – 472.5 m), 262.9 m (95% CI = 32.4 – 478.9 m), and 326 m (95% CI = 66.1 – 494.7 m) respectively (Fig. 8).

The GLM for historic sex ratio identified proportion of forest as the best supported model (lowest AICc), with greater forest cover associated with a higher proportion of males (coefficient = 0.27, SE = 0.17, p-value = 0.012). Three other models also received substantial support (<2 ΔAICc), and each contained a single covariate, wetlands, development, and average daily traffic (Table 10). However, none of those covariates had a significant effect on sex ratio as they all had p-values greater than 0.05 (wetlands p-value = 0.25; development p-value = 0.36; average daily traffic p-value = 0.92). Similar to the historic time period, modern sex ratio was driven by proportion forest (Table 10) with a positive relationship between the amount of forest and the proportion males in the population (coefficient = 0.72, SE = 0.22, p-value = 0.003).

Chemical Pollutants

Two organochlorine pesticides and five synthetic pyrethroids were identified in painted turtle blood. There were no detections of organochlorine pesticides or synthetic pyrethroids in the water samples. None of the organophosphate compounds that were tested for were detected in any blood or water samples.

Five synthetic pyrethroids were detected in turtle blood: bioallethrin-I, bioallethrin-II, bifenthrin, transfluthrin, and tetramethrin-II. The most pervasive synthetic pyrethroid detected was bioallethrin-I which was present in 27 of the 30 blood samples across 11 of the 12 ponds tested. The mean concentration was 4,643.1 ppb with a range from 4463.5 ppb to 5446.7 ppb. Bifenthrin was found at the same 11 ponds and in 19 blood samples with a mean concentration of 3306.7 ppb (ranged from 3301.4 ppb to 3344.6 ppb). Bioallethrin-II was found in 16 blood samples across 10 ponds with a mean concentration of 2088.4 ppb (ranged from 1903.1 ppb to 2227.3 ppb). Transfluthrin and tetramethrin-II were detected at a much lower frequency. Transfluthrin was detected in a single sample with a concentration of 2697.7 ppb and tetramethrin-II was detected in five samples across five ponds with a mean concentration of 2506.6 ppb (ranged from 2499.9 ppb to 2510.9 ppb).

Two organochlorine pesticides were detected in turtle blood: dieldrin and aldrin. These compounds are closely related to one another and aldrin is readily converted into dieldrin by the environment (Centers for Disease Control and Prevention 2021). Dieldrin was found in each of the 30 blood samples across every pond with a mean concentration of 145.3 ppb (ranged from 85.7 ppb to 284.7 ppb). Aldrin was found in nine blood samples across six ponds with a mean concentration of 53.1 ppb (ranged from 13.3 ppb to 135.2 ppb).

The generalized linear models relating the concentrations of both organochlorine pesticides and each of the four synthetic pyrethroids to land cover and land use covariates failed to identify any significant relationships between chemical concentrations and landscape covariates (Tables 12 and 13). The average p-value was 0.63 and the lowest p-value across all chemicals and landscape covariates tested was 0.123. Additionally, the land cover and land use covariates did not explain much of the variation in either the synthetic pyrethroids (Fig. 9 and 10) or the organochlorine pesticides (Fig. 11 and 12). The average R^2 value was 0.05 and the maximum R^2 across all chemicals and landscape covariates tested was 0.32.

DISCUSSION

We used a survey-resurvey approach to determine the effects of landscape context on painted turtle population density and sex ratio over a 20-year period in southeastern New Hampshire. We found that forest cover and road length negatively impacted painted turtle density, while the proportion of wetlands positively impacted turtle density. Moreover, we found that the historic (2001) land cover covariates were a better predictor of modern (2019-2021) turtle density than is the modern (2019) landscape, suggesting a lag time between land cover conversion and a measurable effect on population density. We found no evidence of a lag effect on turtle sex ratio. Rather, current proportion of forest cover was the best predictor of sex ratio, with male-skewed populations associated with highly forested ponds in both time periods. Lastly, we identified pervasive bioaccumulation of synthetic pyrethroids and organochlorine pesticides in painted turtles across the urbanization gradient. Despite having tested for chemical contaminants in turtle blood across the full range of landscape contexts, including ponds with the highest proportion of forest, agriculture, development, and human population density, there was no relationship between the concentration of chemicals detected and any land cover or land use covariates.

Lag in Demographic Response

Turtles are long-lived species, with long generation times and low annual recruitment, and their demographic response to environmental change is often lagged as it may take years for the cumulative reduction in recruitment to substantially impact abundance (Metzger et al. 2009;

Jiménez-Franco et al. 2022). We found historic landscape variables outperformed modern landscape variables as predictors of modern painted turtle density, suggesting a lag in painted turtle response. A 20-year lag is reasonable given painted turtles expected lifespan of 40-50 years (Schwanz et al. 2011) and a time to sexual maturity of four to six years (Frazer et al. 1993). A recent study on spur-thighed tortoise (*Testudo graeca*) populations in southeast Spain, found that it took 30 years following land cover conversion before negative effects on population density could be detected (Jiménez-Franco et al. 2022). The spur-thighed tortoise is longer lived than painted turtles with a lifespan of 127 years (Caglar et al. 2021) and a time to sexual maturity between eight and 12 years (Tiar-Saadi et al. 2022), which is likely why the lag-time we identified for painted turtles is shorter. Studies of long-lived species suggest extinction debt is common, underscoring the need for pro-active management strategies to mitigate declines (Kuussaari et al. 2009).

Effect of Land Cover and Land Use

Forest cover negatively effects painted turtle and is associated with male-skewed sex ratios and lower population densities. These two demographic responses to forest cover are likely linked. Freshwater turtles prefer to nest in open areas with little to no vegetative cover and these ponds are most likely to produce a 50/50 hatchling sex ratio due to temperature-dependent sex determination (Kolbe & Janzen 2002; Mitchell et al. 2013; Roberts et al. 2022). When preferred nesting sites are unavailable, turtles will nest under canopy where incubation temperatures will be lower resulting in a male-skewed hatchling sex ratio (Mitchell et al. 2013). Consistently male-biased hatchlings can lead to a male-biased population over time. Male-biased ponds can have decreased reproductive output (Judson et al. 2020), which could ultimately decrease population density. Our findings are consistent with those of Marchand and Litvaitis (2004), who found the proportion of males in the historic population was positively associated with the proportion of forest within 500 meters of the pond.

The proportion of wetlands was the only covariate found to have a positive effect on turtle density. Wetlands surrounding a pond effectively increase the amount of core habitat available to a freshwater turtle population (Roe et al. 2009). Wetlands also increase connectivity between ponds by facilitating movement without incurring road-mortality risk (Patrick & Gibbs 2010). Wetland networks can be especially beneficial when a local pond experience stochastic environmental events (e.g., drought or flooding) (Roe & Georges 2007; Rees et al. 2009). In contrast to our findings on turtle density, the proportion of wetlands did not have a significant impact on modern or historic painted turtle sex-ratio. A recent study on the spotted turtle suggests that the effects of wetlands on sex-ratio is dependent on both the spatial arrangement of wetlands and the degree of urbanization (Roberts et al. 2022).

Roads negatively impact modern turtle density. Direct mortality from vehicle collisions is likely a primary driver of the negative relationship between population density and road length. High rates of road mortality in turtle populations are well documented (Ashley & Robinson 1996; Haxton 2000a; Steen & Gibbs 2004). We did not detect the same relationship between roads and historic turtle density, which was consistent with the findings of Marchand and Litvaitis (2004). This suggests that the cumulative loss of individuals to road mortality may not have been large enough to result in detectable differences in density at that time. We were surprised to find that average daily traffic was a worse predictor of turtle density than road length, as we had expected that roads with more traffic would pose a greater threat to migrating turtles. Road length may have been the better predictor because it captures the other deleterious effects of roads better than traffic volume. Freshwater turtles readily make use of anthropogenic areas for nesting, including along roadways (Beaudry et al. 2010). However, the females that use these areas and the resulting nests could be at an increased risk of predation. Red foxes, raccoons, and striped skunks, disproportionately use edge habitat along roads to search for prey items (Frey & Conover 2006).

We failed to detect a relationship between the length of roads surrounding a pond and the modern or historic turtle population sex ratio. These findings are in direct contrast to previous studies, including at these same ponds 20 years ago (Steen & Gibbs 2004; Marchand & Litvaitis 2004; Gibbs & Steen 2005; Patrick & Gibbs 2010). However, there is a growing body of research that have also found no relationship between roads and sex ratio (Dorland et al. 2014; Carstairs et al. 2018; Roberts et al. 2022). These conflicting findings suggest the spatial arrangement of other features on the landscape, such as nesting areas and adjacent wetlands, may play a crucial role in determining what effects roads have on a population. For example, in areas where wetlands are less aggregated, male and females turtles may have similar rates of road mortality since males will make inter-wetland movements in search of mates and females will make annual nesting migrations when they must leave the pond in search of nest sites on land (Roberts et al. 2022). Additionally, warming temperatures associated with climate change may be increasing the proportion of female hatchlings which could be having a modulating effect on the disproportionate loss of females by road mortality (Schwanz et al. 2010; Jensen et al. 2018).

When determining how wildlife respond to differences in land cover and land use, it is critical to consider the scale of effect, or spatial extent at which the variable best predicts population response (Jackson & Fahrig 2012). Scale of effect can be influenced by species traits (e.g., body size or dispersal capability) landscape context, and the analytical methods used

(Miguet et al. 2016). Failure to accurately determine the scale of effect may result in errors in inference (Eigenbrod et al. 2011; Jackson & Fahrig 2015). Often, a multi-scale approach is adopted where the effect is assessed across a series of buffers of a predetermined extent (Miguet et al. 2016). However, this approach assumes that the effect of a variable is constant throughout the entire buffer and drops to zero immediately outside of the chosen buffer (Aue et al. 2012). To avoid this limitation, we employed a distance-weighted smoothing method where the effect of a landscape variable diminishes as distance from the focal site increases (Miguet et al. 2017; Moll et al. 2020). We found little variation in the maximal scale of effect estimates between the landscape variables or between time periods (Fig. 8). Mean estimates ranged from approximately 200 m to 325 m from the pond perimeter with nearly all models agreeing that the 95% confidence interval for the maximal scale of effect is between 50 meters and 450 meters. These results suggest that the initial choice of a 2,000-meter buffer was more than sufficient to capture the effect of each landscape feature. The scales of effect we found for forest cover and road length were consistent with those in Marchand and Litvaitis (2004), which did not find evidence for a scale of effect greater than 500 m despite testing scales up to 2000 m. Our results also align with theoretical expectations, where the scale of effect is generally expected to be four to nine times the mean dispersal distance (Jackson & Fahrig 2012). Radio-telemetry on painted turtles in southeastern New Hampshire found that mean dispersal distance ranged from 54 m to 115 m (Baldwin et al. 2009). These findings suggest that any conservation and management activities aimed at bolstering freshwater turtle populations, such habitat manipulation or road-mortality mitigation, should be focused within 500 m of the pond perimeter.

Chemical Contamination

Organochlorine pesticides and synthetic pyrethroids were detected in 100% of the turtles tested. Surprisingly, chemical contaminants were not detected in any water samples across all 31 modern ponds. This pattern could arise if the residence time of chemical contaminants are shorter in water than in turtle tissue or if these chemicals persist at low concentrations in the environment and bioaccumulate in turtle tissue. Given that organochlorine pesticides and synthetic pyrethroids are known to persist in aquatic environments, it is likely that these chemicals are not actually absent in the water samples but are rather present in concentrations below the detection limit of the testing method and are bioaccumulating or biomagnifying in turtles (Lee et al. 2004; Wong et al. 2004; Xie et al. 2022). Organochlorine pesticides can have residence times in freshwater systems greater than 10 years (Catalan et al. 2004) and while synthetic pyrethroids were originally believed to degrade quickly in the environment, recent studies have shown that these compounds can become trapped in sediment rendering them inaccessible to microbial degraders and increasing their environmental persistence (Lee et al. 2004). Chemical laden sediment could be another exposure pathway as painted turtles regularly forage along the pond bottom (Moldowan et al. 2015) and hibernate completely submerged in pond sediment for the duration of winter (Taylor & Nol 1989). These water- and fat-soluble chemicals bioaccumulate in the tissue of aquatic animals when chemical uptake exceeds the body's capacity to process and eliminate the chemicals, resulting in a build-up in the body over time (Franke et al. 1994; Chopra et al. 2011; Xie et al. 2022). The concentration of chemicals accumulated is much greater for animals occupying higher trophic levels due to the biomagnification (Drouillard 2008).

Human land use practices introduced these chemicals into the freshwater systems and can have legacy effects long after their use has been stopped. For example, organochlorine pesticides, the same family that contains the infamous dichlorodiphenyltrichloroethane (DDT), were used extensively in agriculture and residential insect control until they were banned by the USDA in 1970 citing toxic health effects (U.S. EPA 2003). Despite not being used for more than 50 years, we detected organochlorine pesticides in all turtle blood samples we tested. Synthetic pyrethroids, which largely replaced organochlorine pesticides after their ban (Riar 2014), are still used extensively today by commercial mosquito and tick prevention companies and by individuals in the form of aerosol sprays or foggers (Spurlock & Lee 2008). Synthetic pyrethroids were also found in 100% of the turtles we tested. The bioaccumulation of organochlorine pesticides and synthetic pyrethroids in freshwater turtles is concerning because both groups of chemicals have toxic effects on aquatic life and the potential for endocrine disruption (Coats et al. 1989; Willett et al. 1998; Chopra et al. 2011; Wang et al. 2020). Organochlorine pesticides are neurotoxic, carcinogenic, and at low concentrations have sub-lethal effects such as impaired development, immunosuppression, and reduced fertility (Jayaraj et al. 2016). Synthetic pesticides are acutely toxic at high concentrations and have sub-lethal effects such as impaired development, behavioral modifications, and low survivability (Coats et al. 1989). The potential for endocrine disruption is particularly concerning because even in concentrations below the detection limits of the testing used in this study, endocrine disruptors can elicit serious biological response, often with reproductive consequences that can influence fertility and sex ratio (Guillette Jr. et al. 1996; Vandenberg et al. 2012; Iwanowicz et al. 2016; Leslie 2017). Our findings demonstrate that these chemical contaminants are pervasive in freshwater systems in southeastern New Hampshire and further investigations is warranted to determine the effects that these compounds may be having on turtle physiology and their contribution to population declines.

Conclusions

We found strong evidence of a legacy effect of historic landscape context on modern painted turtle density in New Hampshire. Considering the legacy effects of land cover change and land use is essential for effective conservation and management because even populations that appear healthy now could have an elevated risk of extinction in the future.

Painted turtles are widespread and possess several key life-history traits shared by most freshwater turtles, long lifespans, delayed sexual maturity, and dependence on both aquatic and terrestrial habitats, making them a potentially good indicator of freshwater turtle response to environmental change. There is, however, variation among freshwater turtle species in their relative use and dependence on aquatic and terrestrial habitats. Painted turtles fall at the highly aquatic end of this habitat-use spectrum and are relatively small-bodied, thus we must adjust our expectations for other freshwater turtles whose use of the landscape differs in important ways. Semi-aquatic species like the Blanding's turtle may be at even greater risk because they utilize a mosaic of different habitats seasonally and frequently make long-distance and long-duration terrestrial movements (Ross & Anderson 1990; Rowe & Moll 1991; Congdon et al. 2011). Large-bodied species like the snapping turtle travel much further overland in search of nesting areas (Obbard & Brooks 1980). Species that require a mosaic of wetland and terrestrial habitats, and must travel long distances between them, are at greater risk of road-related mortalities, decreased genetic diversity, and male-skewed sex ratios (Reid and Peery 2014); they may also respond to land cover and land use change at different temporal and spatial scales than highly aquatic species like the painted turtle.

The extent to which our findings will translate to other regions is unclear. The relationships we detected may not exist in regions with different land cover or land use regimes, as the rural-urban gradient that exists in our study area is not universally representative of 'urbanization' or agriculture in other regions. Our most intense 'agriculture' ponds had no greater than 40% agriculture within 500 m of the pond and the agricultural land cover is primarily hay cultivation. The effect of large-scale industrial agriculture practices may have different impacts than what we observed. Similarly, we did not have any ponds where high-intensity development comprised more than 50% of the area within 500 m.

TABLES

Table 1. List of the 65 chemical contaminants tested for in turtle blood samples and water samples. Testing was performed by Michigan State Veterinary Diagnostics Laboratory using gas chromatography-mass spectrometry.

Synthetic Pyrethroids	Organophosphate Pesticides	Organochlorine Pesticides
Tefluthrine	Methacrifos	BHC-alpha
Trans-fluthrin	Sulfotepp	BHC-gamma (Lindane)
Bioallethrin-I	Terbufos	BHC-beta
Bioallethrin-II	Tocolofos-methyl	BHC-delta
Resmethrin	Ronnel	Heptachlor
Bifenthrin	Fenthion	Aldrin
Phenothrin	Malathion	Oxychlordane
Tetramethrin-I	Bromophos-ethyl	Chlordane-cis
Tetramethrin-II	Clofenvinfos	Chlordane-trans
cis-Permethrin	Prothiofos	Trans-nonachlor
trans-Permethrin	Tetrachlorvinphos	Chlordane-cis
lambda-Cyhalothrin	Bromfenvinfos	Endosulfan I
Acrinathrin	Profenofos	Endosulfan II
Cyfluthrin-I	Fenamiphos	Dieldrin
Cypermethrin-I	Chlorthiophos	DDE-p,p'
Cyfluthrin-II	Ethion	Endrin
Cyfluthrin-III	Carbophenothion	DDD-p,p'
Cypermethrin-II	Edifenphos	DDT-p,p'
Cypermethrin-III	Leptophos	Methoxychlor
Cyfluthrin-IV	Coumaphos	Endosulfan sulfate Endrinketone
Cypermethrin-IV		
Deltamethrin-I		
Fluvalinate		
Deltamethrin-II		

Table 2. Results of model selection using DIC for Bayesian closed-population capture-mark-recapture painted turtle (*Chrysemys picta*) abundance models. Rhat values were used to assess model convergence.

Model	Mean N	SD	95% CI	Rhat	DIC	ΔDIC
Closed Time Effects	39	4.9	32 - 50	1.005	270.9	0
Closed Individual and Time Effects	42	7.7	37 - 62	1.006	278.2	7.3
Closed Null Model	43	7.9	33 - 64	1.007	361	90.1
Closed Individual Random Effects	68	24.5	37 - 129	1.008	620.4	349.5
Open Jolly-Seber Model	37	11.46	19 – 64	1.006	630.3	359.4

Table 3. Summary of captures by species and year for the 31 ponds sampled in the modern time period (2019-2021) in southeastern New Hampshire, USA.

Species	Year						Total
	2019		2020		2021		
	New Captures	Recaptures	New Captures	Recaptures	New Captures	Recaptures	
Chrysemys picta	236	68	264	66	568	229	1431
Chelydra serpentina	30	7	68	12	63	15	195
Emydoidea blandingii	2	1	2	0	30	10	45
Sternotherus odoratus	10	1	0	0	7	0	18
Clemmys guttata	0	0	0	0	5	0	5
Total	278	77	334	78	673	254	1694

Table 4. Abundance estimates for historic painted turtle populations under a Bayesian closed population capture-mark-recapture model. Model estimates are provided for each of the 32 ponds sampled in 2000 and 2001 (Marchand and Litvaitis 2004). N is abundance and P1-P4 are detection probabilities. Rhat values for all N and P estimates ≤ 1.02. Pond names have been changed to protect the privacy of private landowners.

Pond	Year	N	SD	2.50%	97.50%	P1	P2	P3	P4
Walnut	2000	189.21	23.39	146	233.03	0.1	0.25	0.14	0.13
Beech	2001	175.69	22.4	129	208	0.09	0.03	0.15	0.1
Juniper	2001	159.08	28.05	110	210	0.18	0.11	0.15	0.05
Black Gum	2001	130.16	23.09	91	183	0.12	0.13	0.07	0.27
Pitch Pine	2000	122.09	22.81	87	178	0.28	0.07	0.17	0.12
Basswood	2001	112.56	18.09	85	154	0.3	0.2	0.13	0.14
Hophornbeam	2001	107.63	24.67	70	164	0.12	0.16	0.1	0.17
Black Cherry	2001	83.65	19.26	56	133	0.26	0.1	0.18	0.1
White Spruce	2001	72.87	19.46	47	125	0.14	0.14	0.27	0.1
White Ash	2001	66.99	24.34	37	132	0.11	0.02	0.15	0.27
Bigtooth Aspen	2001	45.07	10.12	32	71	0.35	0.24	0.02	0.22
Quaking Aspen	2001	44.79	15.87	24	84	0.09	0.19	0.19	0.09
Larch	2001	42.52	17.29	23	86.03	0.1	0.15	0.23	0.18
Hickory	2000	36.49	10.42	24	65	0.06	0.47	0.19	0.14
Hawthorn	2001	32.65	16.03	17	77.05	0.07	0.4	0.17	0.03
Yellow Birch	2000	31.38	6.43	23	47	0.25	0.37	0.22	0.19
Silver Maple	2001	28.53	16.14	13	78	0.08	0.12	0.39	0.04
Red Oak	2000	27.24	14.58	12	66	0.08	0.04	0.16	0.32
Dogwood	2001	26.61	7.34	18	45	0.37	0.15	0.4	0.04
Sumac	2000	21.26	7.66	14	43.03	0.05	0.18	0.19	0.6
Hemlock	2001	19.02	4.63	14	31	0.1	0.2	0.64	0.15
Red Maple	2000	18.01	3.1	15	27	0.92	0.67	0.8	0.91
White Pine	2000	16.81	9.84	7	43	0.12	0.25	0.13	0.19
Serviceberry	2000	15.81	9.35	7	43.03	0.07	0.34	0.2	0.13
Sugar Maple	2001	14.19	7.75	8	35	0.07	0.56	0.14	0.07
Mountain Ash	2001	11.23	3.44	8	21	0.08	0.56	0.4	0.08
Black Oak	2000	10.29	4.51	6	23	0.18	0.36	0.27	0.18
White Oak	2000	8.88	3.92	6	19	0.1	0.1	0.59	0.29
River Birch	2000	7.57	2.77	6	15	0.76	0.11	0.22	0.11
Chestnut	2000	7.4	3.85	5	16	0.11	0.58	0.23	0.23
Elm	2001	7.25	3.06	5	16	0.11	0.23	0.58	0.22
Paper Birch	2000	5.16	3.77	3	14.03	0.47	0.32	0.15	0.16

Table 5. Abundance estimates for modern painted turtle populations under the Bayesian closed population mark-recapture model. Estimates are provided for each of the 31 ponds sampled in 2019, 2020, or 2021. N is abundance and P1-P4 are detection probabilities. Rhat values for all N and P estimates ≤ 1.02. Pond names have been changed to protect the privacy of private landowners.

Pond	Year	N	SD	2.50%	97.50%	P1	P2	P3	P4
Black Willow	2021	123.05	22.13	88	173	0.13	0.06	0.24	0.18
Serviceberry	2021	88.92	20.14	75	136.06	0.09	0.23	0.08	0.03
Walnut	2021	76.26	15.92	53	114	0.19	0.23	0.29	0.03
Pin Cherry	2021	72.36	20.79	45	125	0.14	0.23	0.16	0.09
Hophornbeam	2021	71.57	32.78	31	160	0.18	0.18	0.05	0.05
Mountain Ash	2021	67.92	19.04	43	114	0.18	0.3	0.06	0.2
White Ash	2019	67.74	12.37	50	97	0.25	0.13	0.27	0.22
Black Cherry	2021	66.1	24.12	36	130	0.05	0.24	0.15	0.1
Sycamore	2020	59.69	12.27	42	89	0.23	0.29	0.15	0.14
Red Pine	2021	50.43	10.76	36	79	0.24	0.08	0.1	0.2
White Spruce	2021	46.91	17.81	24	95	0.23	0.07	0.11	0.16
Pitch Pine	2021	45.48	20.59	25	99	0.29	0.26	0.14	0.03
Hornbeam	2021	42.04	5.48	34	55	0.3	0.25	0.37	0.32
Quaking Aspen	2020	41.71	20.93	20	99	0.29	0.16	0.03	0.08
Juniper	2021	37.81	18.93	18	92	0.03	0.17	0.07	0.38
Sumac	2019	37.1	16.65	19	84.03	0.29	0.03	0.27	0.03
Yellow Birch	2019	32.89	8.35	22	55	0.09	0.3	0.27	0.24
Bigtooth Aspen	2020	25.37	6.36	18	42	0.38	0.38	0.19	0.07
Beech	2020	24.78	18.17	10	82	0.19	0.34	0.05	0.05
Silver Maple	2021	16.48	8.9	8	41.03	0.19	0.44	0.07	0.06
Sassafras	2021	13.95	7.23	9	33	0.62	0.21	0.07	0.07
Hickory	2019	11.53	3.29	8	20	0.08	0.49	0.25	0.41
White Cedar	2021	10.54	6.64	6	27	0.55	0.09	0.18	0.09
Larch	2020	10.09	3.62	7	19	0.18	0.53	0.18	0.27
Chestnut	2020	10.07	3.95	7	21	0.07	0.63	27	0.18
Dogwood	2020	8.8	2.42	7	16	0.67	0.19	0.19	0.29
Black Oak	2021	7.48	1.94	6	13	0.22	0.65	0.22	0.22
White Oak	2020	7.1	2.9	5	15	0.36	0.58	0.11	0.12
Paper Birch	2019	5.63	3.06	4	13	0.14	0.14	0.57	0.42
River Birch	2019	5.55	2.32	4	12	0.14	0.42	0.57	0.15
Red Maple	2019	5.21	3.83	3	15	0.15	0.47	0.32	0.16

Table 6. Change in painted turtle abundance between initial sampling (2000 or 2001; Marchand and Litvaitis 2004) and resampling (2019, 2020, or 2021) for the 24 resurveyed ponds in southeastern New Hampshire, USA. N is painted turtle abundance. Pond names have been changed to protect the privacy of private landowners.

Pond	Historic		Current		
	Year	N	Year	N	Change in N
Serviceberry	2000	15.81	2021	88.92	73.11
Mountain Ash	2001	11.23	2021	67.92	56.69
Sumac	2000	21.26	2019	37.1	15.84
Chestnut	2000	7.4	2020	10.07	2.67
Yellow Birch	2000	31.38	2019	32.89	1.51
White Ash	2001	66.99	2019	67.74	0.75
Paper Birch	2000	5.16	2019	5.63	0.47
White Oak	2000	8.88	2020	7.1	-1.78
River Birch	2000	7.57	2019	5.55	-2.02
Black Oak	2000	10.29	2021	7.48	-2.81
Quaking Aspen	2001	44.79	2020	41.71	-3.08
Hophornbeam	2001	107.63	2020	98.39	-9.24
Silver Maple	2001	28.53	2021	16.48	-12.05
Red Maple	2000	18.01	2019	5.21	-12.8
Pitch Pine	2000	122.09	2019	105.37	-16.72
Black Cherry	2001	83.65	2021	66.1	-17.55
Dogwood	2001	26.61	2020	8.8	-17.81
Bigtooth Aspen	2001	45.07	2020	25.37	-19.7
Hickory	2000	36.49	2019	11.53	-24.96
White Spruce	2001	72.87	2021	46.91	-25.96
Larch	2001	42.52	2020	10.09	-32.43
Walnut	2000	189.21	2021	76.26	-112.95
Juniper	2001	159.08	2021	37.81	-121.27
Beech	2001	175.69	2020	24.78	-150.91

Table 7. Binomial significance test results for historic (2000-2001) and modern (2019-2021) painted turtle sex ratio. A p-value of less than 0.05 indicated the population sex ratio is biased. NA indicates the pond was not sampled in that time period.

	Historic		Modern	
Pond	Sex Ratio	P-value	Sex Ratio	P-value
Basswood	0.667	0.001	NA	NA
Beech	0.627	0.026	0.300	0.688
Bigtooth Aspen	0.583	0.280	0.400	0.581
Black Cherry	0.438	0.295	0.400	0.424
Black Gum	0.500	1.000	NA	NA
Black Oak	0.833	0.039	0.610	1.000
Black Willow	NA	NA	0.482	0.791
Chestnut	0.833	0.109	1.000	0.031
Dogwood	0.556	0.824	0.800	0.219
Elm	0.385	0.581	NA	NA
Hawthorn	0.471	0.860	NA	NA
Hemlock	0.467	0.804	NA	NA
Hophornbeam	0.390	0.060	0.375	0.281
Hornbeam	NA	NA	0.750	0.007
Juniper	0.532	0.625	0.770	0.006
Larch	0.556	0.845	0.600	1.000
Mountain Ash	0.857	0.039	0.550	0.585
Paper Birch	0.333	0.625	0.672	1.000
Pin Cherry	NA	NA	0.678	0.087
Pitch Pine	0.518	0.804	0.600	0.541
Quaking Aspen	0.588	0.324	0.679	0.344
Red Maple	0.684	0.041	0.500	1.000
Red Oak	0.667	0.099	NA	NA
Red Pine	NA	NA	0.667	0.152
River Birch	0.429	0.727	0.560	1.000
Sassafras	NA	NA	0.800	0.109
Serviceberry	0.500	1.000	0.750	0.001
Hickory	0.545	0.856	0.625	0.508
Silver Maple	0.615	0.607	0.714	0.344
Sugar Maple	0.375	0.383	NA	NA
Sumac	0.857	0.004	0.750	0.077
Sycamore	NA	NA	0.600	0.377
Walnut	0.459	0.329	0.714	0.006
White Ash	0.500	1.000	0.349	0.049
White Cedar	NA	NA	0.429	1.000
White Oak	0.714	0.696	1.000	0.063
White Pine	0.375	0.727	NA	NA
White Spruce	0.486	1.000	0.728	0.064
Yellow Birch	0.476	1.000	0.476	1.000

Table 8. Results of model selection using DIC for historic distance-weighted smoothing painted turtle density models. All models within 3 DIC units of top model shown, full table of results available in appendix B2. Only models within 2 DIC units of the top model are considered equivalent. Covariates are for = proportion forest, wet = proportion wetlands, lid = proportion low-intensity development, opd = proportion open/disturbed, rl = road length, adt = average daily traffic, and hpop = human population density.

Land Cover		Land Use		DIC	ΔDIC	Bayes.P
Year	Covariate	Year	Covariate			
2001	for			107.81	0.00	0.53
2001	for, wet			108.01	0.20	0.54
2001	for, wet	2001	rl	109.40	1.59	0.53
2001	for, opd			109.51	1.70	0.53
2001	wet, lid			109.64	1.83	0.53
2001	for, opd, wet			109.90	2.09	0.54
2001	for	2001	rl	110.05	2.24	0.54
2001	for, lid			110.16	2.35	0.53
2001	for, wet	2001	hpop	110.27	2.46	0.55
2001	for	2001	adt	110.27	2.46	0.53
2001	wet	2001	rl	110.32	2.51	0.53

Table 9. Results of model selection using DIC for modern distance-weighted smoothing painted turtle density models. All models within 3 DIC units of top model shown, full table of results available in appendix B1. Only models within 2 DIC units of the top model are considered equivalent. Covariates are for = proportion forest, wet = proportion wetlands, lid = proportion low-intensity development, opd = proportion open/disturbed, hid = proportion high-intensity development, rl = road length, adt = average daily traffic, and hpop = human population density.

Land Cover		Land Use		DIC	ΔDIC	Bayes.P
Year	Covariate	Year	Covariate			
2001	for, wet	2019	rl	99.56	0.00	0.53
2001	for, wet			99.67	0.11	0.54
2001	wet			99.75	0.20	0.53
2019	wet			100.43	0.87	0.55
2019	for, wet			100.95	1.39	0.54
2001	wet, lid			101.21	1.65	0.54
2001	for	2019	rl	101.22	1.67	0.53
2001	for, opd	2019	rl	101.62	2.07	0.53
2001	wet, hid			101.69	2.13	0.54
2019	for, wet	2019	rl	101.76	2.20	0.53
2001	for, wet	2019	hpop	101.92	2.37	0.54
2019	wet, lid			102.07	2.51	0.53
2001	wet	2019	rl	102.10	2.54	0.54
2001	for, lid			102.15	2.59	0.52
2019	wet, hid			102.17	2.61	0.53
2001	for, opd, wet			102.43	2.88	0.54
2019	for	2019	rl	102.54	2.99	0.54
2001	wet	2019	adt	102.55	3.00	0.53

Table 10. Results of model selection using AICc for generalized linear models of the effect of land cover and land use on the historic proportion of male adult painted turtles. Covariates are for = forest, road = road length, adt = average daily traffic, hpop = human population, dev = development, opd = open/disturbed, wet = wetlands, water = open water, ag = agriculture.

Covariate	Year	AICc	ΔAICc	Likelihood	Weight
for	2001	-1.74	0.00	1.00	0.20
wet	2001	-0.64	1.10	0.58	0.11
dev	2001	-0.11	1.64	0.44	0.09
adt	2001	0.00	1.74	0.42	0.08
hpop	2001	0.26	2.01	0.37	0.07
opd	2001	0.36	2.10	0.35	0.07
road	2001	0.37	2.11	0.35	0.07
for, road	2001, 2001	0.62	2.37	0.31	0.06
water	2001	0.73	2.47	0.29	0.06
ag	2001	0.76	2.50	0.29	0.06
for, hpop	2001, 2001	0.85	2.59	0.27	0.05
for, adt	2001, 2001	1.02	2.76	0.25	0.05
for, hpop, road	2001, 2001, 2001	3.63	5.37	0.07	0.01
for, hpop, adt	2001, 2001, 2001	3.89	5.63	0.06	0.01

Table 11. Results of model selection using AICc for generalized linear models of the effect of land cover and land use on the modern proportion of male adult painted turtles. Covariates are for = forest, road = road length, adt = average daily traffic, hpop = human population, dev = development, opd = open/disturbed, wet = wetlands, water = open water, ag = agriculture.

Covariate	Year	AICc	ΔAICc	Likelihood	Weight
for	2019	8.84	0.00	1.00	0.23
for	2001	9.56	0.72	0.70	0.16
for, road	2019, 2019	10.98	2.14	0.34	0.08
for, adt	2019, 2019	11.06	2.23	0.33	0.07
for, hpop	2019, 2019	11.27	2.44	0.30	0.07
for, adt	2001, 2019	11.66	2.82	0.24	0.06
for, road	2001, 2019	11.76	2.92	0.23	0.05
for, hpop	2001, 2019	11.87	3.04	0.22	0.05
dev	2019	12.48	3.64	0.16	0.04
adt	2019	12.59	3.75	0.15	0.03
for, hpop, adt	2001, 2019, 2019	12.74	3.91	0.14	0.03
for, hpop, road	2001, 2019, 2019	13.09	4.25	0.12	0.03
for, road, adt	2001, 2019, 2019	13.71	4.87	0.09	0.02
hpop	2019	14.24	5.41	0.07	0.02
opd	2019	14.43	5.60	0.06	0.01
dev	2001	14.48	5.65	0.06	0.01
for, hpop, adt	2001, 2019, 2019	14.52	5.69	0.06	0.01
opd	2001	15.53	6.69	0.04	0.01
wet	2019	15.75	6.91	0.03	0.01
wet	2001	16.00	7.17	0.03	0.01
road	2019	18.06	9.23	0.01	0.00
water	2001	18.36	9.52	0.01	0.00
water	2019	18.44	9.61	0.01	0.00
ag	2019	18.73	9.89	0.01	0.00
ag	2001	18.74	9.90	0.01	0.00

Table 12. Generalized linear modeling results relating the concentration of four synthetic pyrethroids in 30 turtle blood samples across 12 ponds to land use and land cover covariates.

Chemical	Landscape Covariate	Coefficient	Standard Error	p-value	R2
Bioallethrin-I	Forest	-1227	1692	0.4847	0.05
Bioallethrin-I	Agriculture	217.6	4424.9	0.962	0.0002
Bioallethrin-I	Open/Disturbed	985.7	5041	0.8488	0.0038
Bioallethrin-I	Wetlands	5166.4	9042	0.58039	0.0316
Bioallethrin-I	Low-intensity Development	-1.642	5822.501	0.99	0.0001
Bioallethrin-I	High-intensity Development	1767.6	3654.6	0.639	0.0229
Bioallethrin-I	Human Density	147.1	237.1	0.549	0.037
Bioallethrin-I	Average Daily Traffic	0.03422	0.134	0.804	0.0065
Bioallethrin-I	Road Length	0.079	0.1615	0.6333	0.0236
Bioallethrin-II	Forest	491.3	943.2	0.6138	0.1363
Bioallethrin-II	Agriculture	961.2	2418.5	0.6993	0.0156
Bioallethrin-II	Open/Disturbed	512.6	2777.1	0.8574	0.0372
Bioallethrin-II	Wetlands	5976.4	4695.2	0.2319	0.1394
Bioallethrin-II	Low-intensity Development	-4605.8	2857.2	0.138	0.0132
Bioallethrin-II	High-intensity Development	-3614	1685.3	0.576	0.0487
Bioallethrin-II	Human Density	-198.1	117.5	0.123	0.0072
Bioallethrin-II	Average Daily Traffic	-0.01539	0.07388	0.839	0.0043
Bioallethrin-II	Road Length	-0.15335	0.07584	0.708	0.0233
Bifenthrin	Forest	-4.568	15.583	0.776	0.0095
Bifenthrin	Agriculture	45.392	101.7	0.666	0.0217
Bifenthrin	Open/Disturbed	-26.359	44.516	0.568	0.0375
Bifenthrin	Wetlands	-7.07	85.54	0.936	0.0008
Bifenthrin	Low-intensity Development	5.636	52.317	0.917	0.0013
Bifenthrin	High-intensity Development	-12.439	33.12	0.716	0.0154
Bifenthrin	Human Density	0.444	2.189	0.844	0.0046
Bifenthrin	Average Daily Traffic	-0.001	0.001	0.623	0.0279
Bifenthrin	Road Length	-0.003	0.001	0.804	0.0072
Tetramethrin-II	Forest	1815.9	1445.5	0.238	0.0264
Tetramethrin-II	Agriculture	-3154.8	3838.8	0.43	0.0633
Tetramethrin-II	Open/Disturbed	2760.2	4441.6	0.548	0.0034
Tetramethrin-II	Wetlands	-2252.8	8204.8	0.789	0.0075
Tetramethrin-II	Low-intensity Development	-1899.3	5183.7	0.7217	0.2063
Tetramethrin-II	High-intensity Development	-2311.5	3231.8	0.4908	0.3151
Tetramethrin-II	Human Density	-58.24	215.81	0.7928	0.2214
Tetramethrin-II	Average Daily Traffic	-0.1405	0.112	0.238	0.1359
Tetramethrin-II	Road Length	-0.0706	0.14476	0.6359	0.2902

Table 13. Generalized linear modeling results relating the concentration of aldrin and dieldrin in 30 turtle blood samples across 12 ponds to land use and land cover covariates.

Chemical	Landscape Covariate	Coefficient	Standard Error	p-value	R2
Aldrin	Forest	-15.59	36.66	0.68	0.0178
Aldrin	Agriculture	-49.88	92.99	0.6034	0.028
Aldrin	Open/Disturbed	152.537	96.225	0.144	0.2008
Aldrin	Wetlands	-255.55	178.38	0.1825	0.1703
Aldrin	Low-intensity Development	111.91	118.93	0.369	0.0813
Aldrin	High-intensity Development	31.04	78.18	0.6997	0.0155
Aldrin	Human Density	3.067	5.058	0.5578	0.0355
Aldrin	Average Daily Traffic	-0.002	0.002	0.4619	0.0553
Aldrin	Road Length	0.002	0.003	0.44	0.0607
Dieldrin	Forest	-2.742	32.98	0.935	0.0007
Dieldrin	Agriculture	23.429	83.805	0.786	0.0078
Dieldrin	Open/Disturbed	23.23	95.74	0.813	0.0059
Dieldrin	Wetlands	97.87	171.93	0.582	0.0314
Dieldrin	Low-intensity Development	0.27	110.69	0.998	0.0001
Dieldrin	High-intensity Development	-9.396	70.221	0.896	0.0018
Dieldrin	Human Density	3.143	4.485	0.449	0.0468
Dieldrin	Average Daily Traffic	-0.002	0.002	0.442	0.0602
Dieldrin	Road Length	0.001	0.003	0.987	0.0001

FIGURES

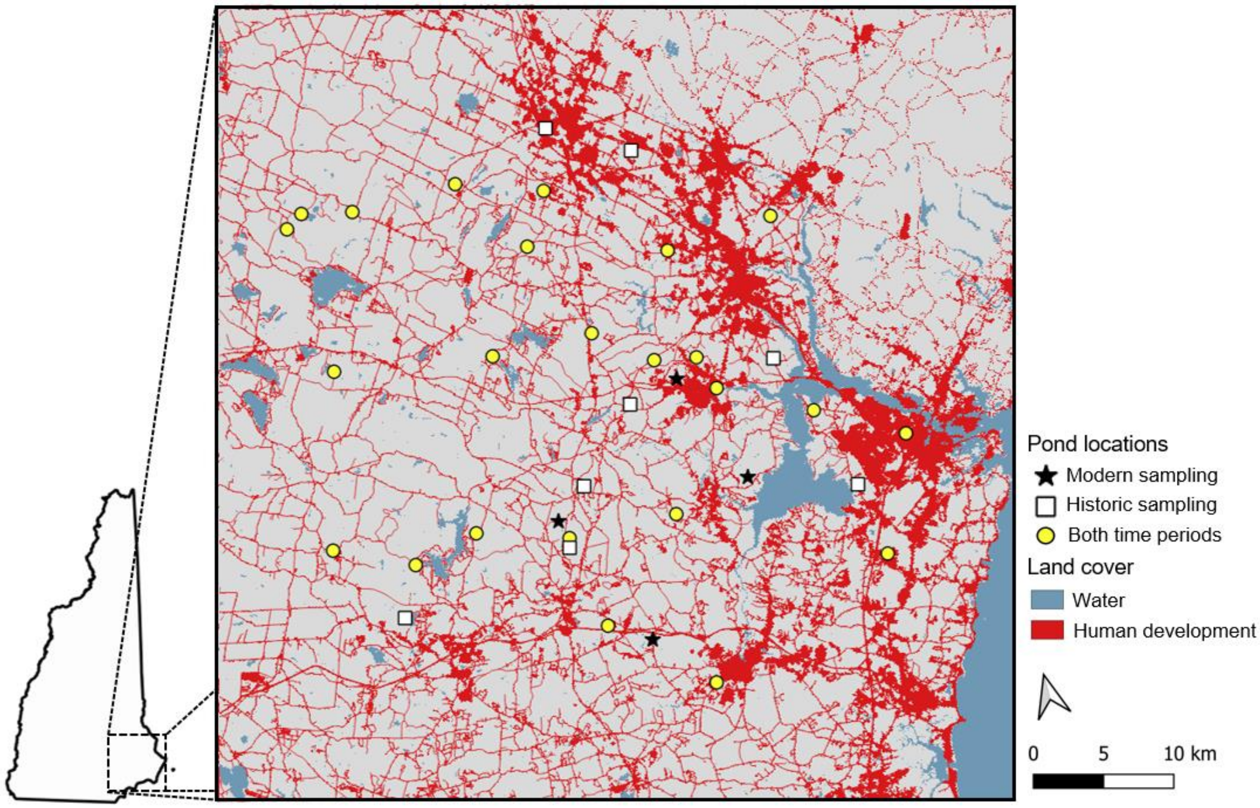

Figure 1. Turtle survey ponds along an urbanization gradient in southeastern New Hampshire, USA. Historic ponds (white squares) were sampled in 2000 or 2001 (Marchand & Litvaitis 2004). Modern ponds (black stars) were sampled in 2019, 2020, or 2021. The majority of ponds were sampled in both time periods (yellow circles). Human development (shown in red) represents areas with greater than 20% impervious surface.

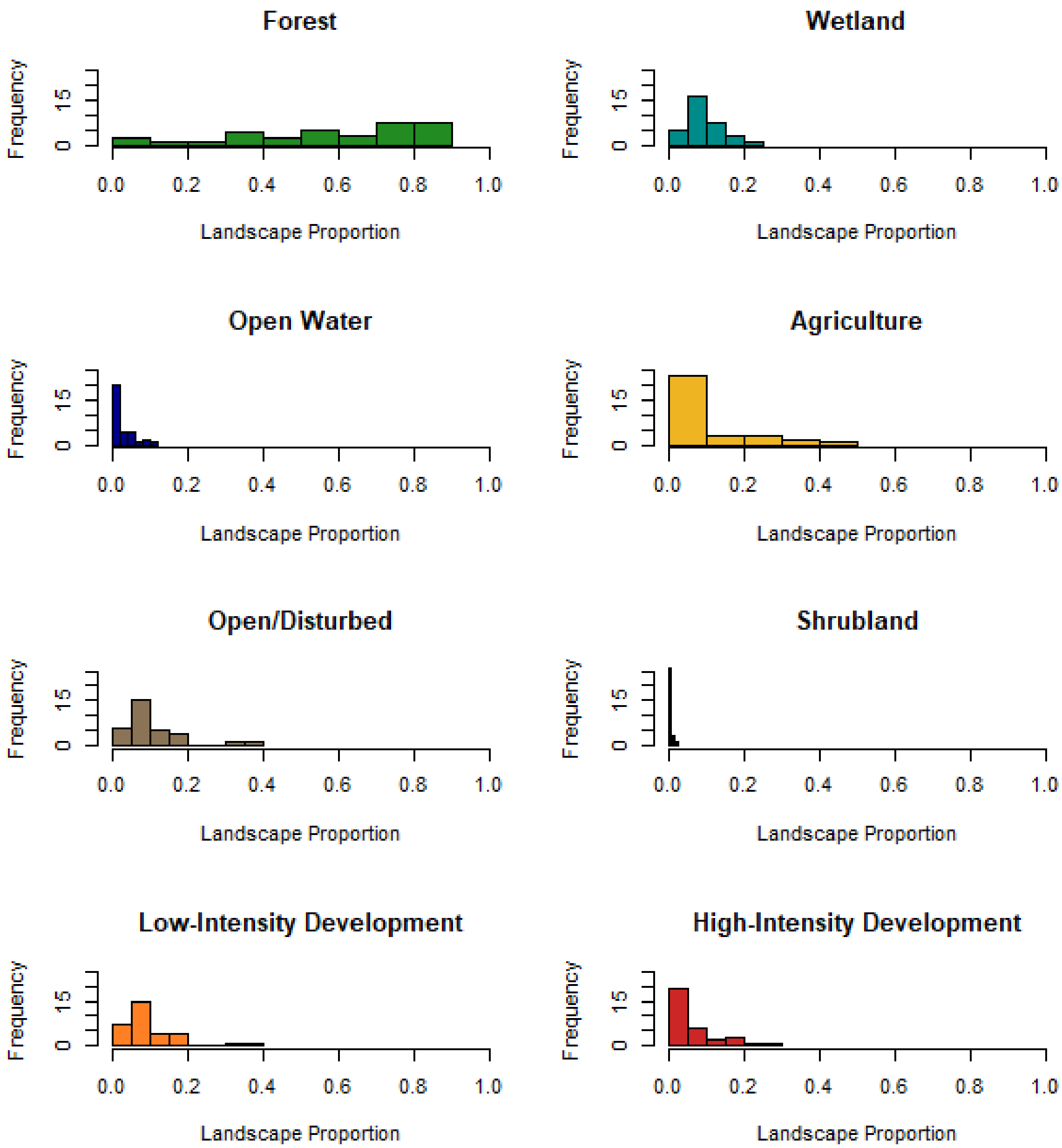

Figure 2. Frequency of historic (2001) land cover proportions within 500 meters of the pond perimeter for the 32 historic ponds in southeastern New Hampshire, USA.

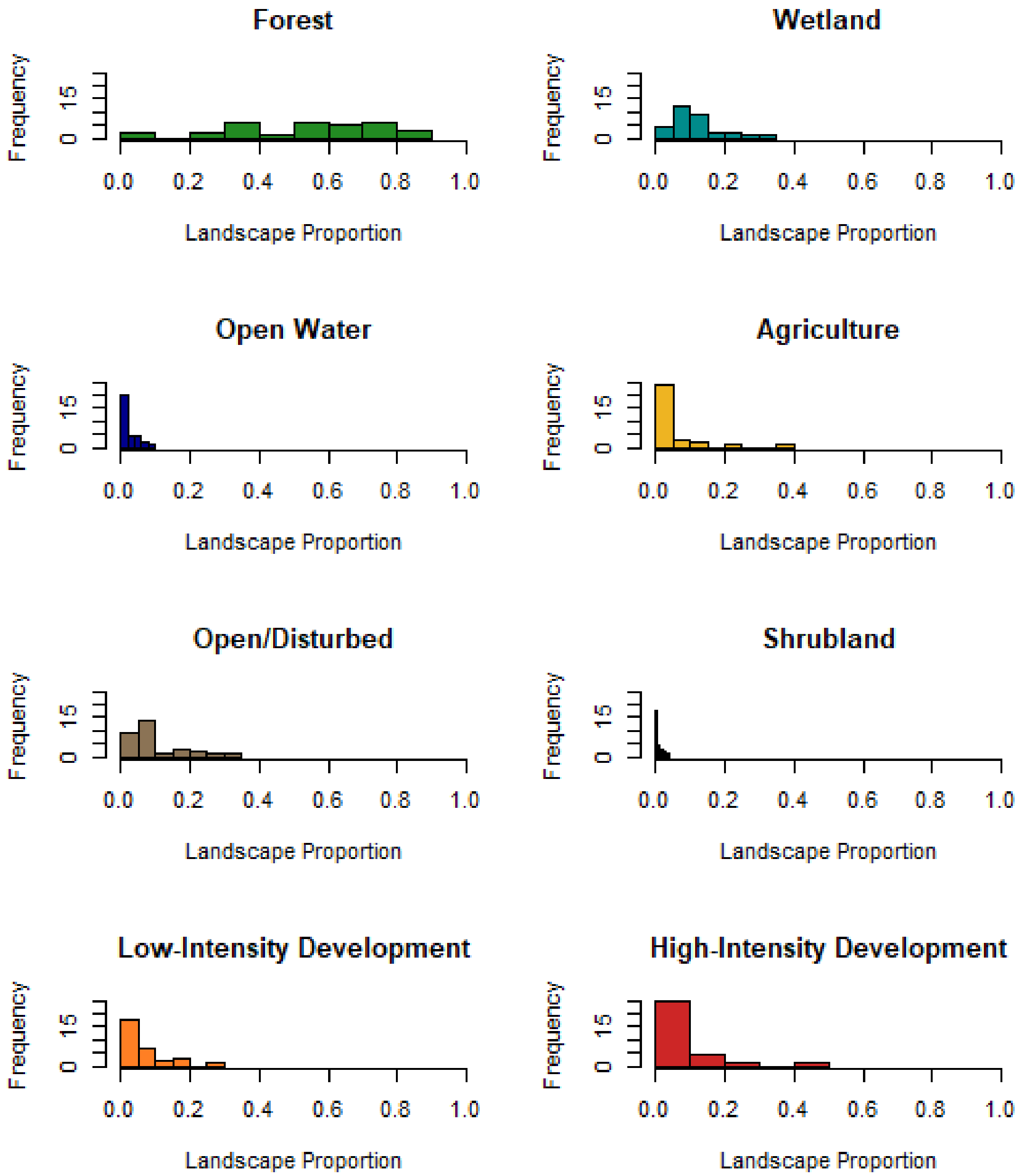

Figure 3. Frequency of modern (2019) land cover proportions within 500 meters of the pond perimeter across 31 modern ponds in southeastern New Hampshire, USA.

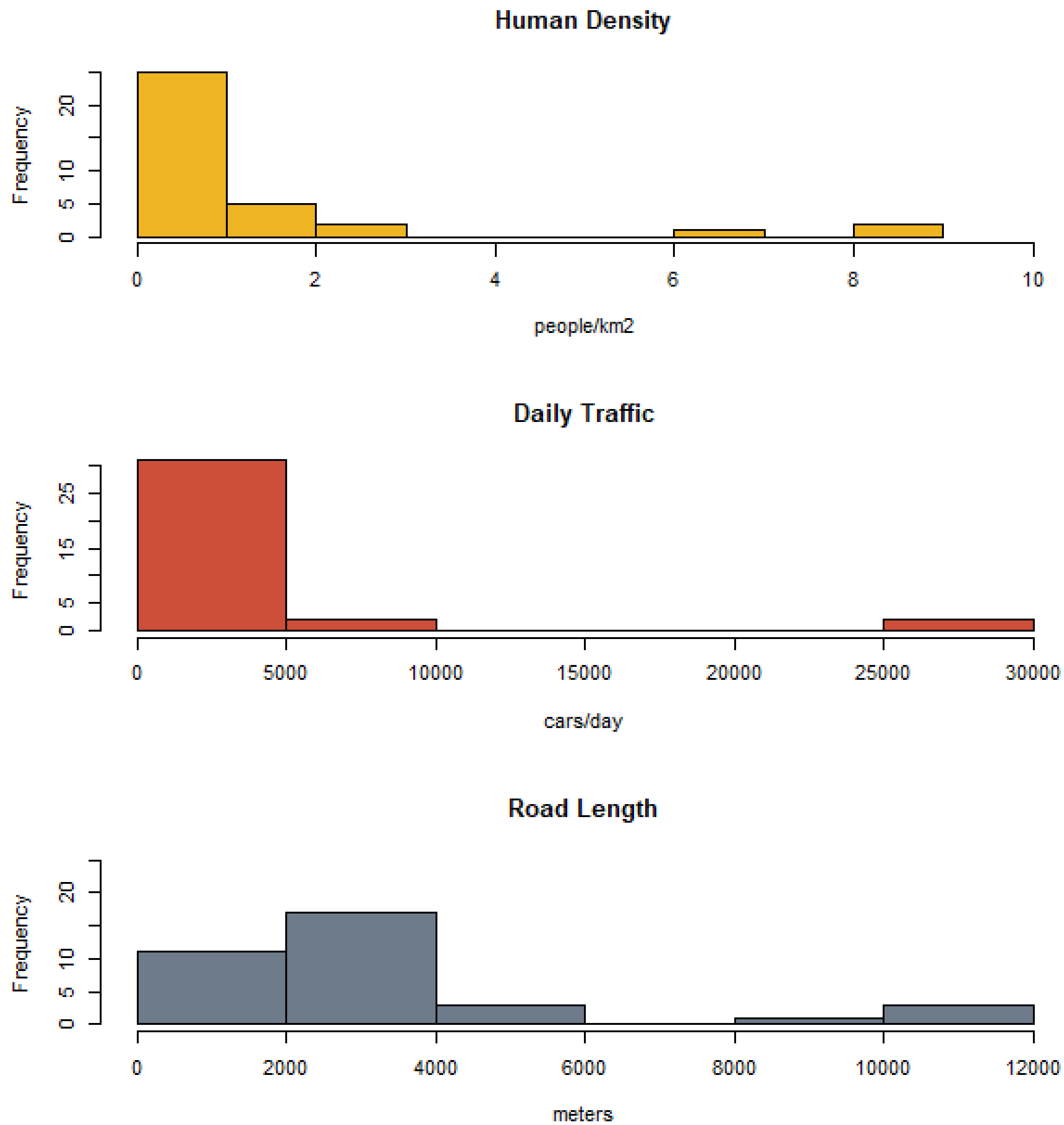

Figure 4. Frequency of historic (2001) land use covariates within 500 meters of the pond perimeter for 32 ponds in southeastern New Hampshire, USA.

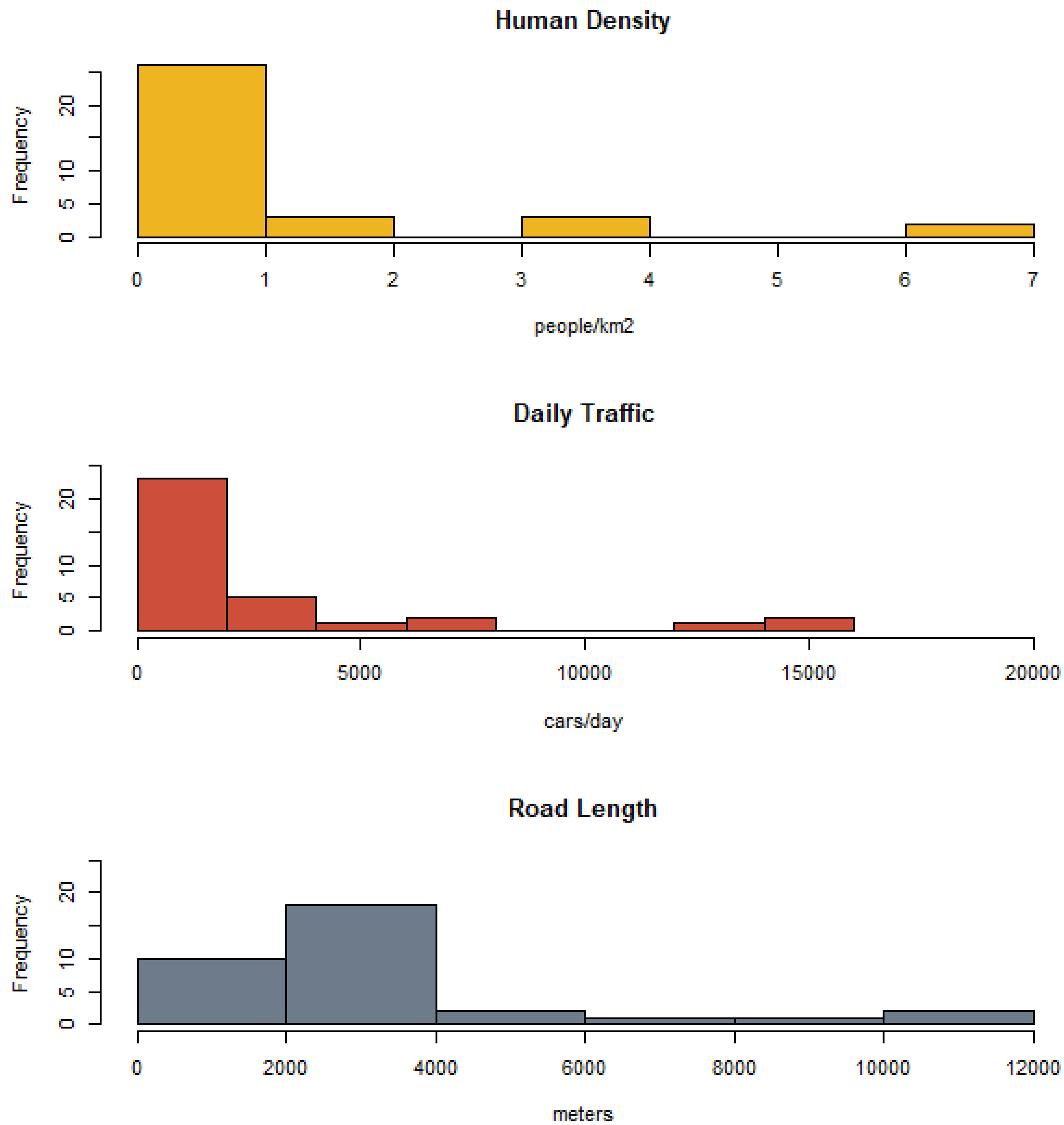

Figure 5. Frequency of modern (2019) land use covariates within 500 meters of the pond perimeter across 31 ponds in southeastern New Hampshire, USA.

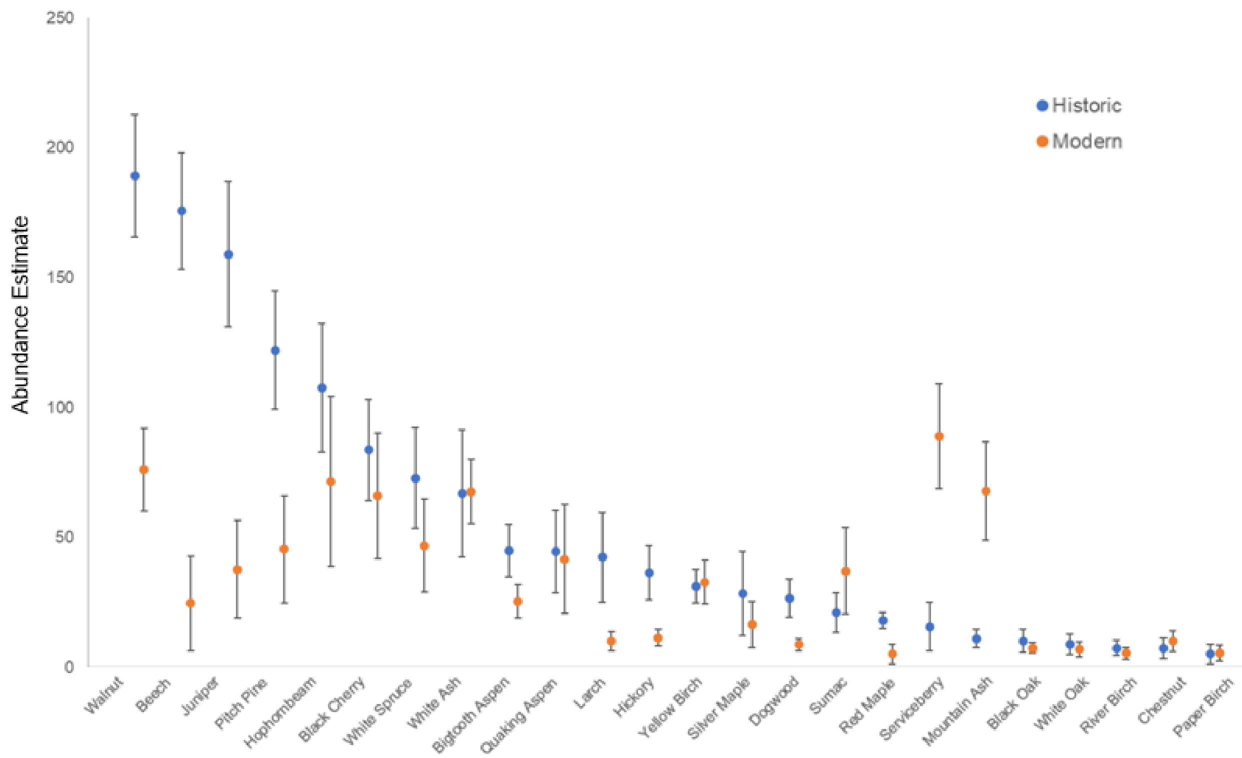

Figure 6. Historic (2000-2001) and modern (2019-2021) painted turtle abundance estimates for 24 ponds sampled in both time periods in southeastern New Hampshire, USA.

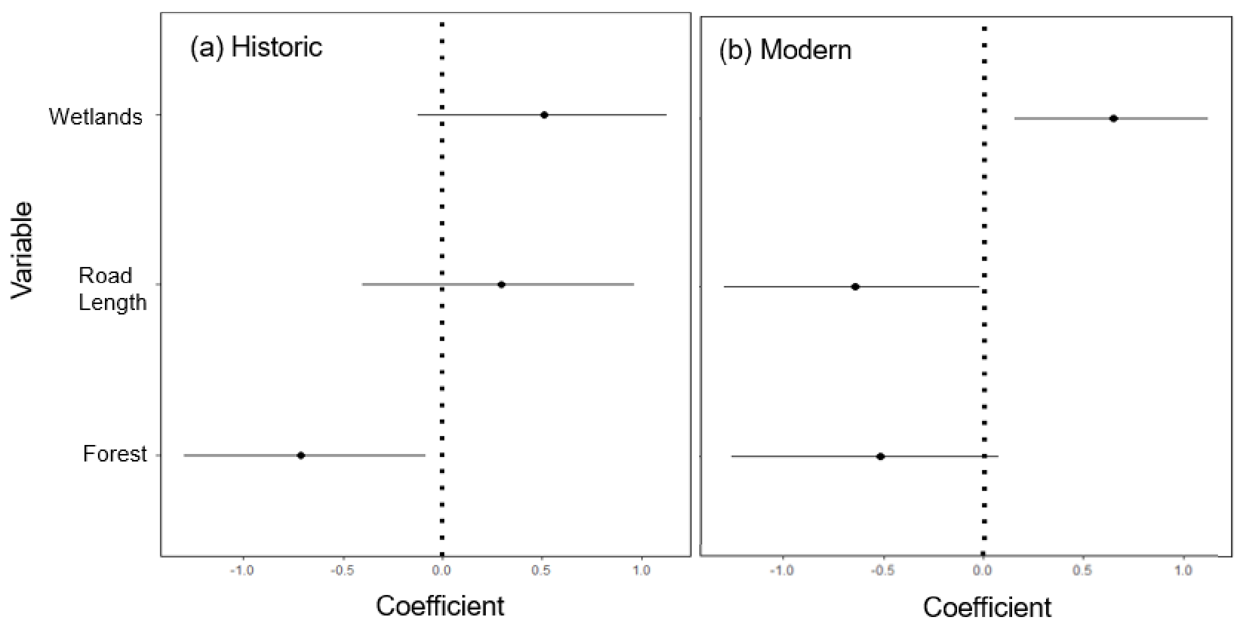

Figure 7. Model averaged parameter estimates of effect of proportion wetlands, road length, and proportion of forest on historic (2000-2001) turtle density (a) and modern (2019-2021) turtle density (b) in southeastern New Hampshire, USA. The dot represents the coefficient estimate and the lines represent the 95% credible interval.

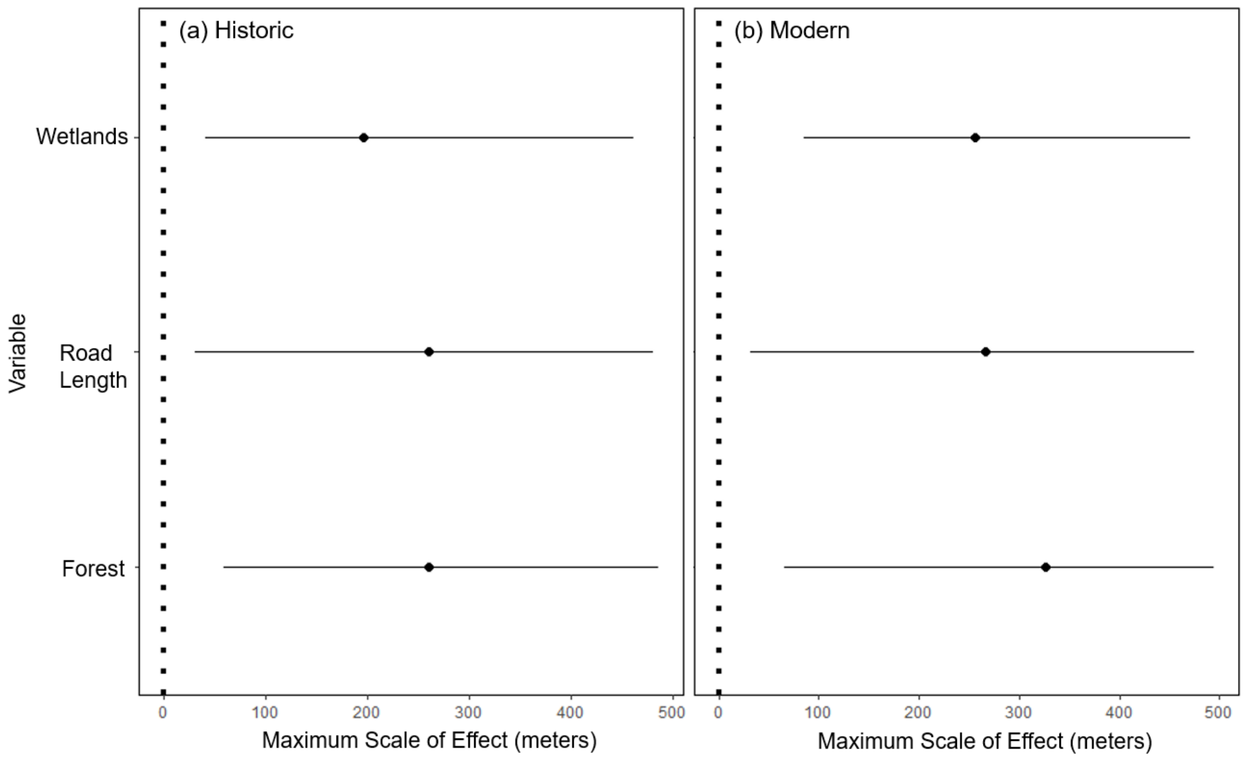

Figure 8. Model averaged parameter estimates of maximal scale of effect for proportion wetlands, proportion forest, and road length on historic (2000-2001) turtle density (a) and modern (2019-2021) turtle density (b) in southeastern New Hampshire, USA. The dot represents the maximum scale of effect estimate and the line represents the 95% credible interval.

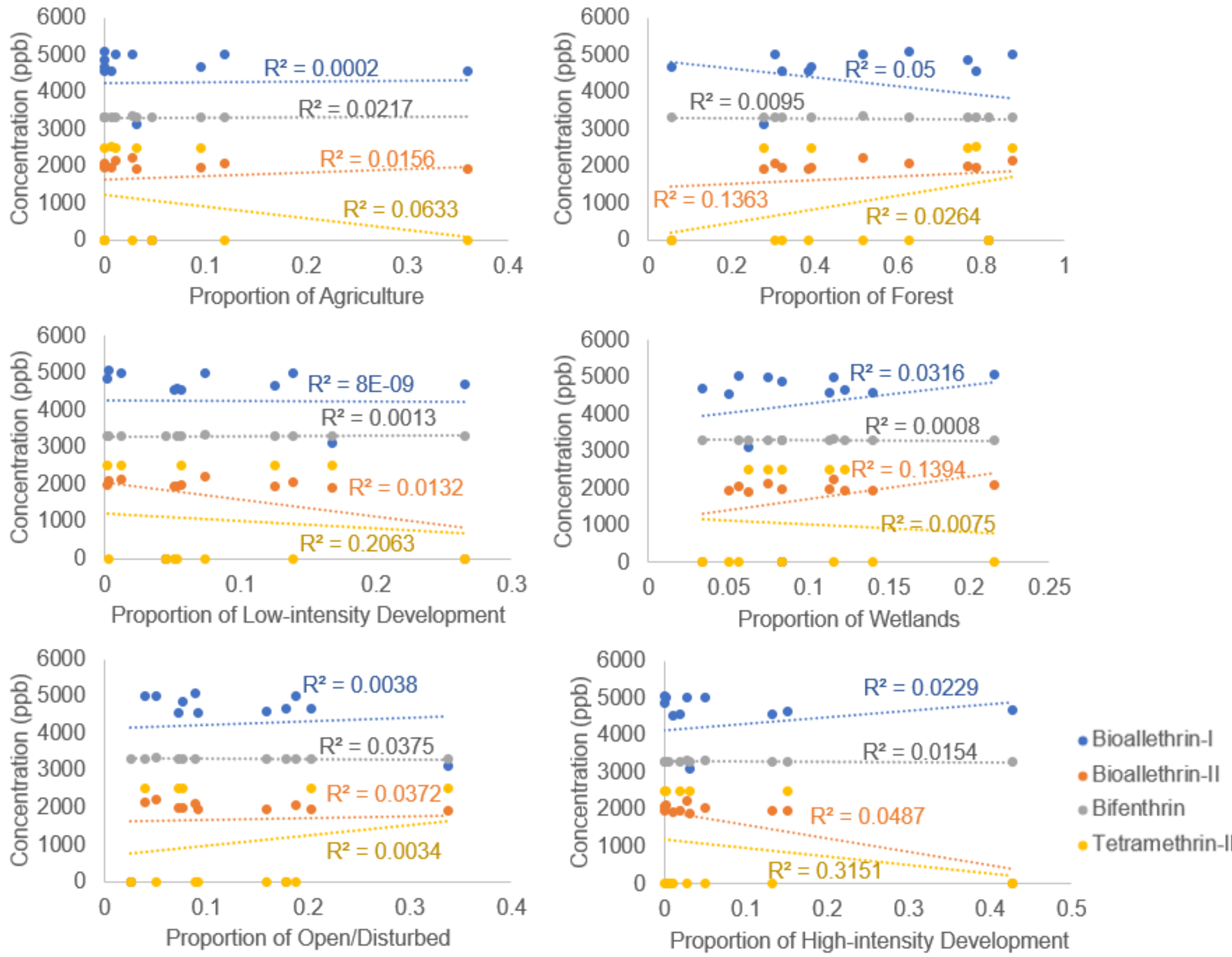

Figure 9. Linear relationship between the concentration of four synthetic pyrethroids in 30 painted turtle blood samples across 12 ponds and the proportion of six land cover classes within 500 meters of the pond perimeter. The proportion of open water and shrubland were not considered due to insufficient variation between the ponds.

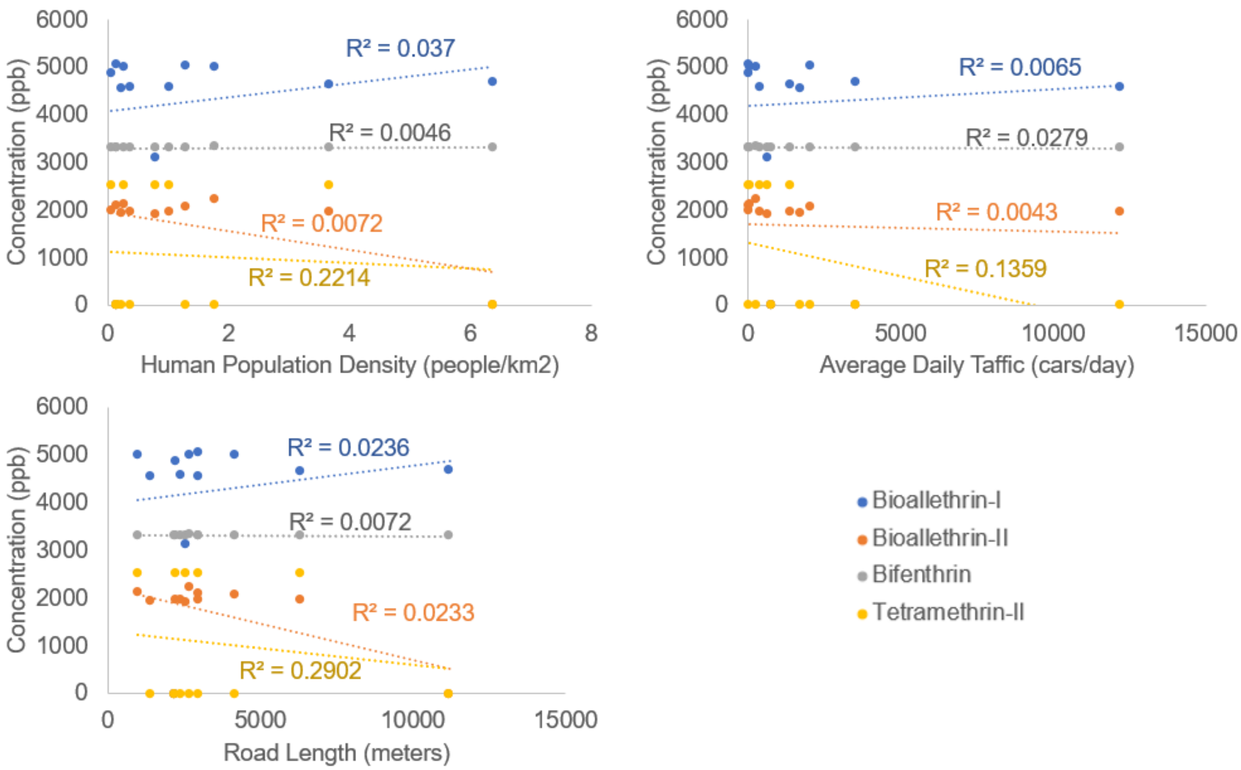

Figure 10. Linear relationship between the concentration of four synthetic pyrethroids in 30 painted turtle blood samples across 12 ponds and three land use variables within 500 meters of the pond perimeter.

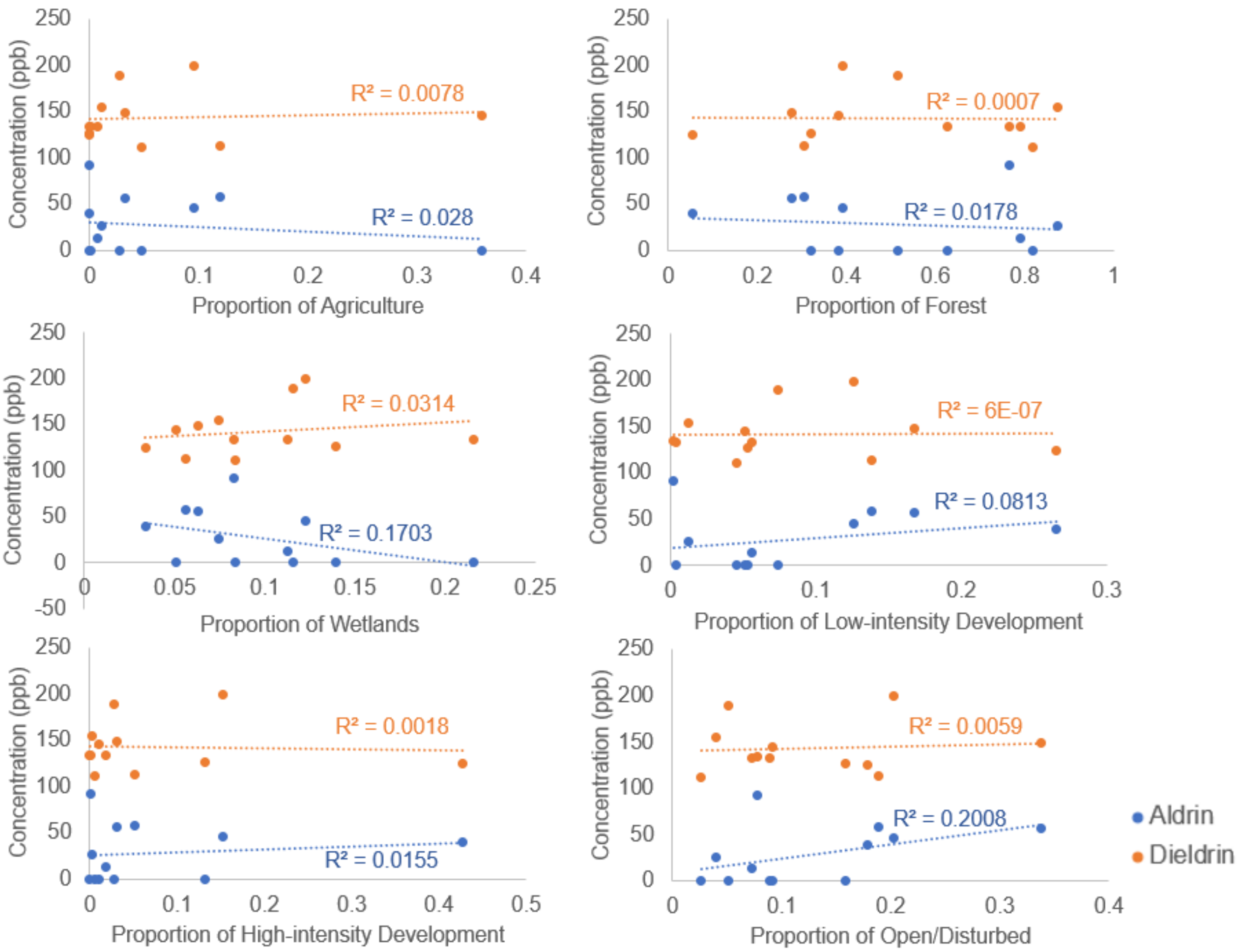

Figure 11. Linear relationship between the concentration of aldrin and dieldrin in 30 painted turtle blood samples across 12 ponds and the proportion of six land cover classes within 500 meters of the pond perimeter. The proportion of open water and shrubland were not considered due to insufficient variation between the ponds.

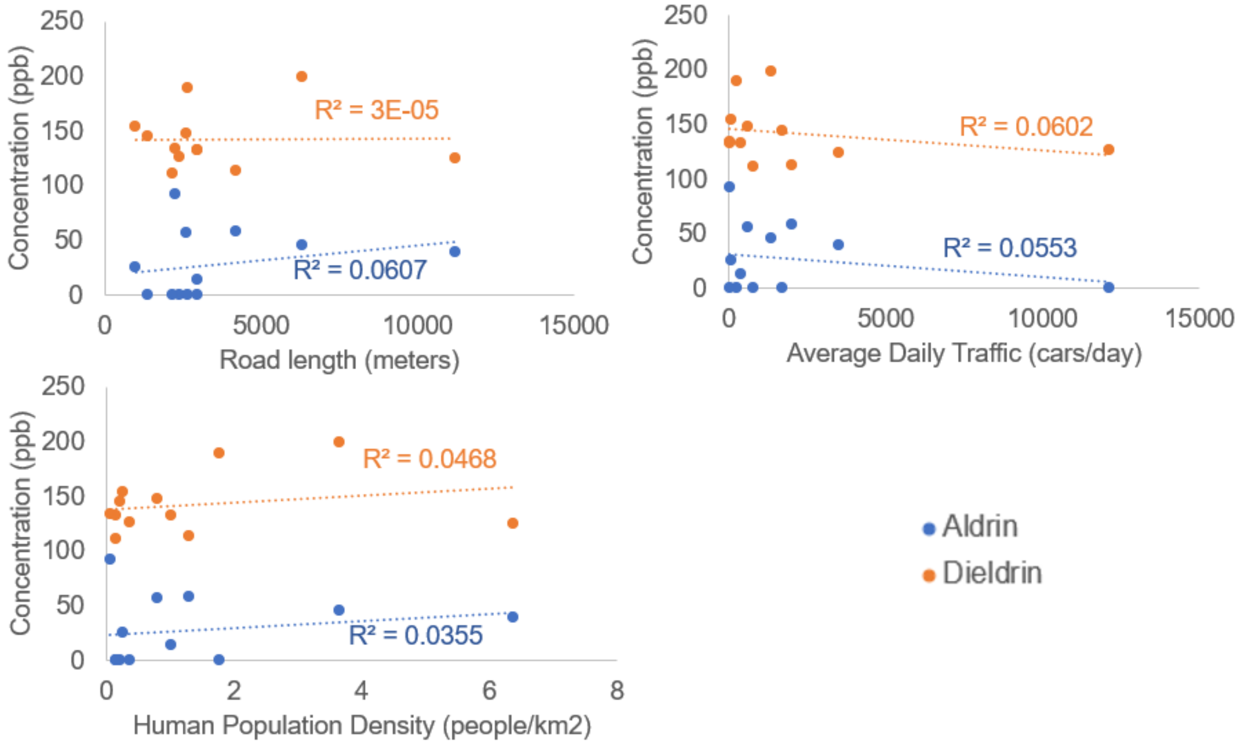

Figure 12. Linear relationship between the concentration of aldrin and dieldrin in 30 painted turtle blood samples across 12 ponds and three land use variables within 500 meters of the pond perimeter.

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APPENDIX A

IACUC Approvals for the use of Animals in Research

University *of* New Hampshire

Research Integrity Services, Service Building
51 College Road, Durham, NH 03824-3585
Fax: 603-862-3564

29-Apr-2019

Purrenhage, Jennifer
Natural Resources and the Environment
114 James Hall
Durham, NH 03824

IACUC #: 190406
Project: Re-survey of Turtle Population Structure and Land Use along an Urbanization Gradient Following 20 Years of Environmental Change
Approval Date: 15-Apr-2019

The Institutional Animal Care and Use Committee (IACUC) reviewed and approved the protocol submitted for this study under Category D in Section V of the Application for Review of Vertebrate Animal Use in Research or Instruction - *Animal use activities that involve accompanying pain or distress to the animals for which appropriate anesthetic, analgesic, tranquilizing drugs or other methods for relieving pain or distress are used.*

Approval is granted for a period of three years from the approval date above. Continued approval throughout the three year period is contingent upon completion of annual reports on the use of animals. At the end of the three year approval period you may submit a new application and request for extension to continue this project. Requests for extension must be filed prior to the expiration of the original approval.

Please Note:

1. All cage, pen, or other animal identification records must include your IACUC # listed above.
2. Use of animals in research and instruction is approved contingent upon participation in the UNH Occupational Health Program for persons handling animals. Participation is mandatory for all principal investigators and their affiliated personnel, employees of the University and students alike. Information about the program, including forms, is available at http://unh.edu/research/occupational-health-program-animal-handlers.

If you have any questions, please contact either Dean Elder at 862-4629 or Julie Simpson at 862-2003.

For the IACUC,

Rebecca Rowe

Rebecca Rowe, Ph.D.
Chair

cc: File

University *of* New Hampshire

Research Integrity Services, Service Building
51 College Road, Durham, NH 03824-3585
Fax: 603-862-3564

22-May-2020

Purrenhage, Jennifer
Natural Resources and the Environment
114 James Hall
Durham, NH 03824

IACUC #: 200503
Project: Population and Habitat Assessment of the Endangered Blanding's Turtle, an Indicator of Environmental Health, in a Human-Altered Landscape
Approval Date: 21-May-2020

The Institutional Animal Care and Use Committee (IACUC) reviewed and approved the protocol submitted for this study under Category C in Section V of the Application for Review of Vertebrate Animal Use in Research or Instruction - *the research potentially involves minor short-term pain, discomfort or distress which will be treated with appropriate anesthetics/analgesics or other assessments.*

Approval is granted for a period of three years from the approval date above. Continued approval throughout the three year period is contingent upon completion of annual reports on the use of animals. At the end of the three year approval period you may submit a new application and request for extension to continue this project. Requests for extension must be filed prior to the expiration of the original approval.

Please Note:
1. All cage, pen, or other animal identification records must include your IACUC # listed above.
2. Use of animals in research and instruction is approved contingent upon participation in the UNH Occupational Health Program for persons handling animals. Participation is mandatory for all principal investigators and their affiliated personnel, employees of the University and students alike. Information about the program, including forms, is available at http://unh.edu/research/occupational-health-program-animal-handlers.

If you have any questions, please contact either Dean Elder at 862-4629 or Susan Jalbert at 862-3536.

For the IACUC,

Julie Simpson

Julie Simpson, Ph.D.
Director

cc: File

APPENDIX B

Supplementary Tables

Table B1. Model selection using DIC for historic distance-weighted smoothing painted turtle density models. All 55 models considered are shown. for = proportion forest, wet = proportion wetlands, lid = proportion low-intensity development, opd = proportion open/disturbed, rl = road length, adt = average daily traffic, and hpop = human population density.

Land Cover		Land Use		DIC	ΔDIC	Bayes.P
Year	Covariate	Year	Covariate			
2001	for			107.81	0.00	0.53
2001	for, wet			108.01	0.20	0.54
2001	for, wet	2001	rl	109.40	1.59	0.53
2001	for, opd			109.51	1.70	0.53
2001	wet, lid			109.64	1.83	0.53
2001	for, opd, wet			109.90	2.09	0.54
2001	for	2001	rl	110.05	2.24	0.54
2001	for, lid			110.16	2.35	0.53
2001	for, wet	2001	hpop	110.27	2.46	0.55
2001	for	2001	adt	110.27	2.46	0.53
2001	wet	2001	rl	110.32	2.51	0.53
2001	for, opd	2001	rl	111.52	3.71	0.53
2001	opd, wet	2001	rl	112.14	4.33	0.53
2001	lid			112.27	4.46	0.54
2001	for, water	2001	rl	112.73	4.92	0.54
2001	wet, hid			112.94	5.13	0.54
2001	dev			113.15	5.34	0.54
		2001	rl	113.23	5.42	0.53
		2001	hpop	113.85	6.04	0.53
2001	wet	2001	adt	114.05	6.24	0.52
2001	wet			114.43	6.62	0.54
2001	water	2001	rl	114.44	6.63	0.52
2001	opd, lid			114.47	6.66	0.54
2001	hid, lid			114.48	6.67	0.54
2001	hid			114.52	6.71	0.53
		2001	adt	114.55	6.74	0.53
2001	lid	2001	rl	114.65	6.85	0.54
1970-80	for	2001	rl	114.69	6.88	0.54
1970-80	wet	2001	rl	115.18	7.37	0.53
1970-80	opd	2001	rl	115.30	7.50	0.54
1970-80	water	2001	rl	115.44	7.63	0.54
2001	opd	2001	rl	115.44	7.63	0.53

1970-80	ag			115.69	7.88	0.54
1970-80	for			115.76	7.95	0.54
2001	water			116.36	8.55	0.54
2001	ag			116.41	8.60	0.53
2001	opd, wet	2001	adt	116.50	8.69	0.54
1970-80	for	2001	adt	116.68	8.87	0.53
2001	opd			116.75	8.94	0.55
1970-80	for, wet	2001	rl	116.91	9.10	0.54
2001	opd, hid			116.93	9.12	0.55
1970-80	for, water	2001	rl	116.97	9.16	0.54
2001	shb			116.98	9.17	0.53
1970-80	wet			117.07	9.26	0.53
1970-80	for, opd	2001	rl	117.10	9.30	0.54
1970-80	dev			117.19	9.38	0.53
1970-80	wet	2001	adt	117.22	9.41	0.54
1970-80	wet, opd	2001	rl	117.24	9.43	0.52
1970-80	for, wet	2001	hpop	117.27	9.46	0.53
1970-80	water			117.30	9.49	0.53
1970-80	opd			117.35	9.54	0.54
1970-80	for, wet			118.10	10.29	0.53
1970-80	for, opd			118.34	10.53	0.53
1970-80	opd, wet	2001	adt	119.51	11.70	0.54
1970-80	for, opd, wet			120.63	12.82	0.54

Table B2. Model selection using DIC for modern distance-weighted smoothing painted turtle density models. All 64 models considered are shown. for = proportion forest, wet = proportion wetlands, lid = proportion low-intensity development, opd = proportion open/disturbed, rl = road length, adt = average daily traffic, and hpop = human population density.

Land Cover		Land Use		DIC	ΔDIC	Bayes.P
Year	Covariate	Year	Covariate			
2001	for, wet	2019	rl	99.56	0.00	0.53
2001	for, wet			99.67	0.11	0.54
2001	wet			99.75	0.20	0.53
2019	wet			100.43	0.87	0.55
2019	for, wet			100.95	1.39	0.54
2001	wet, lid			101.21	1.65	0.54
2001	for	2019	rl	101.22	1.67	0.53
2001	for, opd	2019	rl	101.62	2.07	0.53
2001	wet, hid			101.69	2.13	0.54
2019	for, wet	2019	rl	101.76	2.20	0.53
2001	for, wet	2019	hpop	101.92	2.37	0.54
2019	wet, lid			102.07	2.51	0.53
2001	wet	2019	rl	102.10	2.54	0.54
2001	for, lid			102.15	2.59	0.52
2019	wet, hid			102.17	2.61	0.53
2001	for, opd, wet			102.43	2.88	0.54
2019	for	2019	rl	102.54	2.99	0.54
2001	wet	2019	adt	102.55	3.00	0.53
2019	wet	2019	adt	102.78	3.22	0.54
2001	opd, wet	2019	rl	102.79	3.24	0.54
2019	wet			103.23	3.67	0.53
2001	opd, wet	2019	adt	103.33	3.77	0.53
2019	for, opd, wet			103.62	4.06	0.53
2019	for, lid			103.64	4.08	0.54
2019	for, opd	2019	rl	103.64	4.08	0.55
2019	for, wet	2019	hpop	103.66	4.10	0.54
2019	shb			103.81	4.26	0.53
2001	for, water	2019	rl	104.21	4.65	0.55
2019	opd, wet	2019	rl	104.47	4.91	0.53
2019	for, water	2019	rl	104.63	5.07	0.54
2001	for			105.00	5.44	0.53
2019	opd, wet	2019	adt	105.01	5.45	0.54
2019	for			105.79	6.24	0.54
2001	for, opd			106.28	6.72	0.54
2001	for	2019	adt	106.31	6.75	0.54
2019	for	2019	adt	106.97	7.41	0.54
2019	for, opd			107.39	7.84	0.53

2019	opd			107.76	8.20	0.54
2019	hid			107.88	8.32	0.53
2001	opd			107.95	8.39	0.54
		2019	adt	107.99	8.44	0.53
2019	dev			107.99	8.44	0.54
2001	hid			108.05	8.49	0.53
2001	ag			108.07	8.52	0.53
2019	lid			108.08	8.53	0.54
2019	water			108.19	8.63	0.53
2001	dev			108.21	8.65	0.54
2001	shb			108.21	8.66	0.53
2019	lid	2019	rl	108.22	8.66	0.55
2001	lid			108.22	8.66	0.54
		2019	rl	108.40	8.84	0.54
		2019	hpop	108.50	8.95	0.54
2019	ag			108.55	9.00	0.54
2001	water			108.66	9.11	0.53
2001	lid	2019	rl	108.85	9.29	0.54
2001	opd	2019	rl	109.70	10.14	0.53
2019	opd, hid			109.74	10.19	0.54
2019	opd, lid			110.03	10.48	0.54
2001	opd, hid			110.28	10.72	0.54
2001	adt, lid			110.28	10.73	0.53
2019	adt, lid			110.33	10.77	0.53
2001	opd, lid			110.39	10.84	0.54
2019	hid, lid			110.44	10.89	0.53
2001	hid, lid			110.53	10.97	0.54
2001	water	2019	rl	110.55	11.00	0.53

APPENDIX C

R Code

Appendix C.1 Bayesian closed-population capture-mark-recapture model with detection probability varying by trap occasion.

```
# Parameter-expanded Data Augmentation

nz <- 150 # Number of lines added.

T <- as.numeric(ncol(CH)) # T = Number of sample occasions, CH = site capture history

yaug <- rbind(CH, array(0, dim = c(nz,T)))


# Specify model in BUGS language

sink("model.jags")

cat("

model {

# Priors

omega ~ dunif(0, 1)

for (i in 1:T){

  p[i] ~ dunif(0, 1)

  }


# Likelihood

for (i in 1:M){

  z[i] ~ dbern(omega)

  for (j in 1:T){

    yaug[i,j] ~ dbern(p.eff[i,j])

    p.eff[i,j] <- z[i] * p[j]

    } #j

  } #i


# Derived quantities
```

```
N <- sum(z[])
} # end model
",fill = TRUE)
sink()

# Bundle data
win.data <- list(yaug = yaug, M = nrow(yaug), T = ncol(yaug))

# Initial values
inits <- function() list(z = rep(1, nrow(yaug)), p = runif(T, 0, 1))

# Parameters monitored
params <- c("N", "p", "omega")

# MCMC settings
ni <- 2500; nt <- 2; nb <- 500; nc <- 3

# Call JAGS from R (BRT <1 min)
out <- jags(win.data, inits, params, "model.jags", n.chains = nc, n.thin = nt, n.iter = ni, n.burnin =
nb, working.directory = getwd())
```

Appendix C.2 Example Bayesian distance-weighted smoothing model for the effect proportion forest on turtle density. The same priors and MCMC settings were used for all historic (2000-2001) and modern (2019-2021) turtle density models.

```
# JAGS model

sink("landscape.d_model_for.txt")

cat("

  model {


  # PRIORS

  B0 ~ dnorm(0,0.001)

  B_FOR ~ dnorm(0,0.001)

  sigma_for ~ dunif(1,500)

  tau <- 1/sigma^2

  sigma ~ dunif(0,10)



  # LIKELIHOOD

  for(i in 1:n){

  N[i] ~ dnorm(mu[i],tau)


  mu[i] <- B0 + B_FOR * FOR_w[i]


  # Calculate Ring Weights, (bufs = distance to ring around sites)

  for (u in 1:U){

  w0_for[i,u] <-exp(-bufs[i,u]^2 /(2*sigma_for^2)) * ring_area[i,u]

  }


  w_for[i,1:U] <- w0_for[i,1:U]/sum(w0_for[i,1:U])


  # Calculate Weighted Land Cover / Land Use Covariates
```

```
    FOR_w[i] <- sum(w_for[i,1:U]*for_m[i,1:U])

    # CHECK MODEL FIT
    N.new[i] ~ dnorm(mu[i], tau)

    sq.data[i] <- (N[i] - mu[i])^2
    sq.new[i] <- (N.new[i] - mu[i])^2

    } # i

    # CHECK MODEL FIT - BAYESIAN P-VALUE
    sos.data <- sum(sq.data[])
    sos.new <- sum(sq.new[])
    Bayes.P <- step(sos.new/sos.data -1)

    } # end of model
    ",fill = TRUE)
sink()

win.data <- list(N = log(turtle.dat$N),
                 n  = nrow(turtle.dat),
                 U = ncol(for_rings),
                 for_m = for_rings,
                 bufs = bufs,
                 ring_area = ring_area)



# Initial values
inits <- function()list(B0  = rnorm(1,30),
                        B_FOR  = rnorm(1),
```

```
                        sigma_for = 50)


# Parameters monitored
params <- c("B0",
            "B_FOR",
            "sigma_for",
            "Bayes.P")

# MCMC settings
ni <- 5000; nt <- 1; nb <- 500; nc <- 3

out.d_for <- jags(win.data, inits, params, "landscape.d_model_for.txt",
                n.chains = nc, n.thin = nt, n.iter = ni, n.burnin = nb,
                working.directory = getwd())
```