UNIVERSIDADE FEDERAL DO RIO GRANDE DO SUL

INSTITUTO DE BIOCIÊNCIAS

PROGRAMA DE PÓS-GRADUAÇÃO EM ECOLOGIA

Dissertação de Mestrado

PRESSÃO ANTROPOGÊNICA AFETA A NIDIFICAÇÃO DE TRACAJÁS

(*Podocnemis unifilis*) NA AMAZÔNIA ORIENTAL?

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Porto Alegre, Agosto 2018

Pressão antropogênica afeta a nidificação de Tracajás (*Podocnemis unifilis*) na Amazônia Oriental?

Itxaso Quintana Garcia

Dissertação de Mestrado Apresentada ao Programa de Pós-Graduação em Ecologia, do Instituto de Biociências da Universidade Federal do Rio Grande do Sul, como parte dos requisitos para obtenção do título de Mestre em Ecologia.

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Porto Alegre, Agosto 2018

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Does anthropogenic pressure affect nesting of the yellow-spotted river turtle (*Podocnemis unifilis*) in the eastern Brazilian Amazon?

Short title: Anthropogenic effect on nesting of *Podocnemis unifilis*

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Resumo

As tartarugas na Amazônia são particularmente sensíveis à pressão humana, já que os impactos nas áreas de nidificação podem afetar negativamente a taxa de reprodução das populações. A seleção do local de nidificação é um componente fundamental para o sucesso de desova, no entanto, a associação entre a escolha do local e a aptidão do local escolhido ainda é pouco estudada. Neste trabalho testamos um conjunto de variáveis para explicar os padrões de nidificação em Tracajás (*Podocnemis unifilis*), com base em dados coletados em 73 locais de nidificação ao longo de 118 km de rio na Amazônia Oriental Brasileira. Modelos lineares generalizados (GLMs) foram usados para avaliar as influências antropogênicas e ambientais nos padrões de seleção do local de nidificação (número de ninhos, densidade de ninhos, distância do ninho à água e variação na distância à água) e adequabilidade do local de nidificação (remoção de ninhos por humanos). Como resultado, encontramos que o número e densidade de ninhos foram fortemente explicados por variáveis ambientais, e a distancia do ninho á água diminuiu com a proximidade as casas. A remoção humana foi o principal responsável pela perda de ninhos (47% dos ninhos foram removidos) principalmente nos trechos do rio com maior atividade humana. Nossos dados sugerem que mudanças antropogênicas estão levando a diferenças entre a escolha de local de nidificação e a sua adequabilidade, assim, nos rios onde humanos tem maior acesso, sinais usados pelas fêmeas para selecionar áreas de nidificação não estão permitindo desovas bem-sucedidas. Ao contrário das previsões nas hipóteses, os achados demonstram que as fêmeas não parecem evitar desovar em locais perigosos e inadequados. Portanto, ações diretas, como a proteção de áreas de nidificação, são vitais para a conservação das tartarugas na Amazônia.

Palavras-chave: Áreas protegidas, Réptil, Remoção de ninhos, Conservação de tartarugas, Impacto humano.

Abstract

Amazonian freshwater turtles are particurlarly sensitive to human pressure, since impacts on their nesting areas can negatively affect reproductive rate of populations. Nest-site selection is a fundamental component of freshwater turtle nesting success, however, linking oviposition choices to overall suitability of nest-site selection remains poorly tested. We tested a set of variables to explain nesting patterns in the yellow-spotted river turtle *(Podocnemis unifilis)*, based on data collected from 73 nesting sites along 118 km of river in the eastern Brazilian Amazon. General Linear Models (GLMs) were used to evaluate anthropogenic and environmental influences on patterns in nest-site selection (four responses: number of nests, nest density, distance from nest to water and variation in distance to water) and nest-site suitability (removal of nests by humans). Number and density of nest were largely explained by environmental variables, and distance from nest to water decreased with the proximity to human habitations. Human removal of nests was the primary driver of nest failure (47% of the nests were removed) and removal was higher in sections of river with higher human activity. We show that anthropogenic changes are driving differences between nest-site selection and suitability, whereby the signals used by females to select nesting areas no longer enable successful nesting along rivers accessible to humans. Contrary to predictions from the hypotheses, our findings demonstrate that females do not appear to avoid nesting in dangerous and unfit sites. Therefore, direct actions including the protection of nesting areas are vital for the conservation of Amazonian freshwater turtles.

Introduction

One of the major challenges in conservation is to understand how anthropogenic pressure is affecting wildlife populations (Gill, Sutherland, & Watkinson, 1996). Biodiversity hotspots and tropical wilderness areas are suffering a disproportioned rapid human growth pace (Williams, 2013) and therefore the encounters between humans and wild animals are becoming more frequent in remote areas (De Oliveira, Norris, & Michalski, 2015). It is essential to understand the patterns and effects of human impacts to inform conservation planning, especially in tropical developing countries where data gaps are still a challenge (Collen, Ram, Zamin, & Mcrae, 2008; Meyer, Kreft, Guralnick, & Jetz, 2015).

The Amazon basin has experienced a burgeoning migration and population growth (Carr, Lopez, & Bilsborrow, 2009) mostly concentrated along water-ways (Peres, 2011). Accessibility to freshwater systems brings aquatic fauna into a vulnerable situation (Castello et al., 2013; Antunes et al., 2016), as is the case of river turtles, which are considered endangered in the Amazon (Smith, 1979), and are one of the most threatened group of vertebrates all over the world (Gibbon et al., 2000; Turtle Conservation Coalition, 2018),with almost 52% of the species being classified at some category of threaten (Böhm et al., 2013). Additionally, freshwater turtles represent provisioning (food, source of income) and cultural services (medical, ornamental) for Amazonian communities (Alves et al., 2012;). Hence, they are a highly pertinent example of the challenge facing conservation of common pool resources.

According to Vogt (1994), the understanding of the reproductive ecology plays an important role in conservation strategies. For chelonians, nest-site selection is a key process for reproduction success (Refsnider & Janzen, 2010), since oviposition site affects embryo survival (Pignati, Fernandes, Miorando, Ferreira, et al., 2013a), hatchling performance (Micheli-Campbell, Campbell, Cramp, Booth, & Franklin, 2011), offspring sex-ratio (Vogt &

Bull, 1984), as well as the survival of the nesting female (Spencer, 2002). Several strategies for nest-site selection have been described (Refsnider & Janzen, 2010). Due to the lack of parental care in reptiles, embryonic development depends on incubation conditions (Bujes & Verrastro, 2009), in particular temperature and moisture, which are mainly influenced by landscape and substrate characteristics of the nesting area (Ferreira-Júnior & Castro, 2003; Pignati et al., 2013a).Therefore, females may chose nesting sites that present suitable environmental conditions for development of the embryo (Hughes & Brooks, 2006). However, they must decide between maximizing offspring fitness and minimizing their own mortality (Spencer & Thompson, 2003), thus, factors affecting female survival may ultimately drive maternal nest-site selection, by reducing the time spent on land in high-risk areas (Spencer, 2002; Bermúdez-Romero, Castelblanco-Martínez, Bernhard, Duque, & Vogt, 2014).

Although many studies have examined oviposition decision mechanisms in chelonians, still linking nest-site choice to the overall suitability of the selected sites remains poorly tested (Refsnider & Janzen, 2010), that is, do chosen sites differ in maternal or hatchings survival, especially considering the unprecedented anthropogenic impacts across freshwater habitats (Sala et al., 2000; Dudgeon et al., 2006). Human modifications in nesting habitats can lower the reproductive fitness of females (Kolbe & Janzen, 2002). Moreover, turtles are extremely vulnerable to human disturbances near nesting sites (Alho, 2011), affecting female's reproductive activities (Moore & Seigel, 2006) and suffering displacement from preferred environments (Horne, Brauman, Moore, & Seigel, 2003).

For understanding the consequences of human pressure in the nesting process, we studied the yellow-spotted river turtle (*Podocnemis unifilis*). This Amazonian turtle is the most widespread species of the *Podocnemis* genus, being distributed along the Amazon and Orinoco river basins (Vogt, 2008; Vogt et al., 2015). Although its wide distribution, the

species is categorized as Vulnerable in the IUCN Red List (Tortoise & Freshwater Turtle Specialist Group, 1996) due to human persecution and habitat destruction. *P. unifilis* has been persecuted since pre-colonial times for the eggs and meat (Johns, 1987), and even nowadays eggs and adults are still consumed by indigenous and riverine communities (Smith, 1979; Pantoja-Lima et al., 2014). The yellow-spotted river turtle is a long-lived turtle with a relative fast maturing (i.e., 5 years old females lay eggs) (Norris, Peres, Michalski, & Gibbs, 2018c) compared to some of its congeners (Vogt, 2008). Females are thought to lay eggs once a year, coinciding with the seasonal periods of low water levels (Thorbjarnarson, Perez & Escalona, 1993). Several studies describe nesting ecology and environmental characteristics affecting nest-site selection in *P. unifilis* (Ferreira-Júnior & Castro, 2003; Escalona, Valenzuela, & Adams, 2009; Pignati, Fernandes, Miorando, Ferreira, et al., 2013b).This turtle is considered generalist for nest-site selection (Vogt & Flores-Villela, 1986), since it is not specially demanding about reproductive niches (Pantoja-Lima et al., 2009; Alho, 2011), and nests of *P. unifilis* can be found in a broad variety of substrates (Fachín-Terán & Von-Mülhen, 2003), and in different nesting habitats (Soini, 1994). Despite all its importance and threats there is still a lack of knowledge about the anthropogenic impact in nesting patterns.

We investigated the anthropogenic and environmental effects on nest-site selection as well as nest removal by humans of the yellow-spotted river turtle (*Podocnemis unifilis*) in sustainable use protected areas in the eastern Brazilian Amazon. We hypothesized that: 1) anthropogenically disturbed sites will present lower number and density of turtle nests, because turtles avoid human disturbances, 2) nest distance to water decreases in sites with higher anthropogenic pressure, as a response of turtle females to reduce exposition time, and 3) removal rates of nests by humans is higher closer to human settlements and in areas with more intense human activities. Finally, we identify the most important anthropogenic and environmental variables affecting nesting of the yellow-spotted river turtle, and make some

general points in order to direct and enhance conservation efforts for river turtles in the Brazilian Amazon.

Methods

Study area

The study was conducted in the Araguari river basin, located in the state of Amapá, eastern Amazon, northern Brazil (Figure 1). Data was obtained in two different river stretches, Araguari and Falsino rivers. Both stretches are located between two sustainable-use protected areas, the Amapá National Forest (hereafter FLONA) and the Amapá State Forest (hereafter FLOTA). Falsino and Araguari are important rivers in the region, since they are the main transportation waterways for local communities, as well as a food source and water supply (ICMBIO, 2014). The nearest town, Porto Grande, is located ~50 km from the study area and has 16809 inhabitants (IBGE, 2010). Even if turtle eggs consumption is forbidden by law (ICMBIO, 2014), eggs harvesting is still a threat to the yellow spotted river turtle population in the study area (Norris & Michalski, 2013).

The two river stretches differ in human activity. On one hand, in the Falsino river anthropogenic pressure is relatively low, because it has a lower number of riverine residents (De Oliveira et al., 2015). On the other hand, the Araguari river is characterized by higher boat traffic and more intense fishing activities (De Oliveira et al., 2015), since commercial fishing by the Fishing Fleet from Porto Grande is only allowed in Araguari river (ICMBIO, 2014).

Climate in the area is characterized as "Am" (Equatorial monsoon) according to Köppen-Geiger's classification (Kottek, Grieser, Beck, Rudolf, & Rubel, 2006). Temperature oscillates between 22ºC and 32ºC, remaining constant all over the year (INMET, 2018). The dry season takes place between September and November (total monthly rainfall < 150mm),

and the wet season takes place from February to April (total monthly rainfall > 300mm) (Paredes, Norris, De Oliveira, & Michalski, 2017).

Turtle nests monitoring

Between October and December 2017, field surveys were conducted looking for potential nesting sites, along all the navigable extension of Falsino and Araguari rivers, 72 km and 46 km, respectively. These months correspond to the nesting season of *P. unifilis* in the north-eastern region of the Amazon (Pignati et al., 2013b; Arraes, Cunha, & Tavares-Dias, 2016; Norris, Michalski, & Gibbs, 2018a), when water level decreases, and sand banks appear. Nesting sites were identified as areas of at least 5 m^2 of exposed sand and/or fine gravel, sufficiently raised above the river level not to be waterlogged at a depth of 15 cm (a representative depth that females dig when nesting) (see Figure A1) (Escalona et al., 2009; Pignati et al., 2013b; Norris et al., 2018a). All potential nesting sites were sampled, in total 73 nesting sites, being 28 in the Araguari and 45 in the Falsino river.

In all nesting sites, active nest search was done together with two local residents with over 30 years of knowledge on nesting areas. To minimize possible observer related biases, at least one observer was constantly maintained in the team throughout the entire study period.

Logistical limitations hampered our capacity to visit all nesting sites more than once. Particularly, during the monitoring season some of the sites were difficult to access due to the low water level and remoteness. Thus, in order to standardize the number of visits for each nesting area, we only included data from a single visit done the first fortnight of November, because is when the nesting and human removal reach their peak (see Figure A2). We then are confident to assume that the data collected in those dates were a significant representation of our sampling universe.

Response variables

For analysing nest-site selection and nest-site suitability patterns in yellow-spotted river turtle, we recorded five different nesting variables for each of the nesting sites: (1) nest number, (2) nest density, (3) median distance from nest to water, (4) standard deviation of distance to water, and (5) proportion of nests removed by humans.

For nest-site selection variables, the total number of nests was counted in each nesting site and density of nests was calculated considering the area. Surface nesting area was mapped using a handheld GPS, *in situ*, at the same period of the nesting season. Rocks and dense vegetation were excluded from the calculation of the area in order to only measure the available nesting area. In addition, the distance from each nest to the water's edge was measured with a measuring tape, and the median and standard deviation distance for each nesting site was calculated, to take into consideration the variability in distance to the water within the nesting site.

For nest-site suitability, in each nesting site the total number of nests removed and non-removed by humans was calculated. Human removal of eggs was identified when an open nest was found, with a depth between 10-15 cm but without the presence of eggs. Removal by humans was also associated with signs of human activities in the nesting areas, such as footprints, fire, campsite or trash. Non-removed nests were considered as the intact nests and the nests predated by wild animals. Naturally predated nests were identified by the presence of broken eggshells and/or partially eaten eggs around the nest, and when animal excavation marks were present. For nest removal analysis, four nesting sites, in which nests were protected to avoid natural and human predation, were not taken into account.

Environmental and anthropogenic explanatory variables

Since nesting can be potentially affected by local environmental features and/or

anthropogenic pressure, we obtained data for variables representing both factors.

Four environmental variables describing nesting sites were recorded (Table 1): (1) nesting site type was either island or margin, (2) presence of trees was considered when at least one tree was creating shade into the nesting area (see Figure A1), (3) main substrate was visually determined based on a categorical classification of the dominant type of substrate, defined as fine or coarse sand (see Figure A1), and (4) diversity of substrate was characterized as the number of different substrate types (i.e., fine sand, coarse sand, sand with pebbles and leaf litter) present in the nesting site.

To understand the impacts of human disturbances in nesting patterns, four anthropogenic variables were measured (Table 1). The first one, river stretch, represents different intensities in human activity (i.e., high for Araguari and low for Falsino). The other three variables, related to the riverine settlements, were calculated using Qgis 2.18.13 (QGIS Development Team, 2009): (1) distance from the centroid point of each nest-site to the nearest house (based on GPS fixes obtained *in situ* at the sampling period), (2) presence of houses within a 1 km buffer, and (3) number of houses within a 5 km buffer. Buffer zones were set based on *P. unifilis* movement patterns as females disperse between 3 and 7 km during the dry season (Bock, Páez, & Pérez, 1998; Haller, Guimarães, & Raimo, 2015), when they usually stay close to the nesting sites (Bock et al., 1998).

Data analysis

All analyses were performed in R (R Development Core Team, 2018). The correlation between the five response variables (i.e., nest number, nest density, median distance to water, standard deviation of distance to water, and proportion of nests removed by humans) was tested by a Spearman correlation matrix. Since no collinearity was found, the response variables were modelled separately to examine the relative importance of the anthropogenic

and environmental variables. Except for the proportion of nests removed by humans, which was modelled using a binomial error distribution family with log link; all other response variables were modelled using the Tweedie error distribution family with default log-link power function. Tweedie is a probability distribution family that includes the continuous normal and gamma distributions, the discrete Poisson distribution, and the class of compound Poisson-gamma distributions (Jorgensen, 1997). For each of the four responses, a maximum likelihood method (function "tweedie.profile") was used to profile the Tweedie variance function index parameter p. The Tweedie error distribution family was modelled using the "tweedie" R package (Dunn, 2017).

Generalized linear models (GLMs) were used to evaluate how the explanatory variables influenced nesting patterns in the study area. We controlled for collinearity among explanatory variables by performing a Spearman correlation matrix. Environmental variables were weakly correlated ($r_s < 0.41$), thus we retained all variables in further analyses. When examining anthropogenic variables, two variables (distance to the nearest house and number of houses within 5 km radius) were correlated ($r_s = -0.87$) for distances under 5 km, but not for distances ranging from 5.9 to 37.1 km, as is the case of 42 (57.5%) nesting areas (see Figure A3), therefore we retained both variables for further analysis. All numerical explanatory variables were standardized (centered and scaled by their standard deviation) to make their coefficients comparable (Quinn & Keough, 2002). To examine the relative importance of our eight explanatory variables, we adopted a three-stage approach described below.

First, anthropogenic and environmental variables were modelled separately (see Table A1). Area of the nesting site was maintained as the null hypothesis in both, environmental and anthropogenic models, since we verified that area had a strong influence when modelled alone.

Second, in order to identify the most strongly supported variables for both categories, we calculated the relative importance of each variable by multimodel inference (Burnham & Anderson, 2002), in which our GLMs were ranked and scaled according to the Akaike's Information Criterion (AIC) (implemented in "MuMIn" R package (Barton, 2018). Relative importance of each explanatory variable was measured as the sum of Akaike weights ($\sum$wi) of the models containing that variable (Burnham & Anderson, 2002: pp 75-77, 167-172). A reduced subset of models for a 95% confidence set, based on the sum of Akaike weights across all models from largest to smallest that resulted in the sum of 0.95, was used to calculate variables relative importance (Burnham & Anderson, 2002: pp 169).

Third, a global model for each response variable was obtained by selecting the two variables with the highest support from each category (anthropogenic and environmental) plus nesting area size (see Table A2). This provided a total of five variables that were used to model each response and test the working hypotheses proposed to explain turtle nesting patterns (Table 1). To identify the best predictors for nesting patterns, we adopted a model selection approach based on the AIC (Burnham & Anderson, 2002: pp 60-65), thus, from the global model a final most parsimonious model was obtained for each response variable.

We tested for spatial autocorrelation by plotting semi-variograms of GLMs residuals (Dale et al., 2002; Dormann et al., 2007). None of the unexplained variation (GLM model residuals) was related to the geographic distance among nesting areas (see Figure A4), thus all nesting sites were considered statistically independent. For model validation, Hosmer-Lemeshow goodness of fit test was carried out using the package "sjPlot" (Lüdecke, 2018). In addition, model assumptions were verified by plotting residuals versus fitted values, and the normal distribution of the residuals was checked.

Results

The area of all the surveyed nesting sites ranged from 5.41 m^2 to 2458.41 m^2 (Median (IQR) = 326.31(125.6-711.2) m) (see Figure A5), with the size of the areas equally distributed between site type (margin or island) (Wilcoxon rank sum test, W = 534, p-value = 0.28) and between both rivers (Araguari and Falsino) (Wilcoxon rank sum test, W = 638, p-value = 0.81).Between the potential nest-sites, the proportion of island and margin was similar (Table 2, see Figure A5), on the other hand, sites with presence of trees (89.04%), fine sand as main substrate (84.93%) and only one type of substrate (39.72%) were more abundant than sites without trees, coarse sand and several types of substrates (see Figure A5). Distance from nesting sites to houses ranged from 0.06 km to 37.16 km (Median (IQR) = 7.13 (1.71-14.3) km), however, in the Araguari river the remotest site was 14.32 km far from a riverine house (see Figure A5). In both rivers, a higher proportion of nest-sites had no presence of houses within 1 km (91.11% in Falsino and 71.43% in Araguari) and 5 km (66.67% in Falsino and 42.86% in Araguari) buffer (Figure A5).We found turtle nests in 58 (79.5%) of the 73 nest-sites surveyed along 118 km of river (Table 2). A total of 305 nests were found (Table 2) and the number of nests encountered in a single site ranged from 1 to 26 (Median (IQR) = 3.00 (1.00-6.00) nests). Nest density, calculated for the entire area, varied from 0.0014 nest/m^2 to 0.37 nest/m^2 (Median (IQR) = 0.011 (0.005-0.02) nest/m^2). The distance from the nest to water ranged from 1.35 m to 16.72 m (Median (IQR) = 4.56 (3.32-6.67) m.). In total, 29 (53.7%) nesting-sites presented nest removed by humans, and we recorded a total of 121 (47.27%) nests harvested (Table 2), harvested nests presented no eggs because collectors removed all of them. Proportion of removed nests for each nesting site varied from 0 (none) to 1 (all) nests removed (Median (IQR) = 0.26 (0-0.75)).

There was a discernible effect of the size of nesting sites explaining nest-site selection patterns. Area of nesting site alone explained 20.3% of variation of nest number, 17.1% for

nest density, 15.7% for median distance to water, and 41.5% for standard deviation of distance to water (Table 3). Area was a positive significant predictor of nest number, median and standard deviation distance of nest to water, but negative for nest density (Table 3). Despite its importance for almost all response variables, area was not a significant predictor for explaining nest removal, explaining just 1.7% of the deviance (Table 3). In our information analysis (Figure 2), nesting area was the variable with the highest support for almost all the response variables (i.e., nest number ($\sum$W=1), nest density ($\sum$W=1), median distance to water ($\sum$W=0.98 and 0.89) and standard deviation of the distance ($\sum$W=1)), except for the proportion of removed nests ($\sum$W=0.41 and 0.45).

Besides the importance of area, each response variable showed a different pattern of variables' weight (Figure 2). Environmental variables were clearly the most strongly supported explaining nest number ($\sum$W mean (range) = 0.54 (0.93-0.27)), compared to anthropogenic variables ($\sum$W mean (range) = 0.27 (0.31-0.22)). Environmental and anthropogenic variables were equally supported for explaining nest density ($\sum$W mean (range) = 0.55 (0.85-0.29) and $\sum$W mean (range) = 0.55 (0.84-0.31), respectively). Conversely, anthropogenic variables showed a higher informative strength for explaining median nest to water distance and standard deviation distance ($\sum$W mean (range) = 0.60 (0.93-0.29) and $\sum$W mean (range) = 0.34 (0.45-0.23), respectively), compared to environmental variables ($\sum$W mean (range) = 0.50 (0.79-0.26) and $\sum$W mean (range) = 0 .28 (0.36-0.22), respectively), even if for the latter were weakly informative. Environmental variables showed slightly higher informative strength for proportion of removed nests ($\sum$W mean (range) = 0.54 (0.83-0.23)) than anthropogenic variables ($\sum$W mean (range) = 0.49 (1-0.21)), even if river stretch was the variable with the highest support ($\sum$W=1) (Figure 2).

Overall, a relevant influence of environmental variables explaining nest-site selection pattern was seen, although anthropogenic factors affected distance from the nest to water.

Nests removal was mainly explained by anthropogenic factors, even if environmental ones also influenced (Table 4). However, most parsimonious models differed widely in the strongest predictors for each of the five response variables, and deviance explained by the models ranged from 25.5 to 44.7 (Table 4). For explaining number of nests, diversity of substrate was a positive significant predictor, followed by the presence of trees. Type of nesting site and presence of houses within 1km were important variables explaining nest density; in islands the density tended to be higher and to increase slightly with presence of houses. Distance to houses was a positive significant predictor for median distance from nest to water, moreover distance to water tend to be larger in the Araguari stretch and when trees were present. The distance standard deviation decreased with presence of houses within 1km, meaning that when closer to houses nests were clumped closer to water's edge. For explaining nest removal, river stretch (i.e., intensity of anthropogenic activities) was reinforced as an important variable. Proportion of nests removed was higher in the Araguari river (Median (IQR) = 0.75 (0.50-1.00)), compared to Falsino (Median (IQR) = 0.00 (0.00-0.50)). Nest harvest was also higher in nest-sites closer to houses and in islands.

Discussion

This study, across a large extension of rivers in the eastern Brazilian Amazon and between two adjacent sustainable use protected areas, showed that: (1) nest-site selection by *Podocnemis unifilis* is strongly affected by size of nesting area, (2) number of nests is driven by environmental cues and is not affected by anthropogenic pressure present in the area, while nest to water distance decreased with closeness to human settlements, and (3) nest removal is greater in areas with higher anthropogenic pressure, although environmental variables also played an important role (e.g., type of nesting site). Nesting patterns associated to environmental cues have already been documented for several freshwater turtle species (Janzen, 1994; Wilson, 1998; Janzen & Morjan, 2002; Hughes & Brooks, 2006; Zappalorti,

Lovich, Farrell, & Torocco, 2015), and few studies have documented nesting patterns alteration due to human impacts (Kolbe & Janzen, 2002; Horne et al., 2003; Moore & Seigel, 2006), but combination of both group of variables have been largely overlooked by studies conducted over large scales.

We first turn to discuss variables that influenced the nest-site selection in *P. unifilis* as number and density of turtle nests, and nest distance to water. Then explore how nest removal was influenced by anthropogenic and environmental variables. Finally, we contribute with some general information in order to direct and enhance conservation efforts for river turtles in the Brazilian Amazon.

Nest-site selection: Number, density of nests and distance from nest to water

The number of nests encountered in the monitored nesting sites was small (3 nest/site) in contrast with other *P. unifilis* distribution regions, where number of nest ranged from 24.5 to 43 nest/site (Escalona & Fa, 1998; Ferreira-Júnior & Castro, 2003; Escalona et al., 2009), even 136.5 nest/site reported by Pignati et al. (2013b). However, the density of nests found was much larger (0.011 nests/m^2) compared to previous studies (density ranged from $1.13e^{-4}$ to $3.3e^{-5}$) (Escalona & Fa, 1998; Ferreira-Júnior & Castro, 2003; Escalona et al., 2009; Pignati et al., 2013b). High densities were due to the smaller areas of the monitored nesting sites comparing to the other Amazonian regions, in which nesting sites are sand depositions of approximately 0.18 km^2 to 2.4 km^2 (Escalona & Fa, 1998; Ferreira-Júnior & Castro, 2003; Escalona et al., 2009; Pignati et al., 2013b). Those areas are usually shared with other congeners as the giant Amazon river turtle (*P. expansa*), which needs big sand banks for nesting (Ferreira & Castro, 2005), and is not presented in the Araguari river basin. Contrary, yellow-spotted river turtle nests can be found in a wide variety of sites, including small sand banks eroded by the river (Thorbjarnarson et al., 1993), ravines on the river banks and muddy shores (Soini, 1994). Overall in our study area, bigger areas presented more nests,

corroborating the results found by Escalona & Fa (1998). Nevertheless, while our study shows that smaller areas presented a higher density of nests, Escalona & Fa (1998) found no correlation between area and nest density. This highlights the importance of small areas for *P. unifilis* nesting in the Araguari river basin, which are more abundant (see Figure A5).

Our results showed that environmental features (i.e., substrate diversity and presence of trees) were the strongest predictors of number of turtle nests across surveyed nesting sites. Those variables are important for regulating temperature and moisture, which are important incubation factors(De Souza & Vogt, 1994; Pignati et al., 2013a). Differences in substrate granulometry affect microhabitat temperature due to differences in thermal conductivity (De Souza & Vogt, 1994), however *P. unifilis* eggs are resistant to temperature variation (Packard, Packard, & Boardman, 1982) due to the rigid eggshell (Pritchard & Trebbau, 1984), and no relation has been found between grain size of the sediment and incubation time in *P. unifilis* (Ferreira-Júnior & Castro, 2003). Type of sediment seems not to affect females' preferences, since *P. unifilis* nests have been found in diverse substrates, as mud, sand, leaf litter (Fachín-Terán & Von-Mülhen, 2003), and even in pastures (Dos Santos, 2013). The non preference for one type of substrate was corroborated in this study, since main substrate of nesting site was not an important variable explaining number of nests. Moreover, turtle females selected sites with greater substrate variety.

Vegetation presence in nesting site can provide a variety of microenvironments, from open to shaded areas. Nests located in areas with less plant cover experiment higher temperatures and consequently shorter incubation periods (Vogt & Bull, 1984; Pignati et al., 2013a). On the other hand, small turtles like *P. unifilis* cannot dig deep nests to avoid extreme temperatures (Wilson, 1998), therefore, shade provided by trees avoids extreme increases of incubation temperature (Pignati et al., 2013b). Vegetation can also be the main factor affecting sex ratio, since sex determination depends on the incubation temperature (Vogt &

Bull, 1984), in *P. unifilis* higher proportion of females are produced at higher temperatures (Souza & Vogt, 1994). Results found in this study suggest that the presence of trees is important for *P. unifilis* nesting, although in previous studies nests in open areas (Soini, 1994) as well as near vegetation (Pignati et al., 2013b) have been found. Detailed studies measuring distance to vegetation and vegetation height should assess specific conditions selected by females within the nesting area.

Similarly, turtle nest density was strongly influenced by environmental variables. Islands presented a higher nest density compared to margins, given that the availability of both type of nesting sites was similar (see Figure A5) as well as their size, *P. unifilis* seems to prefer islands to nest. Islands are geographically isolated from the forest and have lower species richness (MacArthur & Wilson, 2001), which in turn can reduce natural predation risks (George, 1987). The main natural predator of *P. unifilis* nests is the Tegu lizard (*Tupinambis sp*.) (Escalona & Fa, 1998; Fachín-Terán & Von-Mülhen, 2003), predation by Tegu lizard was reported in our study area, being higher in margins compare to islands (see Figure A6). This is particularly important for maximizing conservation actions for *P. unifilis* across the Amazon, as restoration of potential nest-sites after submersion due to river flow management and reservoir formation after the construction of new hydroelectric power plants (Norris et al., 2018a).

Contrary to our first hypothesis, anthropogenic variables presented in the area neither decreased number nor density of nests. Conway-Gómez, Reibel, & Mihiar (2014) reported that *P. unifilis* adults avoided areas close to highly populated settlements. However, the communities studied by Conway et al. (2014) had 101 and 70 households, compare to 13 and four houses present in the communities of Araguari and Falsino rivers respectively. De Oliveira et al. (2015) already reported for the Araguari river basin that the impact of human activity is not severe and does not stop wildlife from using the area. Thus, human activity in

our study area does not completely prevent turtles from nesting in anthropogenic disturbed areas.

Nest location within the site was mainly influenced by anthropogenic factors, confirming our second hypothesis. Yellow-spotted river turtles do not concentrate their nests in a specific distance from the shore (e.g. nest to water distance in our study ranged from 1.35 to 16.72 m), but in order to avoid nest flooding, which is the main natural cause of nest failure (Ferreira-Júnior & Castro, 2010; Pignati et al., 2013b), more nests are usually found far from water's edge (Escalona & Fa, 1998; Pignati et al., 2013b). Furthermore, nests that are located closer to the river produce smaller hatchlings, due to unsuitable conditions for incubation, as moist substrate (Ferreira-Júnior, Castro, & Castro, 2007). On the other side, farther from shore maternal mortality risk increases due to exposition to predators (Spencer, 2002). Humans are perceived as possible predators (Frid & Dill, 2002), thus, when females get disturbed by humans they reduce exposure (Moore & Seigel, 2006) and nest near water (Spencer, 2002). Therefore, nesting females face a trade off between maximizing offspring survival or their own survival (Spencer, 2002; Refsnider & Janzen, 2010).

We found that with the proximity to human habitations and in sites with presence of houses within 1km, females nested closer to the water and the variability of the distance decreased, meaning that nests were concentrated closer to water's edge; even if that decreases hatching success. Hence, results presented here suggest that when closer to anthropogenic pressure, *P. unifilis* females nest in potential non-optimal places for offspring survival, but where maternal survival might be higher. Given the limited ability of nesting females to assess nesting environments, sites are likely to be chosen on the basis of maternal survival rather than offspring (Bjorndal & Bolten, 1992).

Nest-site suitability: Proportion of removed nests

Information gathered in this study demonstrated that high rate of nest removal by humans is still a major threat for *P. unifilis* nests in sustainable use protected areas. We found almost half (47.27%) of total nest removed. Nest predation by humans is ubiquitous and the primary driver of *P. unifilis* nest failure (Foote, 1978; Escalona & Fa, 1998; Pignati et al., 2013b). High rates of nest removal have been also recorded across the species range: 70.44% and 50.4% in Southern Venezuela (Hernández, Espinosa-Blanco, Lugo, Jiménez-Oraa, & Seijas, 2010), 51.9% in Peru (Landeo, 1997), and 45.51% in the lower Amazon river in Brazil (Pignati et al. 2013b). Confirming the third hypothesis, nesting success was depressed in places where human activities are higher (e.g., in the Araguari river and closer to riverine houses), due to human removal of eggs.

River stretch was the variable that most explained nest removal. Araguari river suffered a higher proportion of removed nests compared to Falsino river. The two rivers have a relatively low number of sparse riverine houses, 13 in Araguari and four in Falsino river, a total of 17 families living within the study area. Anthropogenic pressure in our study area is lower compared to other areas in the Amazon, communities of 70 and 97 families are found in the Paraguá and Itenéz/Guaporé rivers in the Bolivian Amazon (Conway-Gómez, 2007), in the lower Amazon the Água Preta community has 60 families and is 32 km from Santarém city (294580 inhabitants (IBGE, 2010)) (Pignati et al. 2013b), in the Xingú river two communities of more than 30 families are found riverside close to Altamira city (99075 inhabitants (IBGE, 2010)) (Alcântara, Silva, & Pezzuti, 2013). However, even being a low impacted area, the rate of removal in Araguari river is severe, 86% of the nesting sites presented nests predated by humans and 71.43% of the encountered nest were removed, compare to 26.28% in Falsino river. Araguari river suffers the exploitation by the fishing fleet from Porto Grande town (16809 inhabitants (IBGE, 2010)) (ICMBIO, 2014), thus, extraction

of resources increases by people from outside the riverine community. In addition, campsites and fires encountered during our monitoring period suggested that fishers do trips of several days when they go fishing, and camp in nesting sites, which increases the probability of nest removal.

Harvest rates were not spatially uniform, being higher closer to human settlements. Activities around human settlements are higher because riverine communities fish closer to houses (Escalona & Fa, 1998; Fachín-Terán & Von-Mülhen, 2003; Conway-Gómez, 2007). We observed a threshold of 16 km (see Figure A7) from which human removal of nests decreased significantly. The most likely explanation for the encountered magnitude of impact is that outsiders may be the main contributors for high removal rates. Because even if Norris & Michalski (2013) already reported that more than 50% of locals from the riverine community in Araguari river basin eat turtle eggs, they typically remain close (around 500m) to their houses when fishing. The correlation between proximity to human settlements and the increase in harvest conforms to previous studies, that indicated the depletion of game species because of hunting pressure in the Amazon basin. Peres & Lake (2003) observed a threshold of 9 km, due to reduced access by foot from point access (i.e., road or river), beyond which hunting did not negatively affect wildlife. Conway-Gómez (2007) reported a 10 km threshold for fishers from the city when doing one day trip, in which turtle abundance was being depressed because o hunting.

Nest removal was also affected by environmental variables: a higher removal rate was registered in islands compared to margins. The higher nest density registered in islands (see Table 4) could have lead to an increase in nest harvest, because clumped nests suffer a higher predation rate than scattered nests (Marchand, Litvaitis, Maier, & DeGraaf, 2002; Norris et al., 2018a). In addition, since boat is the only mean of transportation to the nesting sites, margins surrounded by forest and with a small size, as in the case in our study area, are

harder to spot. In contrast, islands are easier to identify and to be access by humans, thus, we expect a higher rate of human visitation to islands.

Implications for conservation

It is expected animals to select nesting habitats that confer fitness benefits (Howerter, Rotella, Anderson, Armstrong, & Devries, 2008). Even if females did not show a clear pattern of nest-selection in our study area and environmental cueing used by the species is described as weak (Escalona et al., 2009; Pantoja-Lima et al., 2009), *P. unifilis* has a placement of eggs differing from a random nesting pattern (Escalona et al., 2009). Nevertheless, in rivers with higher human pressure, signals used by females to choose nesting areas are possibly not longer successful in enhancing individuals' fitness, since reproductive success is being depressed as a result of nest harvest. Due to anthropogenic alterations, nest-site selection is becoming decoupled from suitability, and apparently, nest-site preferences are no longer reliable cues of safe sites because of altered predation risk (Schlaepfer, Runge, & Sherman, 2002; Howerter et al., 2008). *P. unifilis* females do not appear to avoid nesting in dangerous and unfit sites, which makes the species more vulnerable to nest removal.

Turtles are known to have mortality rates inversely related to age (Iverson, 1991), due to natural predation, nests flooding (Hernández et al., 2010) and embryo unfeasibility (Soini, 1994). Hatchling success can fluctuate from year to year depending on natural conditions, for *P. unifilis* nest success can vary from 38% to 92.2% (Soini, 1994; Hernádez et al., 2010; (Ferreira-Júnior & Castro, 2010) mainly due to differences in nest flooding (Soini, 1994; Ferreira-Júnior & Castro, 2010). Nevertheless, considering the high human impacts, especially eggs harvesting, hatchling survival can decrease significantly. Our study suggests that human consumption of freshwater turtle eggs remains a challenge for conservation of the species, since indirect impacts, as nest predation, waterways development and pollution, are likely to generate deleterious effects of *P. unifilis* populations (Norris & Michalski, 2013).

Consumption of eggs has already been reported to cause drastic freshwater turtle population reductions across Amazonia (Vogt, 2008). In addition, Conway-Gómez (2007) and Alcântara et al. (2013) reported 87.18% and 65.77% of decrease in *P. unifilis* adult abundance, due to increasing human population and consequent hunting pressure. Therefore, we anticipate that without direct conservation actions, a reduction in the yellow-spotted river turtle population is likely to occur in areas affected by anthropogenic pressure.

The establishment of protected areas have been the priority for Amazonian biodiversity conservation, however in some of the established protected areas management effectiveness is basic or deficient (Leverington, Hockings, & Costa, 2008; Norris et al., 2018c). Given that protected area alone is insufficient to guarantee the conservation of turtle species, direct action is needed to preserve Amazonian freshwater turtles. Although in sea turtles conservation of eggs might not be enough for ensuring population growth, due to high fluctuating rates of eggs and juvenile mortality (Crouse, Crowder, & Caswell, 1987), headstarting could be an effective tool for managing freshwater turtles, thanks to the short time to reproductive maturity (Spencer, Van Dyke, & Thompson, 2017). Translocation of eggs has been one of the most common conservation strategies used in chelonians; however, it has low success and natural incubation conditions could be altered (Fischer & Lindenmayer, 2000; Pantoja-Lima et al., 2009). Therefore, *in situ* protection of nests by nesting area protection might be a key process for ensuring conservation of the species. Norris et al. (2018c) showed that community-based management is perhaps the only way to ensure long-term conservation of overexploited wildlife species in low-governance regions as the Amazon. In the same study they demonstrate that increasing first-year survival by the 50% *P. unifilis* adult population could double within a decade. Local community-based management has already been successful decreasing nest harvesting rate (Caputo, Canestrelli, & Boitani, 2005; Norris, Michalski, & Gibbs, 2018b).

For an effective management, conservation efforts should be enhanced in more vulnerable areas identified in this study, as sites with high density of nests, nests that are not under the risk of flooding, and in more vulnerable sites (i.e., the ones closer to human settlements). This strategy could be effective for local community-based management, given that sites are more accessible, besides, tools and the support needed should be offered in order to reach distant affected sites. Riverine communities do not depend anymore on river turtles for their daily nutritional requirements or economic well-being (Norris & Michalski, 2013; De Jesus Silva, Garavello, Nardoto, Mazzi, & Martinelli, 2017) and because nest harvesting by humans is not specifically targeted, not conflict should be generated (Norris et al., 2018b). Nevertheless, approaches as providing payments for protecting nests and/or the selective harvest of nests that would otherwise be flooded have been used to engage local communities (Caputo et al., 2005). For the success of conservation activities within sustainable-use protected areas, collaborative development between local communities, researchers and conservationists are needed.

Acknowledgements

invaluable assistance during fieldwork.

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Table 1. Working hypotheses and variables used to explain nesting patterns in *Podocnemis unifilis* in the eastern Brazilian Amazon. For variable support: (++) $p < 0.05$ in the final most parsimonious model; (+) $p > 0.05$ but retained in the final most parsimonious model; (-) not retained in the most parsimonious final model.

Category	Working hypothesis	Variable name	Variable description	Variable support				
				Nest number	Nest density	Dist. to water (median)	Dist. to water (SD)	Removal
Null	Area will highly influence number of nest and nest distance to water.	Area	Continuous - Size (m^2) of nesting area.	++	++	++	++	-
Anthropogenic	Nesting areas with presence of houses will present higher anthropogenic pressure, affecting nest site selection and nesting suitability.	House 1km	Categorical – Presence of house (yes/ no) within one km of nesting area	-	+	-	+	-
Anthropogenic	Nesting areas with higher amount of houses have higher human disturbances that will affect nest site selection and nesting suitability.	Houses 5km	Continuous – Number of houses within a 5km radius of nesting area.	-	-	-	-	-
Anthropogenic	Since Araguari river suffers higher anthropogenic pressure, different rivers will present differences in nest site selection and nest removal.	River	Categorical – River stretch (Falsino or Araguari)	-	-	++	-	++
Anthropogenic	Closer to houses human disturbances will increase, affecting nest site selection and nest removal.	Dist. house	Continuous – Distance (km) to nearest riverine house	-	-	++	-	++
Environmental	Type of nesting site could influence the access of predators affecting nest site selection.	Type	Categorical – Located along the river bank or island.	-	++	-	-	++
Environmental	Shade created by trees can alter incubation characteristics affecting nest site selection.	Tree	Categorical –Presence of tree (yes/no).	+	-	++	-	-
Environmental	Different types of substrate can experiment different incubation conditions, affecting nest site selection.	Substrate diversity	Continuous – Ranked scale of substrate diversity (from 1 to 4).	++	-	-	-	-
Environmental	Different types of substrate can experiment different incubation conditions, affecting nest site selection.	Main substrate	Categorical – Predominant substrate cover (fine or coarse sand).	-	-	-	-	-

Table 2. River length (km), number of nesting areas, and number of *Podocnemis unifilis* nests recorded along the Araguari and Falsino rivers, in the eastern Brazilian Amazon. * Protected nest present in the four protected nesting sites.

	Araguari	Falsino	Total
River length (km)	46	72	118
Nesting sites count	28	45	73
Nest present	22	36	58
No nest present	6	9	15
Margin	11	22	40
Island	17	23	33
Sites with removed nests	18	11	29
Total number of nest	122	183	305
Non protected nests *	119	137	256
Number of removed nests	85	36	121

Table 3. Area effects on nesting patterns. Relationships between size of nesting sites (m^2) and *Podocnemis unifilis* nesting in the eastern Brazilian Amazon. Acronyms: B: Model slope estimate, CI: confidence interval of estimate, p: significance levels (<0.05 are showed in bold).

	Nest number			Nest density			Dist. to water (median)			Dist. to water (SD)			Removal		
	B	CI	p	B	CI	p	B	CI	p	B	CI	p	B	CI	p
(Intercept)	1.32	1.08–1.54	<.001	-4.18	-4.54– -3.76	<.001	1.65	1.52–1.80	<.001	0.09	-0.10–0.32	.389	0.68	0.46–0.90	<.001
Area	0.43	0.25–0.61	<.001	-0.72	-1.11– -0.29	.007	0.24	0.09–0.46	.006	0.65	0.38–1.02	<.001	-0.09	-0.25–0.08	.312
Observations	73			73			58			38			54		
AIC	363.5			-246.1			266.6			91.6			166.7		
Deviance %	20.3			17.1			15.7			41.5			1.7		
Hosmer-Lemeshow-X^2	-16.60;p=1.00			0.60;p=1.00			-4.28;p=1.00			12.59;p=.71			0.98;p=1.00		

Table 4. Final most parsimonious models. Relationship between environmental and anthropogenic explanatory variables and *Podocnemis unifilis* nesting in the eastern Brazilian Amazon. In categorical variables reference level as, Houses 1km: no, River: Araguari, Tree: no, Type: island. Acronyms: B: Model slope estimate, CI: confident interval of estimate, p: significance levels (<0.05 are showed in bold).

	Nest number			Nest density			Dist. to water (median)			Dist. to water (SD)			Removal		
	B	*CI*	*p*	*B*	*CI*	*p*	*B*	*CI*	*p*	*B*	*CI*	*p*	*B*	*CI*	*p*
(Intercept)	0.01	-0.95–0.91	.982	-4.07	-4.47–-3.65	<.001	1.38	1.02–1.81	<.001	0.15	-0.04–0.39	.176	1.34	0.93–1.77	<.001
Area	**0.31**	**0.11–0.50**	**.003**	**-0.67**	**-0.96–-0.35**	**<.001**	**0.21**	**0.07–0.40**	**.011**	**0.64**	**0.38–1.00**	**<.001**			
Houses 1km (yes vs no)				0.72	-0.01–1.48	.061				-0.42	-0.83–0.09	.072			
Dist. house							**0.20**	**0.08–0.33**	**.007**				**-0.22**	**-0.45–-0.00**	**.048**
River (Falsino vs Araguari)							**-0.31**	**-0.59–-0.05**	**.027**				**-0.60**	**-1.14–-0.06**	**.029**
Tree (yes vs no)	0.66	-0.15–1.54	.132				**0.46**	**0.07–0.76**	**.009**						
Substrate diversity	**0.35**	**0.07–0.63**	**.018**												
Type (bank vs island)				**-0.71**	**-1.30–-0.10**	**.024**							**-0.50**	**-0.95–-0.05**	**.030**
Observations	73			73			58			38			54		
AIC	357.2			-252.2			257.6			91.5			146.8		
Deviance %	29.0			25.5			35.2			44.7			41.3		
Hosmer-Lemeshow-X^2	-13.88;p=1.00			.32;p=1.00			-1.73;p=1.00			5.46;p=.71			1.27;p=1.00		

Figure Legends

Figure 1. Study area. (A) State of Amapá in Brazil. (B) Location of the FLONA and FLOTA within Amapá. (C) Yellow circles showing the location of the sampled *Podocnemis unifilis* nesting sites in the Araguari and Falsino rivers. The nearest town (Porto Grande) is shown by a solid red triangle.

Figure 2. Ranking of support of explanatory variables grouped into two categories (Anthropogenic and Environmental). The support of each variable within each model category was ranked (ranks in parenthesis) according to the sum of the Akaike weight ($\sum$Wi) for their contribution to explain variation in five response variables (nest number, nest density, distance to water (median), distance to water (SD), and removal).
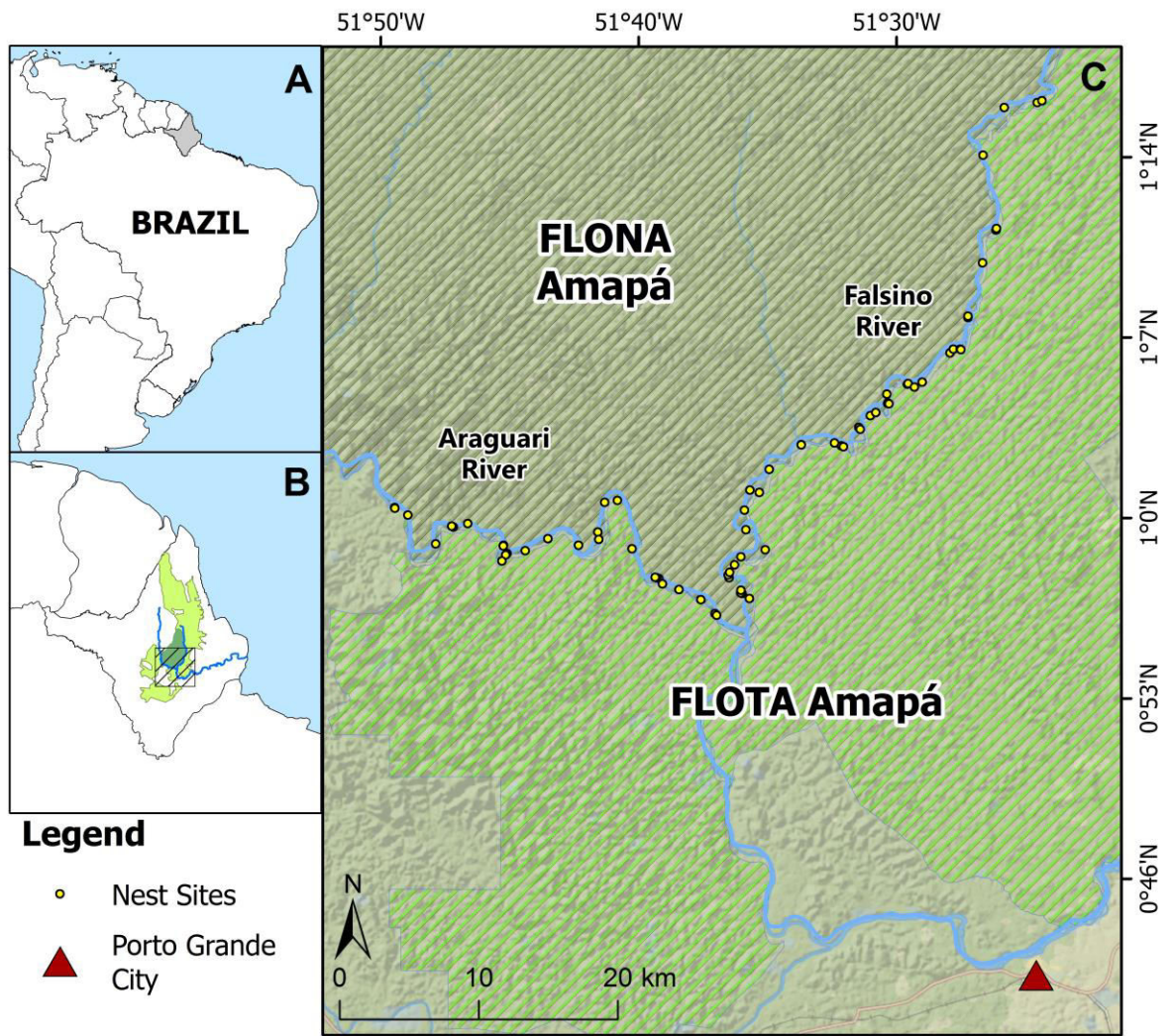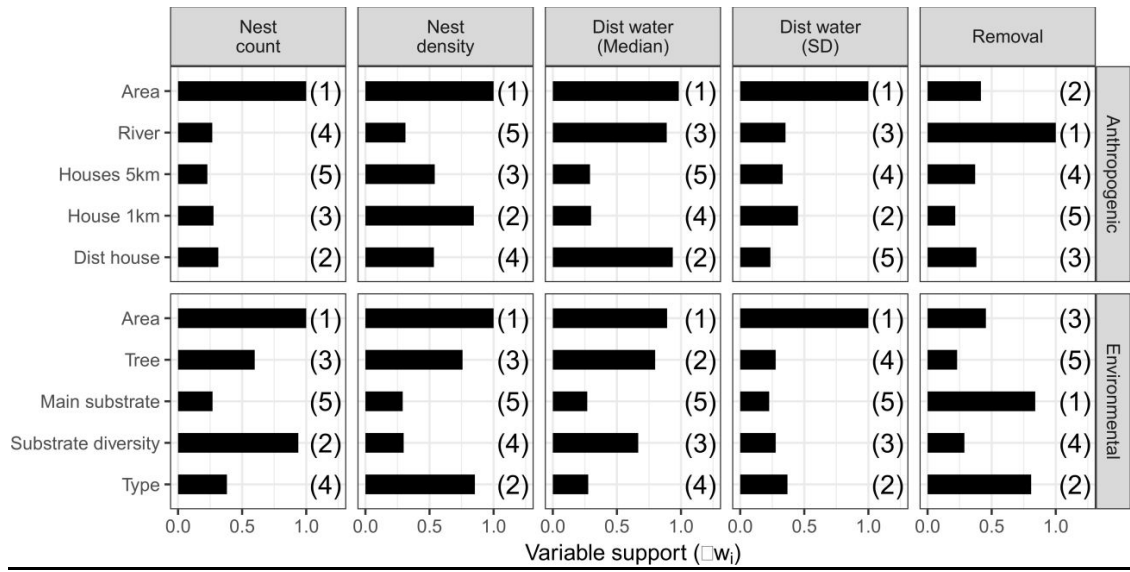
Figure 1

Figure 2

Appendices

Figure A1. Photos characterizing *Podocemis unifilis* nesting areas in the eastern Brazilian Amazon. Representative examples showing, (A-D): Nesting areas with suitable habitat conditions for nesting; (E-F): Nesting areas with presence of trees; (G-H): Nesting areas without trees; (I-J): Nesting areas with coarse sand as dominant substrate. (K-L): Nesting areas with fine sand as dominant substrate. Photo credit Itxaso Quintana.

Figure A2. Nesting peak and human removal peak in *Podocnemis unifilis* in the eastern Brazilian Amazon. Cumulative proportion of new nests found and nests found removed by humans through the multiple visit during the study period. Both graphs show that the nesting and human removal peak was during the first fortnight of November in 2017.
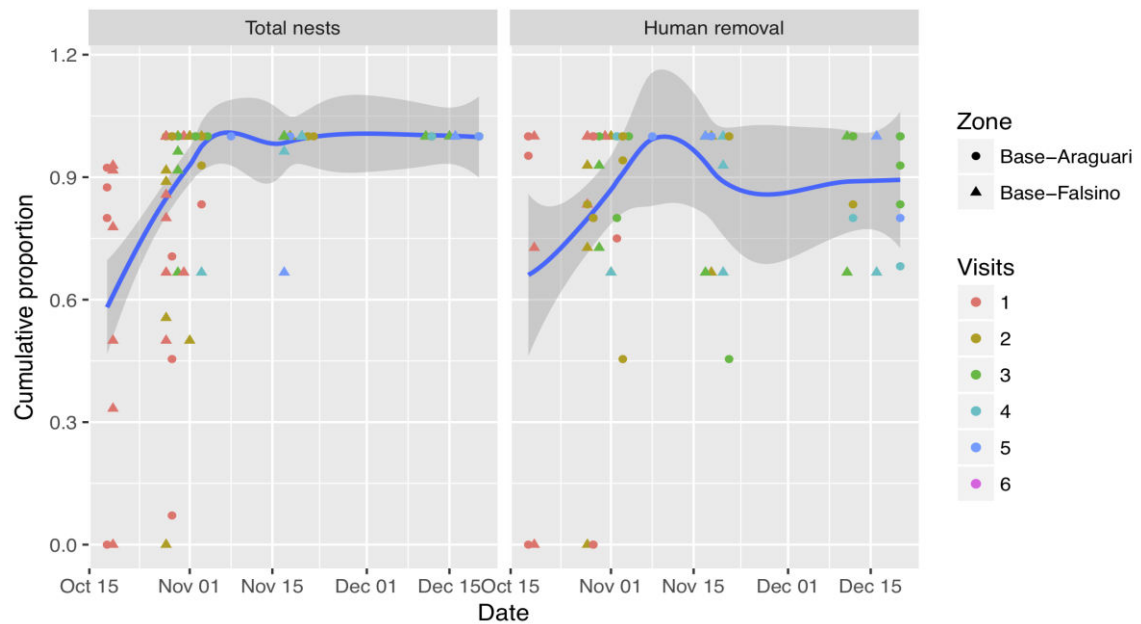
Figure A3. Relationship between the number of houses within 5km and distance to nearest house. Loess (local polynomial regression fit) trend line added to aid the visual interpretation of the non-linear relationship.
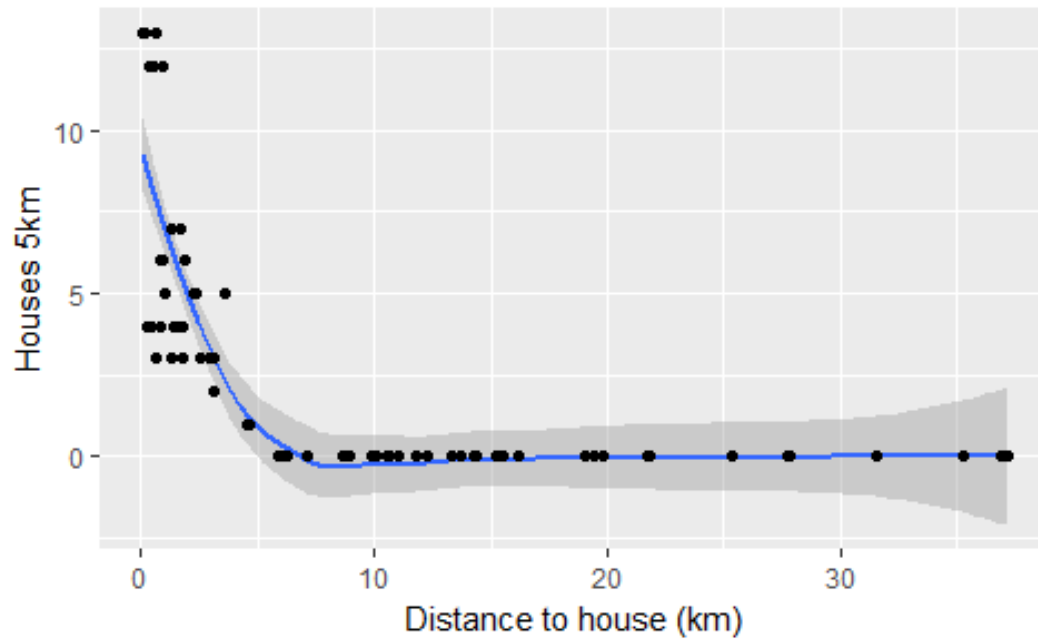
Figure A4. Sample semi-variograms and simulation envelopes under random permutation of GLM residuals of A) Anthropogenic models and B) Environmental models. Distance calculated from geographic coordinates (decimal degrees, $0.1 \approx$ 9000m).

Uncorrelated residuals should give a more or less flat semi-variogram, while unmodelled spatial auto-correlation (spatial dependence) usually results in a semi-variogram which increases sharply before eventually plateauing. In the current case, the semi-variograms for all models suggest no autocorrelation.

A) Anthropogenic models

Figure A4. Continued.

B) Environmental models

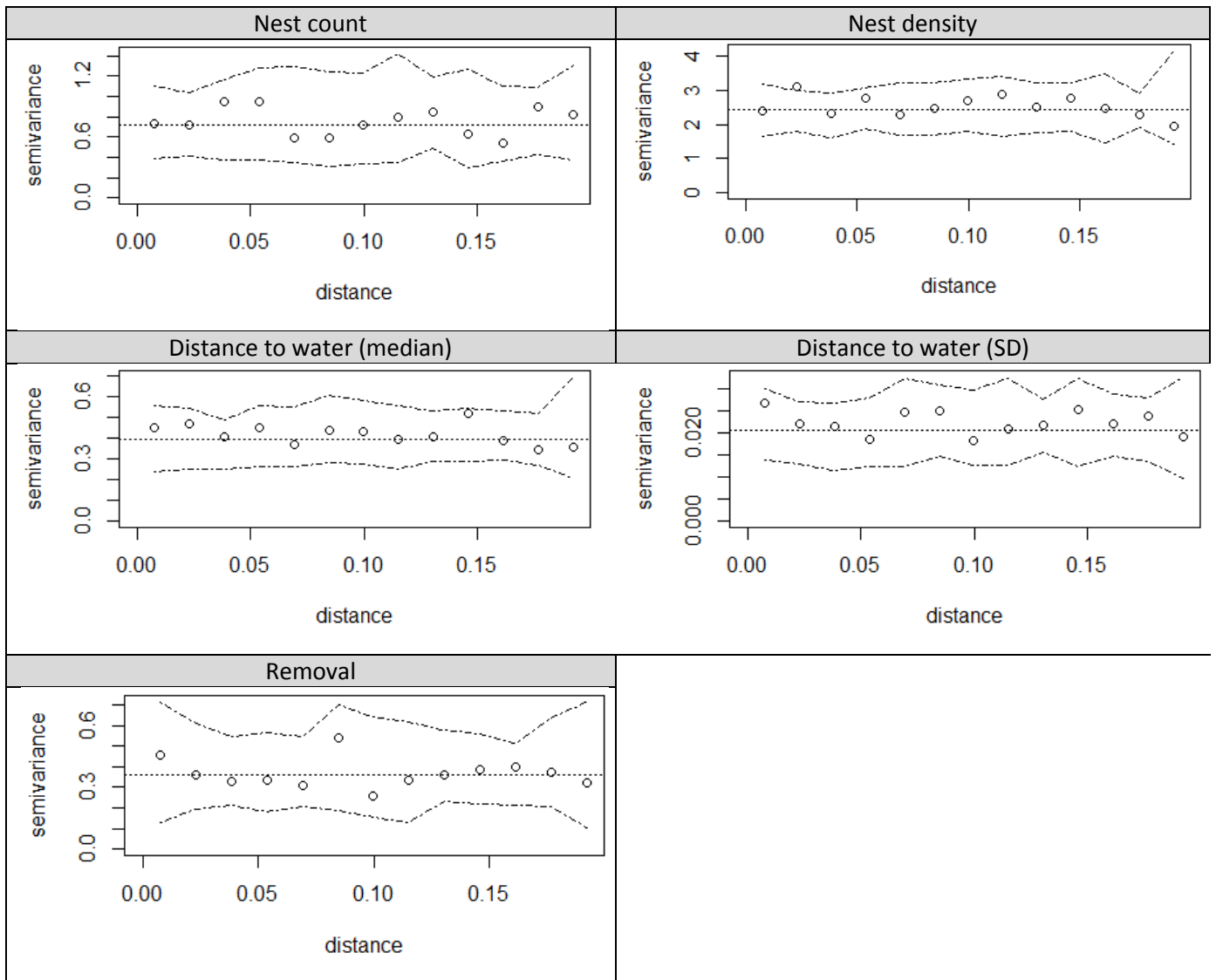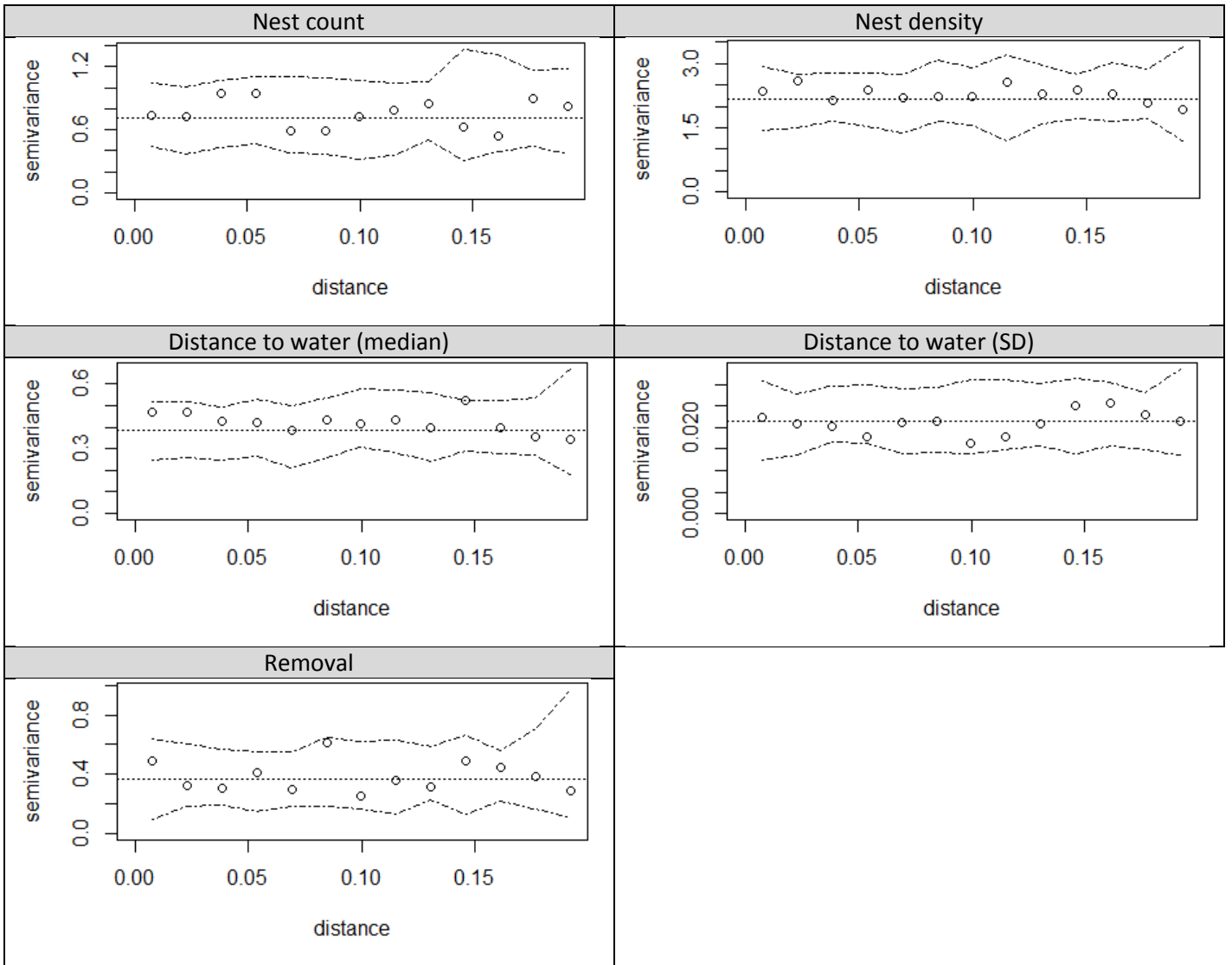**Table A1**. Results from generalized linear models (GLMs) performed separately for each group of variables (i.e., anthropogenic and environmental).GLMs of five responses evaluated against additive effects of anthropogenic and environmental variables. In categorical variables reference level as, Houses 1km: no, River: Araguari, Tree: no, Type: island, Main substrate: fine. Acronyms: B: Model slope estimate, CI: confidence interval of estimate, p: significant level.

Anthropogenic	Nest number			Nest density			Dist. to water (median)			Dist. to water (SD)			Removal		
	B	*CI*	*p*	*B*	*CI*	*p*	*B*	*CI*	*p*	*B*	*CI*	*p*	*B*	*CI*	*p*
(Intercept)	1.40	1.00–1.79	<.001	-4.54	-5.04–-4.03	<.001	1.85	1.63–2.13	<.001	0.32	-0.04–0.78	.092	1.28	0.85–1.73	<.001
Area	0.45	0.26–0.64	<.001	-0.67	-0.97–-0.35	.001	0.21	0.06–0.41	.014	0.70	0.35–1.22	<.001	-0.10	-0.30–0.10	.328
Houses 1km (yes vs no)	-0.24	-1.13–0.63	.599	1.09	0.11–2.09	.052	-0.19	-0.64–0.31	.355	-0.30	-1.01–0.45	.405	-0.08	-1.00–0.87	.869
Houses 5km	-0.02	-0.41–0.36	.919	-0.45	-0.95–0.06	.071	0.07	-0.13–0.29	.466	-0.09	-0.41–0.24	.603	0.15	-0.29–0.62	.516
River (Falsino vs Araguari)	-0.07	-0.57–0.43	.772	0.17	-0.51–0.85	.612	-0.33	-0.66–-0.05	.028	-0.31	-0.84–0.22	.214	-0.82	-1.38–-0.28	.003
Dist. house	-0.12	-0.42–0.17	.443	-0.31	-0.67–0.06	.136	0.20	0.05–0.39	.025	0.03	-0.20–0.32	.826	-0.09	-0.36–0.18	.513
Observations	73			73			58			38			54		
AIC	370.2			-247.8			263.7			96.3			153.7		
Deviance %	21.4			25.2			30.4			46.4			36.6		
Hosmer-Lemeshow-X^2	-19.89; *p*=1.00			.34; *p*=1.00			-2.01; *p*=1.00			-2.65; *p*=1.00			.97; *p*=1.00		

Table A1. Continued.

Environmental	Nest number			Nest density			Dist. to water (median)			Dist. to water (SD)			Removal		
	B	CI	p	B	CI	p	B	CI	p	B	CI	p	B	CI	p
(Intercept)	0.13	-0.86–1.06	.795	-3.50	-4.44–-2.53	<.001	0.77	0.30–1.36	<.001	-0.49	-1.24–0.98	.280	0.30	-0.67–1.30	.553
Area	0.30	0.10–0.50	.003	-0.73	-1.05–-0.39	<.001	0.20	0.01–0.48	.037	0.70	0.38–1.11	<.001	-0.18	-0.38–0.02	.080
Type (bank vs island)	-0.21	-0.64–0.22	.346	-0.70	-1.29–-0.12	.019	0.08	-0.19–0.36	.554	0.25	-0.22–0.75	.272	-0.51	-0.95–-0.07	.024
Tree (yes vs no)	0.64	-0.17–1.53	.144	-0.87	-1.73–-0.03	.043	0.56	0.09–0.90	.003	0.38	-1.02–1.07	.376	0.22	-0.62–1.03	.599
Substrate diversity	0.34	0.06–0.62	.022	0.19	-0.20–0.59	.333	0.16	-0.01–0.36	.075	0.06	-0.17–0.30	.665	0.17	-0.12–0.46	.248
Main substrate (coarse vs fine)	0.02	-0.56–0.57	.946	-0.31	-1.10–0.51	.450	0.04	-0.31–0.54	.820	0.02	-0.59–0.78	.959	0.74	0.07–1.48	.038
Observations	73			73			58			38			54		
AIC	360.1			-249.1			267.0			97.7			159.7		
Deviance %	29.8			26.2			26.2			44.4			26.6		
Hosmer-Lemeshow-X^2	-7.44; p=1.00			.32; p=1.00			-2.09; p=1.00			-5.16; p=1.00			1.13; p=1.00		

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Table A2. Global model for the five response variables, with the four variables of each group (anthropogenic and environmental) that contributed most to explain the variation in nesting pattern in *Podocnemis unifili*s in the eastern Brazilian Amazon, plus the area. In categorical variables reference level as, Houses 1km: no, River: Araguari, Tree: no, Type: island, Main substrate: fine. Acronyms: B: Model slope estimate, CI: confidence interval of estimate, p: significant level (<0.05 are showed in bold).

	Nest number			Nest density			Dist. to water (median)			Dist. to water (SD)			Removal		
	B	CI	p	B	CI	p	B	CI	p	B	CI	p	B	CI	p
(Intercept)	0.06	-0.90–0.97	.893	-3.76	-4.82–-2.69	<.001	1.31	0.65–2.00	<.001	0.18	-0.40–0.84	.568	1.25	0.76–1.77	<.001
Area	**0.32**	**0.13–0.52**	**.002**	**-0.68**	**-0.97–-0.37**	**<.001**	**0.20**	**0.05–0.40**	**.025**	**0.77**	**0.43–1.20**	**<.001**	-0.10	-0.29–0.09	.314
Houses 1km (yes vs no)	-0.14	-0.79–0.49	.669	0.78	-0.32–1.91	.172				-0.45	-0.89–0.09	.072			
Houses 5km				-0.16	-0.61–0.28	.467									
Dist. house	-0.13	-0.38–0.11	.313				**0.19**	**0.06–0.34**	**.010**				-0.20	-0.44–0.04	.101
River (Falsino vs Araguari)							**-0.29**	**-0.62–0.02**	**.043**	-0.27	-0.74–0.19	.190	-0.56	-1.16–0.01	.059
Tree (yes vs no)	0.59	-0.21–1.47	.169	-0.41	-1.42–0.62	.362	**0.47**	**0.07–0.78**	**.008**						
Type (bank vs island)				**-0.62**	**-1.21–-0.03**	**.043**				0.22	-0.16–0.64	.269	-0.43	-0.89–0.03	.066
Substrate diversity	**0.37**	**0.08–0.65**	**.014**				0.03	-0.16–0.23	.752	0.03	-0.18–0.25	.824			
Main substrate (coarse vs fine)													0.45	-0.27–1.22	.234
Observations	73			73			58			38			54		
AIC	360.0			-249.9			259.5			95.4			148.3		
Deviance %	29.9			26.8			35.3			47.6			45.5		
Hosmer-Lemeshow-X^2	-3.54;p=1.00			.24;p=1.00			-1.94;p=1.00			32.64;p=.00			1.19;p=1.00		

Figure A5. Measures of the relative availability of characteristics of potential sites for *Podocnemis unifilis* nesting in the study site. Recorded environmental and anthropogenic characteristics are showed. Anthropogenic characteristics are showed for Araguari and Falsino rivers.
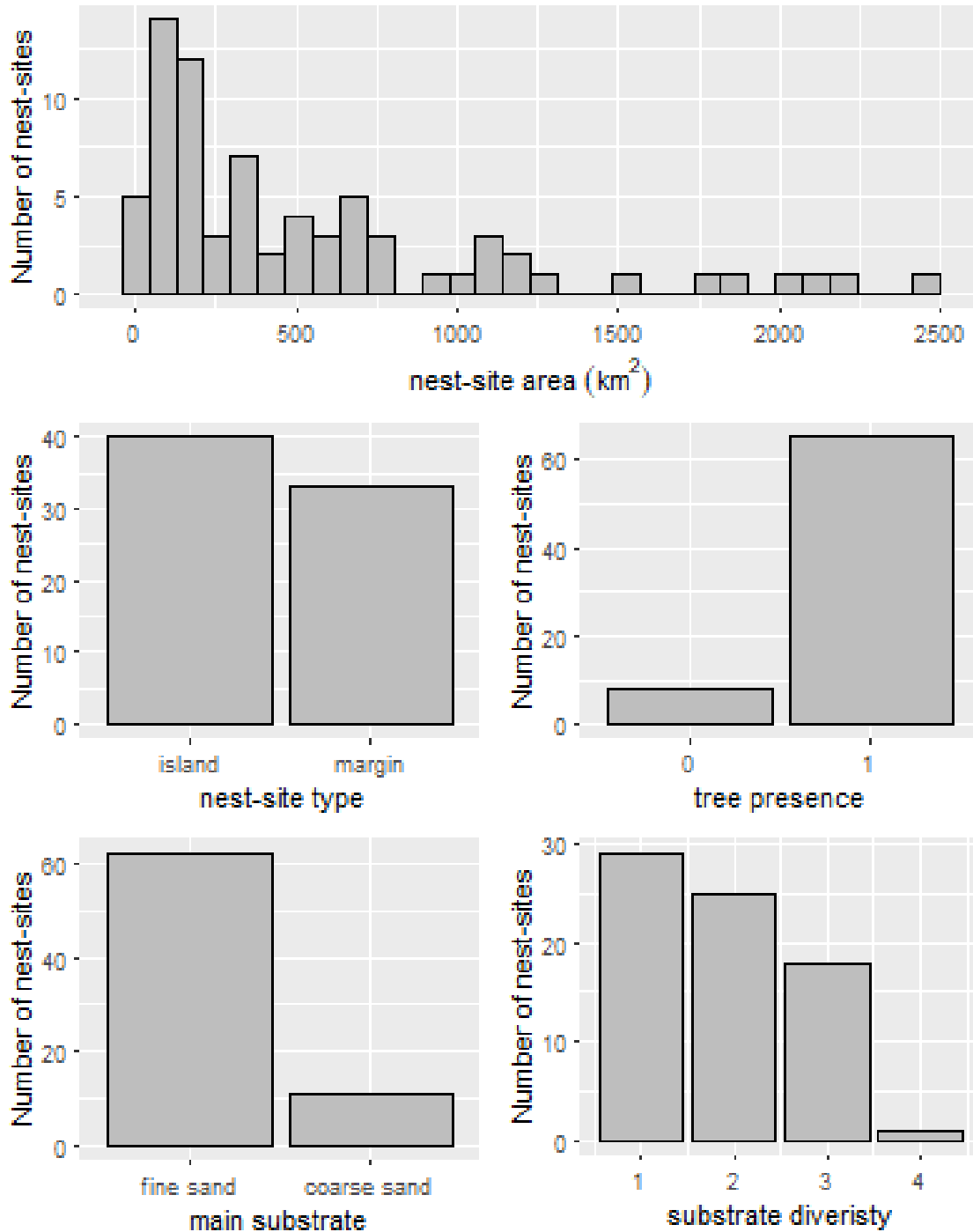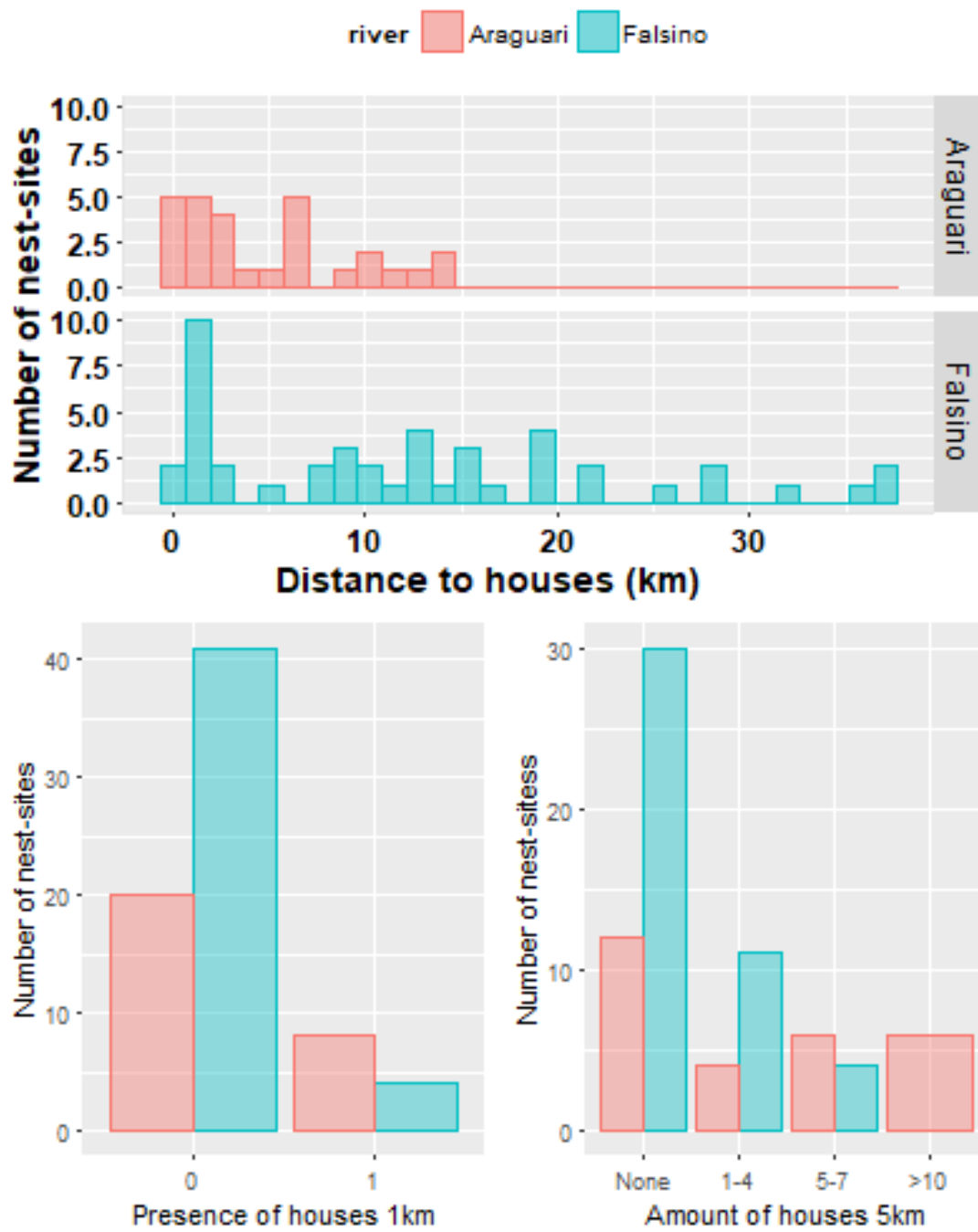
Figure A6. Proportion of naturally predated *Podocnemis unifilis* nests/site in the eastern Brazilian Amazon are showed, for different type of nesting sites (i.e., island and margin). Median and mean of the proportion of naturally predated nests/site is represented by a thick black line and a thick black point, respectively.

Figure A7. Relationship between the proportion of nests removed and the distance from the nesting site to the nearest riverine house. Loess (local polynomial regression fit) trend line added to aid the visual interpretation. Notice that 16 km in the threshold for nests to be removed around human settlements.