



PRISCILA DA SILVA LUCAS

**HISTÓRIA DE VIDA, DEMOGRAFIA E
CONSERVAÇÃO DE QUELÔNIOS: DA
PERSPECTIVA LOCAL À GLOBAL**

LAVRAS-MG

2016

PRISCILA DA SILVA LUCAS

**HISTÓRIA DE VIDA, DEMOGRAFIA E CONSERVAÇÃO DE
QUELÔNIOS: DA PERSPECTIVA LOCAL À GLOBAL**

Tese apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Ecologia Aplicada, área de concentração em Ecologia e Conservação de Recursos Naturais em Paisagens Fragmentadas e Agroecossistemas, para a obtenção do título de Doutor.

Prof. Dr. Alex Bager

Orientador

LAVRAS-MG

2016

**Ficha catalográfica elaborada pelo Sistema de Geração de Ficha Catalográfica da Biblioteca
Universitária da UFLA, com dados informados pelo(a) próprio(a) autor(a).**

Lucas, Priscila da Silva.

História de vida, demografia e conservação de quelônios : da perspectiva local à global / Priscila da Silva Lucas. – Lavras : UFLA, 2016.

129 p. : il.

Tese(doutorado)–Universidade Federal de Lavras, 2016.

Orientador(a): Alex Bager.

Bibliografia.

1. Quelônios. 2. Viabilidade populacional. 3. Tráfico de animais silvestres. 4. Estradas. 5. Avaliação global. I. Universidade Federal de Lavras. II. Título.

PRISCILA DA SILVA LUCAS

**HISTÓRIA DE VIDA, DEMOGRAFIA E CONSERVAÇÃO DE
QUELÔNIOS: DA PERSPECTIVA LOCAL À GLOBAL**

**LIFE HISTORY, DEMOGRAPHY AND TURTLE CONSERVATION:
FROM LOCAL TO GLOBAL PERSPECTIVE**

Tese apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Ecologia Aplicada, área de concentração em Ecologia e Conservação de Recursos Naturais em Paisagens Fragmentadas e Agroecossistemas, para a obtenção do título de Doutor.

APROVADA em 12 de setembro de 2016.

Dra. Clara Bentes Grilo	UFLA
Dr. Luiz Fernando da Silva Magnago	UFLA
Dr. Marco Aurélio Leite Fontes	UFLA
Dr. Vinícius Xavier da Silva	UNIFAL

Prof. Dr. Alex Bager

Orientador

LAVRAS-MG

2016

AGRADECIMENTOS

À Universidade Federal de Lavras, ao Centro Brasileiro de Estudos em Ecologia de Estradas, ao programa de Pós-graduação em Ecologia Aplicada, ao Setor de Ecologia e Conservação, Departamento de Biologia pelo acolhimento, oportunidades, desafios, estrutura e ensino disponibilizados para a minha formação acadêmica e profissional.

À Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) pela bolsa de estudos concedida durante estes anos e também durante o Programa de Doutorado Sanduíche no Exterior que foi realizado na Alemanha.

À Universität Leipzig e o German Centre for Integrative Biodiversity Research–iDiv pela infraestrutura e disponibilidade de seus funcionários em ajudar prontamente com a burocracia e por terem me recebido tão bem.

Ao meu orientador, Alex Bager, pelos bons momentos de convivência, boa conversa, ensinamentos, discussão sobre tartarugas, estradas e outras coisas que muito contribuíram para minha formação profissional. Ao meu outro orientador do doutorado sanduíche, professor Henrique Miguel Pereira por ter me recebido na Alemanha.

Aos meus colegas de CBEE, os antigos que não estão mais aqui, mas não menos importantes: Hélio, Aline e Cristiane, e aos atuais, em especial o Ramon por toda ajuda, Rafaela, Priscilla, Flávio, Fernando. Pessoas especiais que levo comigo pra sempre! Aos meus colegas do grupo BIOCON - iDiv (Biodiversity Conservation), Laetitia Navarro, Miguel Fernandez, Silvia Ceausu, Inês Martins, Max Hoffmann, Alexandra Marques, Andrea Perino e em especial à Ana Ceia-Hasse, Ana Bárbara Gonçalves, Florian Wolf e Murilo Miranda pelos momentos de discussão científica de alto nível e conversa fiada. Foi sensacional!

Aos meus colegas de disciplinas, do departamento, amigos fora da UFLA (Albinha, Walter...), conhecidos que passaram pelo meu caminho nessa fase e que deixaram mensagens de apoio e de alegria.

À minha família, pai, mãe e irmãs (Milton, Magali, Stéfani e Bruna) pelo suporte, paciência nos momentos de ausência e carinho. Ao meu parceiro de vida, Bruno pelo apoio, sem ele acho que essa tese não teria saído!

Aos répteis, seres lindos que eu amo!

Aos que amam e trabalham com herpetologia. Aos estagiários do projeto de tartarugas no sul pelas horas incontáveis de campo para construir uma exorbitante base de dados (mania de grandeza do Alex) para que no futuro (agora) eu pudesse utilizar. Aos pesquisadores ao redor do mundo pelos seus trabalhos...eles foram a base para a construção deste trabalho. Por favor, não parem!

Meu muito obrigada!!

RESUMO

A biodiversidade enfrenta crescente pressão derivada da ação humana, incluindo alteração e degradação do hábitat, fragmentação, mudanças climáticas, exploração de espécies e poluição. Como consequência, avaliações sobre vários grupos de espécies mostram taxas de extinção extremamente altas. Estradas e outras infraestruturas lineares são um dos maiores modificadores da paisagem, especialmente em regiões tropicais. A formação dessas clareiras lineares causam vários impactos que são melhor compreendidos, mas não exclusivos, em escala local. Em uma escala mais ampla um aumento na densidade de rodovias gera um aumento de perda de áreas naturais na paisagem e conseqüentemente, há uma redução significativa no tamanho das manchas restantes que afetam a persistência das populações. Um bom conhecimento da história de vida das espécies é o primeiro passo e essencial para o desenvolvimento de estratégias de conservação para manter a persistência a longo prazo e recuperação de populações em declínio. Assim, a presente tese ao longo dos capítulos busca quantificar parâmetros básicos de história de vida, principalmente os relacionados à reprodução, as relações entre si e tamanho da fêmea. Utilizar informações de história de vida juntamente com parâmetros demográficos para estimar taxas de crescimento populacional para *Trachemys dorbigni* em uma região com vários tipos de ameaça à persistência da espécie. E, finalmente, usamos dados de história de vida, dinâmica populacional, mortalidade por atropelamento em estradas e dados de uso do solo (malha viária) em um modelo espacialmente explícito que quantifica valores críticos de densidade máxima e tamanho mínimo de manchas margeadas por estradas que permitem a persistência de populações de espécies de quelônios em paisagens fragmentadas. De um modo geral as estratégias reprodutivas da tartaruga tigre d'água variam entre as populações e com o tamanho da fêmea. Populações da espécie próximo à rodovia BR-471 na ESEC Taim estão declinando devido aos impactos observados na área (atropelamento de fauna e remoção de ovos). Fêmeas adultas é o principal estágio onde devemos focar esforços para a conservação da espécie em locais dentro da sua distribuição e que sofrem os mesmos potenciais impactos. Na avaliação global, observamos muitas regiões, principalmente na América do Norte e Ásia, que muitas espécies já possuem manchas de habitat menor que o tamanho viável para a sua persistência. Essas regiões são as que possuem as maiores densidades de rodovias. Esperamos que os resultados deste trabalho possam direcionar estudos futuros, serem usados por órgãos públicos para implementação de medidas específicas de mitigação, ações de conservação e no planejamento da melhoria e/ou construção de novas estradas.

Palavras-chave: *Trachemys dorbigni*. História de vida. Dinâmica populacional. Viabilidade Populacional. Estradas. Exploração ilegal. Avaliação global.

ABSTRACT

Biodiversity faces increasing pressure derived from human action, including habitat alteration and degradation, fragmentation, climate change, exploitation of species and pollution. Consequently, assessments of various species show extremely high extinction rates. Roads and other linear infrastructures are one of the greatest landscape modifiers nowadays, especially in tropical regions. These linear clearings cause numerous impacts that are better understood, but not exclusive, on a local scale. In a broader scale, an increase in road density leads to an increased loss of natural habitats and then, there is a distinguished decrease on the remaining patch size that affects population persistence. A good knowledge of the species life history is the first step and essential for the development of conservation strategies, to keep the long-term persistence and recover declining populations. Thus, this thesis throughout the following chapters seeks to quantify basic life history parameters, especially those related to reproduction in different populations, the relationship between reproductive parameters and between them and maternal body size. In addition, we aimed to use this life history information with demographic parameters to estimate population growth rates for *Trachemys dorbigni* in a region with many sources of threat to the species persistence. Finally, we used life history parameters, population dynamics, road mortality data and land use (road network) information and species distribution data in a spatially explicit model that quantifies critical values of maximum density and minimal patch size that allow the persistence of populations of turtle species in landscapes fragmented by roads. In general, reproductive strategies (mainly egg size) of D'Orbigni slider vary between populations and with body size. Species populations near the highway BR-471 at ESEC Taim may be declining due to the threats observed in the area (turtle road-kill and removal of eggs for pet trade). Adult females is the main stage where we should focus efforts for conservation programs of the species in locations within the distribution and that suffers with the same potential impacts. In the global assessment, we observed many regions, especially in North America and Asia, many species already have smaller patch size than the viable size to their persistence. These regions are those with the highest density of highways. We hope that the results of this study may direct future studies, be used by government agencies for implementation of specific mitigation measures, conservation actions and planning the improvement or construction of new roads.

Keywords: *Trachemys dorbigni*. Life history. Population dynamic. Population viability. Roads. Pet trade. Global assessment.

LISTA DE FIGURAS

Figura 1 -	A) Espécime de <i>Trachemys dorbigni</i> . B) Fêmea e ovos da espécie sendo mensurados. C) Indivíduos recém-eclodidos em criadouro clandestino no sul do país. D) Fêmea de atropelada na BR-471 rodovia que corta a Estação Ecológica do Taim.....	13
Artigo 1 - Figura 1	Location of the three populations of D'Orbigny's slider sampled in South of Brazil.....	50
Figura 2 -	Carapace length frequency histograms of reproductive females. (A) Arroio Grande; (B) ESEC Taim; (C) Pelotas.....	51
Figure 3 -	Maternal body size and reproductive traits of D'Orbigny's slider measured in three populations. Means with different letters were significantly different based on the Bonferroni adjusted <i>P</i> -value. The error bars represent 95% confidence limits.....	52
Figure 4 -	Linear relationship between clutch size and maternal body size at the three populations of D'Orbigny's slider. A) The linear model in Arroio Grande is described by $Y = \exp^{(0.012x - 0.508)}$. B) In ESEC Taim by $Y = \exp^{(0.007x + 0.651)}$. C) In Pelotas by $Y = \exp^{(0.0145x - 0.931)}$	53
Artigo 2 - Figure 1	Proportion of D'Orbygni slider turtle individuals in each stage in a stable distribution.....	82
Figure 2 -	The effect of different threats and levels of a) road mortality, b) nest predation and c) egg poaching on the population size of D'Orbygni slider turtle in Brazil. Baseline is the deterministic matrix projection.....	83
Figure 3 -	Best and worst case scenarios including lowest and highest values of all threats and their effects on the time to D'Orbygni slider population extirpation.....	84
Artigo 3 - Figure 1	Number of species assessed classified by IUCN status and their distribution within regions worldwide.....	112
Figure 2 -	Turtle species most affected by road density. Values of RD_{mx}/RD_{ob} comparing species within the 5th percentile with a species not affected (<i>Kinosternon flavescens</i>). Dashed red line include the boundary of road density values for all the species within the 25th percentile. LC = least concern, EN = endangered, CR = critically endangered, NT = near threatened status in IUCN.....	112

Figure 3 - Turtle species most affected by patch size within the 5th percentile. <i>Hardella thurjii</i> is among the 5% species with lower $P[PA_{ob} > PA_{mn}]$. In contrast, <i>Trachemys scripta</i> has high $P[PA_{ob} > PA_{mn}]$. Fewer patches larger than PA_{min} (areas shaded in grey) reveal a high vulnerability for <i>H. thurjii</i>	113
Figure 4 - Number of turtle species affected within 100 x 100 km grid cell where RD_{ob} is higher than RD_{mx} and/or $PA_{ob} > PA_{mn}$	114

LISTA DE TABELAS

Artigo 1 - Table 1	Variation of female size and reproductive traits of D'Orbigny's slider populations in Brazil. X^2 values of Kruskal-Wallis tests.....	47
Table 2 -	Linear and quadratic models fitted for three D'Orbigny's slider populations. a, b and c are parameters estimates. Indications of parameters estimates for linear models are followed by _(L) and for quadratic models by _(q) . * $P < 0.05$. EW = egg width, EL = egg length and EV = egg volume.....	48
Table 3 -	Response of clutch size to egg size to test for a possible trade-off for the three population of D'Orbigny's slider in Brazil.....	49
Artigo 2 - Table 1	Parameters used in the baseline deterministic matrix of D'Orbigny's slider turtle at the ESEC Taim, Brazil.....	81
Table 2 -	D'Orbygni slider turtle population growth rate (λ) response to simulations based on the threats road mortality and nest predation pressure. Scenario represents the percentage of the threat levels.....	81
Artigo 3 Table 1	Turtle species most affected by minimal patch size. P [$PA_{ob} > PA_{mn}$] are the lowest values inside the 5th percentile. IUCN Red List status are CR: Critically Endangered, EN: Endangered, LC: Least Concern and VU: Vulnerable.....	111

SUMÁRIO

	PRIMEIRA PARTE.....	12
1	INTRODUÇÃO GERAL.....	12
2	REFERENCIAL TEÓRICO.....	15
2.1	Quelônios.....	15
2.2	Ameaças à perda de biodiversidade e o impacto de rodovias....	16
	REFERÊNCIAS.....	21
	SEGUNDA PARTE - ARTIGOS	26
	ARTIGO 1 - Contrasting reproductive strategies of D’Orbigny’s slider turtle at the same latitude range in subtropical wetlands.....	26
	ARTIGO 2 - Consequence of threats to freshwater turtle’s viability in subtropical wetlands.....	54
	ARTIGO 3 - Global road density jeopardize turtle persistence	87

PRIMEIRA PARTE

1 INTRODUÇÃO GERAL

A presente tese é um dos vários resultados obtidos de alguns projetos de Quelônios do Rio Grande do Sul desenvolvidos entre 1995 à 2006. Durante os anos de pesquisa, os projetos buscavam identificar os padrões de morfologia, reprodução, comportamento de nidificação, dieta e história de vida de quatro espécies de quelônios de ocorrência no estado: *Trachemys dorbigni* (Duméril & Bibron, 1835) (Figura 1A), *Hydromedusa tectifera*, *Acanthochelys spixii* e *Phrynops williamsi*. A ideia deste trabalho se originou a partir de uma base de dados de monitoramento de longo prazo feito em vários locais para *T. dorbigni* e posteriormente foi extrapolada para várias espécies de quelônios mundiais.

No primeiro capítulo, abordamos informações sobre a reprodução de *T. dorbigni* coletadas em três localidades do Rio Grande do Sul entre 1995 a 2006 (Figura 1, Artigo 1). Fêmeas adultas foram mensuradas e os ninhos quantificados em relação ao número e tamanho dos ovos (Figura 1B). Essas informações foram usadas no intuito de comparar se os atributos reprodutivos variam entre diferentes populações, testar a relação entre os atributos reprodutivos para identificar possíveis *trade-offs* e com o tamanho corporal das fêmeas. Os resultados deste estudo fornecem informações base de história de vida que são essenciais para estudos demográficos e de conservação dessa espécie.

O segundo artigo utiliza informações sobre reprodução (Artigo 1) e parâmetros demográficos em uma abordagem de modelagem populacional baseada no indivíduo para estimar a taxa de crescimento populacional de *T. dorbigni* na região da Estação Ecológica do Taim (ESEC Taim) no extremo sul do Rio Grande do Sul. Na região da estação (dentro e fora dela) a espécie sofre diferentes pressões (intensa predação, exploração para o tráfico ilegal de animais silvestres e mortalidade devido ao atropelamento de fauna) (Figura 2A e 2B) que

ameaçam sua persistência a longo prazo. Os resultados deste estudo fornecem subsídios para recomendação de diferentes estratégias de conservação atuando em estágios de vida críticos da espécie.

O terceiro capítulo através de uma compilação da literatura e bases de dados disponíveis utiliza parâmetros de história de vida, dinâmica populacional, mortalidade por atropelamento em estradas e dados de infraestrutura utilizando um modelo espacialmente explícito que quantifica valores críticos de densidade máxima de estradas e tamanho crítico de manchas rodeadas por estradas que permitem a persistência de populações em paisagens fragmentadas por estas.

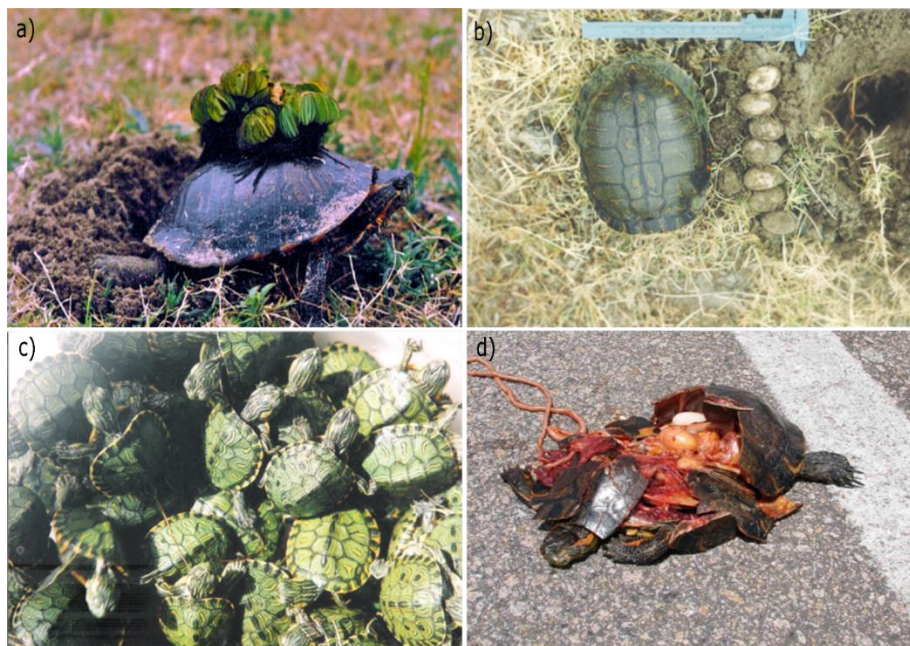


Figura 1: A) Espécime de *Trachemys dorbigni*. B) Fêmea e ovos da espécie sendo mensurados. C) Indivíduos recém-eclodidos em criadouro clandestino no sul do país. D) Fêmea de atropelada na BR-471 rodovia que corta a Estação Ecológica do Taim. Fotos: A. Bager.

Este modelo foi aplicado para as espécies de quelônios terrestres e de água doce distribuídos ao redor do mundo. A partir da quantificação dos valores críticos, esta abordagem permite o ranqueamento das espécies com maior vulnerabilidade e mapear onde dentro de suas áreas de distribuição existam áreas críticas para a espécie em relação ao impacto das estradas. Essa abordagem também permite a identificação de espécies e locais que necessitem de medidas específicas de mitigação, ações de conservação além de poder ser usada para informar esquemas de priorização para a melhoria e/ou construção de novas estradas.

2 REFERENCIAL TEÓRICO

2.1 Quelônios

Os quelônios formam um grupo monofilético que compreende a ordem Testudines. Até o momento, 335 espécies e mais 118 subespécies são documentadas (VAN DIJK et al. 2014) ocorrendo desde regiões temperadas até tropicais do planeta. O grupo é morfologicamente bastante diverso, incluindo espécies de habitat terrestres, semi-aquáticas (por exemplo, rios, córregos, lagos, lagoas, pântanos e outros habitats úmidos) e marinhas. Aproximadamente 55% das espécies estão listadas na Lista Internacional Vermelha de Espécies Ameaçadas (IUCN), e muitas destas incluídas classificadas em categorias como Quase Ameaçada, de Menor Interesse ou com Dados Deficientes (IUCN 2016).

Grande parte das espécies possuem características de história de vida como crescimento corporal lento, maturidade tardia, alta fecundidade (reproduzindo por vários anos), alta mortalidade nos juvenis, baixa mortalidade nos adultos e longevidade (CONGDON et al. 1993). Há grande variação no tamanho corporal das espécies desde tamanhos pequenos como nos gêneros *Kinosternon* e *Sternotherus* (10 cm de comprimento de carapaça) até algumas espécies como as do gênero *Podocnemis* e família Tryonichidae que atingem 60-70 cm de tamanho corporal (IVERSON 1982). Da mesma forma, as espécies de tartarugas exibem grande variação na quantidade de ovos depositados, desde um único até centenas (ERNST; LOVICH 2009).

Espécies de tartarugas são conspícuas nos ecossistemas que ocupam e podem servir como indicadoras de qualidade ambiental. Em muitos dos locais as populações de várias espécies estão encolhendo e o risco de extinção aumenta exponencialmente com a poluição, fragmentação, perda de hábitat, remoção de indivíduos, introdução de espécies e disseminação de doenças (GIBBONS et al. 2000). Por mais que tartarugas tendam a ser um táxon não prioritário em

amostragens de vertebrados, o estudo destas espécies podem fornecer informações úteis sobre a saúde do ecossistema em que vivem, tendências populacionais e risco de extinção frente aos impactos que atingem organismos com histórias de vida tão particulares.

2.2 Ameaças à perda de biodiversidade e o impacto de rodovias

Atualmente, a biodiversidade enfrenta crescente pressão derivada da ação humana, incluindo alteração e degradação do hábitat, fragmentação, mudanças climáticas, exploração de espécies e poluição (NEWBOLD et al. 2014, 2015). Em uma escala de comparação, os impactos decorrentes da alteração no uso do solo são maiores que os impactos por exemplo, da exploração e da poluição (PEREIRA et al. 2012). Como consequência, avaliações sobre vários grupos de espécies mostram taxas de extinção extremamente altas (CHAPIN III et al. 2000; PEREIRA et al. 2010; PIMM et al. 2014), consideravelmente mais altas que taxas de perda estimadas para o século passado (PIMM et al. 2014) e períodos ainda mais remotos (PIMM et al. 1995). Previsões para o fim deste século projetam taxas de extinção de aproximadamente 18 % devido ao desmatamento em áreas de *hotspots* de florestas tropicais (PIMM; RAVEN 2000) e perdas globais de riqueza de espécies de 3.4% concentradas em regiões ricas mas com países economicamente pobres (NEWBOLD et al. 2015).

Estradas e outras infraestruturas lineares são um dos maiores modificadores da paisagem na atualidade, especialmente em regiões tropicais (LAURANCE et al. 2009; 2011). A formação dessas clareiras lineares causam vários impactos que são melhor compreendidos, mas não exclusivos, em escala local (BAGER; FONTOURA 2013; JAEGER 2015). Por exemplo, altas taxas de mortalidade de fauna devido à colisões com veículos (GRILO et al. 2009; BAGER; ROSA 2011; COELHO et al. 2012), alterações na luminosidade,

umidade e regimes de ventos devido ao efeito de borda (FORMAN; ALEXANDER 1998), distúrbios do tráfego (barulho e emissão de poluentes) (PARRIS 2015) e facilitação da propagação e dispersão de espécies exóticas (TROMBULAK; FRISSEL 2000) são alguns dos principais mecanismos pelos quais a biodiversidade é afetada.

O entendimento destes efeitos em níveis de organização maiores é de extrema importância (VAN DER REE et al. 2011) até porque alguns efeitos negativos das estradas ocorrem na escala de paisagem, como por exemplo, a fragmentação de habitats (JAEGER 2015). Estradas contribuem neste processo através da densidade da malha viária. Um aumento na densidade de rodovias gera um aumento de perda de áreas naturais na paisagem (LEE et al. 2004; GOOSEM 2007) e conseqüentemente, há uma redução significativa no tamanho das manchas restantes que afetam a persistência das populações (JAEGER; HOLDEREGGER 2005; JAEGER et al. 2005; JAEGER 2015). Com a diminuição do tamanho efetivo das manchas, o arranjo espacial destas também muda, aumentando a distância entre habitats adequados e disponíveis perdendo em termos de conectividade da paisagem (FAHRIG 2003). A perda de conectividade aumenta o efeito barreira e atropelamento de fauna silvestre, limitando possibilidades de dispersão (ROEDENBECK et al. 2007). Logo, estes efeitos a nível de paisagem geralmente estão atrelados à persistências populacionais a longo prazo (JAEGER 2015), porque persistência populacional é regulado por processos que operam em escalas maiores (ROEDENBECK et al. 2007). Uma simples contagem do número de colisões entre veículos e fauna silvestre ou a mensuração da zona de efeito na escala local de uma rodovia, sozinhos não irão informar o quanto estes empreendimentos lineares estão impactando a existência de populações ou espécies (VAN DER REE et al. 2011). Em uma escala espacial mais ampla, entender como a densidade e configuração da malha viária (HEILMAN et al. 2002) afetam as relações funcionais dentro e entre ecossistemas e as persistências

populacionais são parâmetros de prioridade para melhor subsidiar estratégias de planejamento do transporte (VAN DER GRIFT 2005; VAN DER REE et al. 2011; JAEGER 2015).

A malha viária global expandiu a níveis relevantes desde o último século (VAN DER REE et al. 2015). Pelo menos 25 milhões de quilômetros de novas estradas estão previstos até 2050 (DULAC 2013), expansão principalmente em regiões que compreendem muitos países em desenvolvimento (LAURANCE et al. 2014; VAN DER REE et al. 2015). A América Latina, principalmente o Brasil se encaixam neste contexto (DULAC 2013; BAGER et al. 2015). O governo brasileiro nos últimos anos tem priorizado a construção de novas rodovias e no ano de 2014 a estimativa era de que aproximadamente 7000 km de estradas e 3000 km de ferrovias fossem construídas (BAGER et al. 2015).

As áreas para expansão da malha viária para promover crescimento social e econômico no Brasil vão de encontro à regiões de elevada biodiversidade (MITTERMEIER et al. 2015). Especificamente répteis, há uma alta riqueza de espécies e endemismos na região tropical da América Latina (UNEP 2010) onde uma em cada cinco espécies de répteis são ameaçadas de extinção e essa mesma proporção de espécies são classificadas como “Data Deficient” (BÖHM et al. 2013). Perda e degradação do habitat novamente são apontados como os fatores mais importantes (PEREIRA et al. 2010; 2012) induzido principalmente pela da alteração e redução de habitats terrestre e aquático (GIBBONS et al. 2000). O mecanismo pelo qual as espécies estão declinando permanecem desconhecidos mas são principalmente relacionados à degradação geral da qualidade do habitat a partir da conversão antrópica do uso da terra (TODD et al. 2010). Existe uma imensa lacuna em determinadas regiões, incluindo o Brasil, que limita o entendimento de como estas ameaças afetam as espécies de répteis (BÖHM et al. 2013). Além disso, o grupo é ainda insuficientemente representado em avaliações feitas pela Lista Vermelha de Espécies Ameaçadas da IUNC. Apenas 37% das

espécies descritas foram avaliadas e a maior parte delas carece de informações básicas sobre tendências populacionais e ameaças, o que os tornam um grupo de alto interesse para conservação (BÖHM et al. 2013).

Um bom conhecimento da história de vida das espécies é o primeiro passo e essencial para o desenvolvimento de estratégias de conservação para manter a persistência a longo prazo e recuperação de populações em declínio (SHINE; CHARNOV 1992; FERGUNSSON; LARIVIÈRE 2002; FOSTER; VINCENT 2004). Os atributos de história de vida, por exemplo, número de crias produzidos, idade na primeira reprodução, crescimento, longevidade frequentemente variam consistentemente em relação ao habitat ou às condições do ambiente (ROFF 1992), ou seja, a estratégia de história de vida de uma espécie pode influenciar como as populações dessa espécie respondem à variação ambiental. Programas de conservação, muitas vezes são formulados sem dados adequados de atributos de história de vida para espécies alvo (CONGDON et al. 1994). Na ausência deste tipo de dado adequado, conhecimento dos limites realistas das características demográficas (principalmente os documentados em natureza e que mantem populações estáveis ou mesmo aumente em tamanho) podem ser extremamente importantes para a tomada de decisão sobre estratégias de manejo em populações de vertebrados (DUNHAM; OVERALL, 1994).

Recentemente, vários estudos utilizando essas informações predizem como o risco de extinção de uma espécie pode estar relacionado com suas características de história de vida (HUTCHING 2002; PEARSON et al. 2014). Por exemplo, espécies com baixa fecundidade, taxas de crescimento lentas e maturação tardia são mais vulneráveis à extinção porque apresentam maior dificuldade em compensar altas taxas de mortalidade, principalmente nos estágios adultos, como é o caso de várias espécies de tartarugas (CROUSE et al. 1987; CONGDON et al. 1993; 1994). Diante do cenário de ameaças nos quais as espécies estão expostas, informações sobre a história de vida são determinantes

fundamentais na dinâmica das populações e como estas respondem aos vários distúrbios causados pelo homem (FERGUNSSON; LARIVIÈRE 2002).

Este trabalho teve como finalidade quantificar e identificar parâmetros básicos de história de vida e dinâmica populacional atuando tanto em escala local para a espécie *Trachemys dorbigni* e em escala global através do levantamento dessas informações para várias outras espécies de quelônios de água doce. O uso dessas informações foi integrado em modelos populacionais mecanicistas e espacialmente explícitos para identificar tendências e discutir estratégias de conservação possíveis para mitigar os impactos de exploração para o comércio de animais silvestres e mortalidade em estradas.

REFERÊNCIAS

BAGER, A.; BORGHI, C. E.; SECCO, H. **The influence of economics, politics and environment on road ecology in South America.** Em: VAN DER REE, R.; SMITH, D.J.; GRILO, C. (Eds.). Handbook of Road Ecology, pp. 407-413. Wiley Blackwell, Oxford, 2015.

BAGER, A.; ROSA, C. A. Influence of sampling effort on the estimated richness of road-killed vertebrate wildlife. **Environmental Management**, v. 47, n. 5, p. 851-858, 2011.

BAGER, A.; FONTOURA, V. Evaluation of the effectiveness of a wildlife roadkill mitigation system in wetland habitat. **Ecological Engineering**, v. 53, p. 31-38, 2013.

BÖHM, M. et al. The conservation status of the world's reptiles. **Biological Conservation**, Washington, v. 157, p. 372-385, 2013.

CHAPIN, F. S.; ZAVALETA, E. S.; EVINER, V. T.; NAYLOR, R. L.; VITOUSEK, P. M.; REYNOLDS, H. L.; HOOPER, D. U.; LAVOREL, S.; SALA, O. E.; HOBBIIE, S. E.; MACK, M. E.; DÍAZ, S. Consequences of changing Biodiversity. **Nature**, London, v. 405, n. 6783, p. 234-242, 2000.

COELHO, I. P.; TEIXEIRA, F. Z.; COLOMBO, P.; COELHO, A. V. P.; KINDEL, A. Anuran road-kills neighboring a peri-urban reserve in the Atlantic Forest, Brazil. **Journal of Environmental Management**, v. 112, p. 17-26, 2012.

CONGDON, J. D.; DUNHAN, A. E.; VAN LOBEN SELS, R.C. Delayed sexual maturity and demographics of Blandingii's turtle (*Emydoidea blandingii*): implications for conservation and management of long-lived organisms. **Conservation Biology**, Washington, v. 7, n. 4, p. 826-833, 1993.

CONGDON, J. D.; DUNHAN, A.E., VAN LOBEN SELS, R.C. Demographics of the common snapping turtles (*Chelydra serpentina*): implications for conservation and management of long-lived organisms. **American Zoologist**, v. 34, 397-408, 1994.

CROUSE, D. T.; CROWDER, L. B.; CASWELL, H. A stage-based population model for loggerhead sea turtles and implications for conservation. **Ecology**, Washington, v. 68, n. 5, p. 1412-1423, 1987.

DULAC, J. Global Land Transport Infrastructure Requirements: Estimating road and railway infrastructure capacity and costs to 2050. França, 2013. 54 p.

DUNHAM, A. E.; OVERALL, K. L. Population responses to environmental change: Life history variation, individual-based models, and the population dynamics of short-lived organisms. **American Zoologist**, v. 34, n. 3, p. 382-396, 1994.

ERNST, C. H.; LOVICH, J. E. (Eds). **Turtles of the United States and Canada**. JHU Press, 2009.

FAHRIG, L. Effects of Habitat Fragmentation on Biodiversity. **Annual Review of Ecology, Evolution and Systematics**, v. 34, p. 487-515, 2003.

FERGUSON, S. H.; LARIVIERE, S. Can comparing life histories help conserve carnivores? **Animal Conservation**, Cambridge, v. 5, n. 1, p. 1-12, 2002.

FORMAN, R.T.T.; ALEXANDER, L. E. Roads and Their Major Ecological Effects. **Annual Review of Ecology, Evolution and Systematics**, v. 29, p. 207-231, 1998.

FOSTER, S. J.; VINCENT, A. C. J. Life history and ecology of seahorses: implications for conservation and management. **Journal of Fish Biology**, v. 65, n. 1, p. 1-61, 2004.

GIBBON, J.W.; SCOTT, D. E.; RYAN, T. J.; BUHLMANN, K. A.; TUBERVILLE, T. E.; METTS, B. S.; GREENE, J. L.; MILLS, T.; LEIDEN, Y.; POPPY, S.; WINNE, C. T. The Global Decline of Reptiles, Déjà Vu Amphibians. **Bioscience**, Oxford, v.50, p. 653-666, 2000.

GOOSEM, M. Fragmentation impacts caused by roads through rainforests. **Current Science**, Bengaluru, v. 93, p. 1587-1595, 2007.

GRILO, C.; BISSONETTE, J. A.; SANTOS-REIS, M. Spatial–temporal patterns in Mediterranean carnivore road casualties: consequences for mitigation. **Biological Conservation**, Washington, v. 142, n. 2, p. 301-313, 2009.

HEILMAN, G.E. et al. Forest Fragmentation of the Conterminous United States: Assessing Forest Intactness through Road Density and Spatial Characteristics. **Bioscience**, Oxford, v. 52, p. 411-422, 2002.

HUTCHINGS, J. A. **Life histories of fish**. Em: HART, P. J. B.; REYNOLDS, J. D. (Eds.). Handbook of fish biology and fisheries, pp. 149-174. Blackwell Science, Oxford, 2002.

INTERNATIONAL UNION FOR CONSERVATION OF NATURE. The IUCN **Red List of Threatened Species**. Cambridge, 2016. Disponível em: <<http://www.iucnredlist.org/>>. Acesso em: 20 jan. 2016.

IVERSON, J. B. Biomass in turtle populations: a neglected subject. **Oecologia**, Berlin, v. 55, p. 69-76, 1982.

JAEGER, J.A.G. **Improving Environmental Impact Assessment and Road Planning at the Landscape Scale**. Em: VAN DER REE, R.; SMITH, D.J.; GRILO, C. (Eds.). Handbook of Road Ecology, pp. 32-42. Wiley Blackwell, Oxford, 2015.

JAEGER, J.A.G.; HOLDEREGGER, R. Thresholds of landscape fragmentation (in German: Schwellenwerte der Landschaftszerschneidung). **GAIA**, v. 14, p. 113-118, 2005.

JAEGER, J.A.G.; BOWMAN, J.; BRENNAN, J.; FAHRIG, L.; BERT, D.; BOUCHARD, J.; CHARBONNEAU, N.; FRANK, K.; GRUBER, B.; VON TOSCHANOWITZ, K. T. Predicting when animal populations are at risk from roads: An interactive model of road avoidance behavior. **Ecological Modelling**, v. 185, p. 329-348, 2005.

LAURANCE, W. F.; COCHRANE, M. A.; BERGEN, S.; FERANSIDE, P. M.; DELAMÔNICA, P.; BARBER, C.; DANGELO, S.; FERNANDES, T. The future of the Brazilian Amazon. **Science**, New York, v. 291, p. 438-439, 2001.

LAURANCE, W. F.; GOOSEM, M.; LAURANCE, S.G.W. Impacts of roads and linear clearings on tropical forests. **Trends in Ecology and Evolution**, Cambridge, v. 24, n. 12, p. 659-669, 2009.

LAURANCE, W. F.; CLEMENTS, G. R.; SLOAN, S.; O'CONNEL, C.S. A global strategy for road building. **Nature**, London, v. 513, p. 229-232, 2014.

LEE, P. F.; HSU, F. H.; GENG, S. Breeding bird species richness in Taiwan: distribution on gradients of elevation, primary productivity and urbanization. **Journal of Biogeography**, v. 31, p. 307-314, 2004.

MITTERMEIER, R.A.; VAN DIJK, P. P.; RHODIN, A. G. J.; NASH, S. D. Turtle Hotspots: An Analysis of the Occurrence of Tortoises and Freshwater Turtles in Biodiversity Hotspots, High-Biodiversity Wilderness Areas, and Turtle Priority Areas. **Chelonian Conservation and Biology**, v. 14,n. 1, p. 2-10, 2015.

NEWBOLD, T.; HUDSON, L.N.; PHILLIPS, H.P.; HILL, S.L.L.; CONTU, S. A global model of the response of tropical and sub-tropical forest biodiversity to anthropogenic pressures. **Proceedings of the Royal Society of London B: Biological Sciences**, London, v. 281, n. 1792, p. 20141371, 2014.

NEWBOLD, T.; HUDSON, L.N.; HILL, S.L.L. Global effects of land use on local terrestrial biodiversity. **Nature**, London, v. 520, p. 45-50, 2015.

PEARSON, R. G.; STANTO, J. C.; SHOEMKER, K. T.; AIELLO-LAMMENS, M. E.; ERSTS, P. J.; FORDHAM, D. A.; RAXWORTHY, C. J.; RYU, H. Y.; MCNEES, J.; AKÇAKAYA, H. R. Life history and spatial traits predict extinction risk due to climate change. **Nature Climate Change**, v. 4, n. 3, p. 217-221, 2014.

PEREIRA, H.M.; LEADLEY, P.; RPOENÇA, V. Scenarios for global biodiversity in the 21st century. **Science**, New York, v. 330, n. 6010, p. 1496-1501, 2010.

PEREIRA, H. M.; NAVARRO, L. M.; MARTINS, I. S. Global biodiversity change: the bad, the good, and the unknown. **Annual Review of Environment and Resources**, v. 37, p. 25-50, 2012.

PIMM, S. L.; RUSSELL, G. J.; GITTLEMAN, J. L.; BROOKS, T. M. The future of biodiversity. **Science-AAAS-Weekly Paper Edition**, v. 269, n. 5222, p. 347-349, 1995.

PIMM, S. L.; JENKINS, C. N.; ABELL, R.; BROOKS, T. M.; GITTLEMAN, L.; JOPPA, L. N.; RAVEN, P. H.; ROBERTS, C. M.; SEXTON, J. O. The biodiversity of species and their rates of extinction, distribution, and protection. **Science**, New York, v. 344, n. 6187, p. 1246752, 2014.

PIMM, S. L.; RAVEN, P. Biodiversity: extinction by numbers. **Nature**, London, v. 403, n. 6772, p. 843-845, 2000.

ROEDENBECK, I. A.; FAHRIG, L.; FINDLAY, C. S.; HOULAHAN, J. E.; JAEGER, J. A. G.; KLAR, N.; KRAMER-SHADT, S.; VAN DER GRIFT, E.

The Rauschholzhausen agenda for road ecology. **Ecology and Society**, v. 12, n. 11, p. 241-252, 2007.

ROFF, D. A. (Ed). **The evolution of life histories: theories and analysis**. Chapman & Hall, New York, 1992.

SHINE, R.; CHARNOV, E. L. Patterns of survival, growth, and maturation in snakes and lizards. **The American Naturalist**, v. 1992, p. 1257-1269, 1992.

TODD, B. D.; WILLSON, J. D.; GIBBONS, J. W. **The global status of reptiles and causes of their decline**. Em: Sparling, D. W.; Linder, G.; Bishop, C. A.; Krest, S. (Eds.). *Ecotoxicology of amphibians and reptiles*, pp. 47-67. CRC Press, New York, 2010.

TROMBULAK, S. C.; FRISSELL, C. A. Review of ecological effects of roads on terrestrial and aquatic communities. **Conservation Biology**, Washington, v. 14, n. 1, p. 18-30, 2000.

VAN der REE, R.; JAEGER, J. A. G.; VAN DER GRIFT, E.; CLEVINGER, A. P. Effects of roads and traffic on wildlife populations and landscape function: Road ecology is moving toward larger scales. **Ecology and Society**, v. 16, n. 1, p. 48, 2011.

VAN der GRIFT, E. A. Defragmentation in the Netherlands: a success story? **GAIA - Ecological Perspectives for Science and Society**, v. 14, n. 2, p. 144-147, 2005.

VAN DIJK, P. P.; IVERSON, J. B.; RHODIN, A. G. J.; SHAFFER, H. B.; BOUR, R. *Turtles of the World, 7th Edition: Annotated Checklist of Taxonomy, Synonymy, Distribution with Maps, and Conservation Status*. **Chelonian Research Monographs**, v. 5, p. 243-328, 2014.

SEGUNDA PARTE - ARTIGOS**ARTIGO 1****Comparando estratégias reprodutivas da tartaruga Tigre d'água na mesma amplitude latitudinal em zonas úmidas subtropicais.**

Preparado de acordo com as normas da Revista Zoologischer Anzeiger.

Lucas P.S., Bager A. (2016) Contrasting reproductive strategies of D'Orbigny's slider turtle at the same latitude range in subtropical wetlands.

Priscila da Silva Lucas^{a,b,c} & Alex Bager^{a,b}

^aPrograma de Pós-Graduação em Ecologia Aplicada, Departamento de Biologia, Universidade Federal de Lavras, Campus Universitário, CP 3037, Lavras, CEP 37200 000. Brazil. Tel.: +55 35 3829 1924.

^bCentro Brasileiro de Estudos em Ecologia de Estradas, Universidade Federal de Lavras, Campus Universitário, CP 3037, Lavras, CEP 37200 000. Brazil. Tel.: +55 35 3829 3129.

^cCorresponding author: Priscila da Silva Lucas: Tel.: +55 35 3829 3129, Fax: +55 35 3829 1924; E-mail: prilucass@gmail.com

Abstract: Comparing how reproductive attributes and investment vary between individuals and species, and how they can be apportioned among offspring, are two central issues in life history theory. Maternal body size plays an important role in reproductive variation, however, we usually observe variable investment in offspring by females, and optimal egg size may not be achieved in some populations or species. Here, we tested if reproductive traits differed between populations of D'Orbigny's slider turtle in close geographical areas in Brazil. We evaluated the relationship between reproductive traits to maternal body size and clutch size to egg size to determine possible trade-offs across populations. Population had a significant effect on maternal body size and reproductive traits even in geographically close areas. In general, reproductive traits were larger in populations in which females were relatively larger. Maternal body size had a positive effect on clutch size, but not on egg size, except in Arroio Grande. We did not observe a negative correlation between clutch and egg size across any population. Although body size had relevant effects in different populations, we discuss other possible factors that could also cause variation in life history traits in these populations.

Keywords: turtle reproductive traits; geographic variation; optimal egg size; *Trachemys dorbigni*; life history.

1. Introduction

Classical life-history theory is based on optimization models (Smith and Fretwell, 1974, Stearns, 2000) that try to explain how evolution designs organisms to reach reproductive success (Stearns, 2000). The theory has a number of assumptions as consequence of the prediction of a single optimal offspring size that maximizes parental fitness in a given environment (Smith and Fretwell, 1974). Understand why some reproductive traits vary and how reproductive investment can be apportioned among offspring to optimize the trade-off between offspring size and number are one of the main concerns among life history (Roff, 1992, Stearns, 2000).

Life history traits exhibit extensive variation in many organisms and between–population of the same species (e.g. Iverson et al., 1997, Ashton et al., 2007, Macíp-Rios et al., 2013, Blew et al., 2016, Salguero-Gómez et al., 2016). For instance, in freshwater turtles, temperature, seasonality and productivity (as a proxy of resource availability) have already been inferred as proximate causes of reproductive variations within species range (Iverson and Smith, 1993; Ashton et al., 2007). Although there are evidences that environmental factors explain reproductive variations in populations at broader geographical scales (mostly at different latitudes), little is known on populations in close proximity. Individual traits such as, breeding age, genetic, home range quality, foraging places and maternal body size may explain reproductive variations at small scales (Wallis et al., 1999, Iverson and Smith 1993, Rowe 1994a, 1994b, Valenzuela 2001, Litzgus and Mousseau 2006; Naimi et al., 2012).

The predicted assumption that females will expend their available energy to reproduce eggs of an optimal size in the Smith and Fretwell (1974) model has been supported by some turtle species (Lovich et al. 2012; Macíp-Rios et al. 2012). On the other hand, this simple concept could not apply within to all species or populations of conspecifics and females can invest variably in offspring size (Rowe et al. 2003; Wilkinson and Gibbons 2005; Rollinson and Brooks 2008; Macíp-Rios et al. 2012, 2013). There are, at least three general categories of explanation in which theoretical or empirical models can predict maternal correlation with egg size (Rollinson and Rowe, 2015) and some could apply to turtles. For instance, female morphological structures can pose some constraints on egg size, i.e., egg size increases with female body size until they become large enough to produce eggs of an optimal size (Congdon and Gibbons 1987; Rollinson and Brooks 2008). In addition, in species that experience high depredation in a short time period of the life cycle (such as some turtles) lay larger clutches of small offspring size is beneficial (McGinley 1989). Females with larger maternal resources level should also produce larger offspring size even when larger clutches increase the probability of offspring survival (McGinley 1989, Rollinson and Rowe 2015).

Here, we first tested if reproductive traits differ between geographically close populations of the freshwater turtle, *Trachemys dorbigni* (D'Orbigny's slider) (Duméril and Bibron 1835) (Testudines: Emydidae) in South of Brazil. Second, we evaluated the relationship between reproductive traits and maternal body size. We tested 1) whether maternal body size explain variation in egg size and clutch size in populations of D'Orbigny's slider with different gradient of body sizes. We

hypothesized a relationship in which egg size does not vary with body size. With increasing female body size within populations, we expected to find larger clutch size rather than larger egg size as the reproductive strategy adopted by females. These would be exhibited by a positive correlation between clutch size and maternal body size, but not for egg size. In addition, we investigated 2) how clutch size vary with egg size expecting for a trade-off between these traits (Iverson and Smith 1993; Rowe 1994a; Rasmussen and Litzgus 2010).

2. Material and Methods

2.1 Species model

D'Orbigny's slider is a medium-sized (250 mm of carapace length) freshwater turtle species, abundant in many wetlands in Rio Grande do Sul, Brazil (the only Brazilian state where this species has not been introduced) (Bujes and Verrastro, 2008). There is sexual dimorphism in both size (males are smaller than females) and color pattern (Bager et al., 2010). Previous studies have shown that females attaining carapace length of at least 176 mm (Fagundes et al., 2010) can reproduce, but their size at maturity can vary across populations (Bager et al., 2007).

2.2 Study area

We studied three populations of D'Orbigny's slider in the Rio Grande do Sul state, Brazil between 1995 and 2006: Arroio Grande, Pelotas and at the Ecological Station of Taim - Santa Marta Farm, a Protected Area (hereafter ESEC Taim) (Figure 1). All three sites are located near the coastal plain, which encompasses extensive marshes forming the largest

South American lagoon complex (Bager et al., 2007 and Fagundes et al., 2010). Mean annual temperature ranged from 18°C to 19°C and mean annual precipitation ranged from 1200 to 1400 mm (Matzenauer et al., 2011). All study areas are under human pressure such as: intensive egg harvest and exploitation for illegal pet trade (A. Bager, personal observation), agricultural activities near wetlands (Bager and Rosado, 2010), and collisions with vehicles (Hahn et al., 2014).

2.3 Life-history traits

Female turtles were manually captured in the field during reproductive activity season, their maximum carapace length was measured with a caliper to the nearest 0.1 mm, the individuals were weighed with Pesola spring scales of 1 g, and individually marked (Cagle, 1939). We also search for turtle nests. Only intact nests were excavated to determine clutch and egg size (length and width).

2.4 Data Analysis

Exploratory analyses were performed to assess the normality and homogeneity assumptions for all variables and to detect possible extreme outliers (Zuur et al., 2007). We computed an equation based on the upper and lower quartiles for all variables (Bager et al., 2012) to minimize errors from measurements and exclude outliers. We estimated clutch volume and egg volume based on the equation described by Maritz and Douglas (1994) accounting for the bicone format. Here, we used egg volume as a proxy of egg mass in all populations since the two variables are highly correlated in this species ($R^2 = 0.91$; Bager, 2003).

To detect differences between reproductive traits and maternal body size among populations, we used a non-parametric Kruskal-Wallis test to check the population effect on clutch and egg size measurements. When we detected significant differences, Dunn *post hoc* tests were used to check pairwise differences between populations.

We applied an analysis of covariance to assess whether there was a population effect on these attributes after removal of the maternal body size effect. Each clutch and egg measure was included as an independent variable, the female carapace length as a covariate and the population as a treatment.

We fitted Generalized Linear Models (GLM) to assess the role of maternal body size on egg and clutch size, with Gaussian and Poisson distributions, respectively. We fitted linear and quadratic models and used Akaike Information Criteria (AIC) to select the model that better explain the relation between reproductive traits and maternal body size (Burnham and Anderson, 2002).

To find a trade-off between clutch size and egg size we run a Generalized Linear Mixed Model (GLMM). We treated clutch size as the response variable, egg size measures as fixed covariates and population as a random factor. We used clutch average ($n = 571$ clutches of the 2.185 eggs measured) as a unit of measure in all analysis. All analysis were performed with R software (R Core Team 2015) and the “nlme” package was used to run the GLMM (Pinheiro et al., 2016).

3. Results

3.1 Body size and life history traits among populations

Sizes class of reproducing female varied among three populations (Figure 2). Most of reproductive females in Arroio Grande had between 198 mm and 218 mm of carapace length whereas in Pelotas, we recorded between 192 and 198 mm and in ESEC Taim, most had between 224 and 235 mm.

We found a population effect on maternal body size regarding maximum carapace length ($\chi^2 = 154.04$, $gl = 2$, $P < 0.0001$) (Table 1). Dunn *post hoc* tests showed that females of ESEC Taim (mean: 233.6 ± 13.72) were significantly larger than females from other populations ($P < 0.01$ in all comparisons). Females of Arroio Grande and Pelotas had similar body sizes ($P > 0.05$; Figure 3).

We also observed a population effect on clutch size ($\chi^2 = 110.42$, $gl = 2$, $P < 0.01$) (Table 1). Females at the ESEC Taim laid larger clutches than those in Arroio Grande and Pelotas ($P < 0.05$), and the latter showed the smallest clutch sizes (Figure 3). Excluding the maternal body size effect, clutch size was similar among all populations (one-way ANCOVA: $F_{2,296} = 2.49$, $P = 0.08$). However, we did not observe the same trend for clutch volume, whose values were greatest at ESEC Taim, followed by Arroio Grande, and Pelotas (one-way ANCOVA: $F_{2,122} = 32.4$, $P < 0.0001$).

A significant population effect on egg size in general was also observed (Table 1). Egg width and volume were significantly different in all populations, and egg length was similar between Arroio Grande and ESEC Taim, but smaller at Pelotas (Figure 3). Significant differences were observed even after removing maternal body size effect for egg length (one-way ANCOVA: $F_{2,122} = 7.85$, $P < 0.01$) and width (one-way ANCOVA: $F_{2,122} = 42.4$, $P < 0.0001$). Tukey's *post hoc* tests for mean values

adjusted to maternal body size indicated that egg width and volume were similar between ESEC Taim and Arroio Grande, but smaller at Pelotas. Egg length in Arroio Grande was larger than other populations.

3.2 Relationship between maternal body size and reproductive traits

Maternal body size affected positively clutch size in all populations (Figure 4). The variation explained was approximately 47%, 29%, and 28% at Arroio Grande (GLM: $Z = 2.94$, $P = 0.003$), ESEC Taim (GLM: $Z = 8.91$, $P < 0.0001$), and Pelotas (GLM: $Z = 2.45$, $P = 0.01$), respectively. In contrast, maternal body size did not explain variation of any reproductive trait of egg size for the populations ESEC Taim and Pelotas ($P > 0.05$) (Table 2). However, we observed a linear increase between egg width ($F = 4.72$, $P = 0.03$, $df = 1, 45$) and volume ($F = 4.13$, $P = 0.04$, $df = 1, 45$) to maternal body size at Arroio Grande. Quadratic terms in the models did not produce better estimates than linear models in any of the relationships (Table 2).

3.3 Trade-off between egg size and clutch size

No trade-off was observed between egg size and number of eggs. Mixed models did not show any significant linear or quadratic relationship between the number of eggs and egg length, width or volume (Table 3).

4. Discussion

Our study of D'Orbigny's life history slider provides strong support to a variation on reproductive traits among populations at the same latitude range and at a small geographic scale. We also find that clutch size increase

with maternal phenotype while egg size has no relationship, except in Arroio Grande. None of the populations show decreasing egg size in large clutch size and the evidence of a trade-off is not observed in these populations.

It is plausible to argue that variations in the traits can occur due to changes in other factors at small scales, e.g., resource availability and other sources that could lead to a trade-off between reproduction and survival (Wallis et al., 1999, Iraeta et al., 2013, Zeng et al., 2013). In fact, the region comprising the studied populations has high productivity due to the complex environment of lakes connected to the ocean (Niencheski et al., 2007). Even close, the different populations exhibit different productivity levels, although this variation is quite small (Attisano et al., 2013). In a qualitative gradient, the region near Pelotas and the ESEC Taim has one of the highest productivities (Attisano et al., 2013) in relation to Arroio Grande. However, reproductive trait and maternal size from Pelotas are smaller and in the another end, all measured traits are larger in the ESEC Taim.

Demographic effects associated with small local variations in resource availability may provide insights into how populations might be varying (Congdon and van Loben Sels, 1991). Reproductive traits and maternal body size might be larger in ESEC Taim because of lower population density found in this area compared to, for example, Arroio Grande (P.S.L unpublished results). In fact, in conditions such as Arroio Grande a faster juvenile growth rate could be expected (Rowe, 1994a) and we believe that this happens for two main reasons. First, juvenile growth rates could vary due to density if resources are a limiting source in the same

place. In populations with high densities, the amount of resources directed to each individual would be lower or of poorer quality (Rowe 1994a) and the further growth could be compromised. Even the ESEC Taim area with high resource availability, the population faces an unquantified impact from agricultural activities, mainly through the introduction of agrochemicals on irrigated rice cultivation affecting nesting sites (Bager and Rosado, 2010). If environmental contaminants reduce habitat quality and affect reproductive traits making the offspring survival dubious (Alford, 2010), there may be a faster growth in the juvenile stage in this population. Thus, experimental studies assessing the cumulative effects of toxins of this species' eggs in these circumstances, and subsequent evaluation of the survival of offspring could help to better understand this effects.

Second, egg size variation may be associated to a differential intensity rate of egg mortality in early stages. Some studies have shown that egg size is highly correlated with hatchling size (Congdon et al., 1983, Janzen and Morjan, 2002, Warner et al., 2010). Thus, larger offspring (here larger eggs) may have faster juvenile growth rates, which is important to avoid predators (Brockeman, 1975). Faster growth rates may decrease exposure time during this stage when mortality is high (Congdon et al., 1994, Spencer and Janzen, 2010). For example, Gonçalves et al. (2007) observed high predation rates of D'Orbigny's slider nests at ESEC Taim, where egg size was larger than any other population. Of the nests monitored in ESEC Taim, 98% were depredated within the 48 hours after nesting. Nests of the same species monitored by Fagundes et al. (2010) in Pelotas, where we observed the smallest egg sizes, showed about 82% of survival.

Therefore, larger egg size might be required during high juvenile or early stages mortality in environments where predation pressure is high (Reznick et al., 2001).

After removing the maternal size effect from all analyses related to geographic variation, we still observed changes in traits related to egg size, but not in clutch size. The maternal body cavity considerably influences the number of eggs that a female can carry (Rowe, 1994a, Daza and Paez, 2007, Lin et al., 2012, Naimi et al., 2012, Macíp-Rios et al., 2013) and thus explains much of the variation in offspring size as supported by the linear relationships (Figure 4).

Maternal body size seems to have a minor effect on egg size. Although the relationship tended to increase, the slope was not significantly different from zero in ESEC Taim and Pelotas. This is in agreement with previous models (Smith and Fretwell, 1974) in which egg size was invariant with maternal phenotype within populations, in this way resulting in a single investment per offspring optimum for all parents. However, in Arroio Grande, we observed that egg width and volume were related to maternal body size. This pattern is often found in many turtle species (Congdon and Gibbons, 1987, Rollinson and Brooks, 2008, Lovich et al., 2012, Macíp-Rios et al., 2013) and since the early model developed, there are many lines of investigation that try to explain this incongruence (Parker and Begon, 1986, Congdon and Gibbons, 1987, McGinley, 1989, Sakai and Harada, 2001, Filin, 2015). We could infer here that, in Arroio Grande, females with larger maternal resources (as maternal resources often increase with body size) should produce larger offspring size even when clutch size is high (McGinley, 1989). This could be favored when large

clutch size also increase offspring survival. According to McGinley (1989), this could be the case when hatchlings experience high predation risk and offspring from larger clutches could have better chance during initial dispersion. To our knowledge, no study provides estimates of nest and hatchling predation in this population. However, illegal harvesting of this species in Rio Grande do Sul may exceed 300,000 hatchlings/year of individuals taken from nature (Bager and Rosado, 2010), and Arroio Grande is one of the most impacted areas by this threat (A. Bager pers. observ.). We believe this is a potential explanation to increased egg size with larger clutch size and maternal phenotype.

In the three populations, the expected negative relationship between clutch size and egg size was not observed. The existence of trade-offs is a central concept in evolutionary biology (Lloyd, 1987, Stearns, 2000, Uller and Olson, 2005, Rollinson and Rowe, 2015). Despite this, the trade-off would not be expected if resource availability and reproductive allocation by females vary greatly among individuals or species (Uller and Olson, 2005, Warne and Charnov, 2008). Experimental studies have showed that with unlimited food availability females could produce more and larger hatchlings (Stahlschmidt et al., 2014). As all the three populations are located in a region with high productivity potential (see above), it is not hard to think how females can invest in clutch size and offspring fitness.

In summary, our comparative study shows variation in life history attributes in nearby populations of D'Orbigny's slider. Several factors may play important roles in interpopulation variation, and this could be, for instance, the results of phenotypic plasticity of the species in response to environmental differences (Zeng et al., 2013) and pressures coupled with

population dynamics. Some of the predictions tested are in accordance with recent studies on optimization models in turtles (Lovich et al., 2012); however, others were not supported in specific populations. This study highlight how interactions between morphological and reproductive attributes add complex variations in clutch and egg size and to what degree egg size is optimized, or not in different species or populations

Acknowledgements

Field support, both logistical and financial, was provided by Brazilian Agency CNPq for A. Bager. PSL was supported by a PhD grant from Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (Capes) in Brazil. Suggestions from Murilo Dantas de Miranda and Clara Grilo were very helpful.

References

- Alford, R.A., 2010. Declines and the global status of amphibians, In: Sparlling, D.W., Linder, G., Bishop, C.A., Krest, S. (Eds.), *Ecotoxicology of reptiles and amphibians*. SETAC Press, Pensacola, Florida, pp.13–45.
- Ashton, K.G., Burke, R.L., Layne, J.N., 2007. Geographic Variation in Body and Clutch Size of Gopher Tortoises. *Copeia* 2007, 355–363. [http://dx.doi.org/10.1643/0045-8511\(2007\)7\[355:GVIBAC\]2.0.CO;2](http://dx.doi.org/10.1643/0045-8511(2007)7[355:GVIBAC]2.0.CO;2).
- Attisano, K.K., Santos, I.R., Andrade, C.F.F.D., Paiva, M.L.D., Milani, I.C.B., Niencheski, L.F.H., 2013. Submarine groundwater discharge revealed by radium isotopes (Ra-223 and Ra-224) near a paleochannel on the Southern Brazilian continental shelf. *Braz. J. Oceanogr.* 61, 195–200. <http://dx.doi.org/10.1590/S1679-87592013000300004>

- Bager, A., 2003. Aspectos da biologia e ecologia da tartaruga Tigre D'Água, *Trachemys dorbignyi* (Testudines – Emydidae) no extremo sul do Estado do Rio Grande do Sul – Brasil. In: Ph.D. Thesis, Universidade Federal do Rio Grande do Sul.
- Bager, A., Freitas, T.R.O., Krause, L., 2007. Nesting ecology of a population of *Trachemys dorbignyi* (Emydidae) in southern Brazil. *Herpetologica* 63, 56–65. [http://dx.doi.org/10.1655/0018-0831\(2007\)63\[56:NEOAPO\]2.0.CO;2](http://dx.doi.org/10.1655/0018-0831(2007)63[56:NEOAPO]2.0.CO;2)
- Bager, A., Rosado, J., 2010. Estimation of core terrestrial habitats for freshwater turtles in Southern Brazil based on nesting areas. *J. Herpetol.* 44, 658–662. <http://dx.doi.org/10.1670/09-036.1>.
- Bager, A., Freitas, T.R.O., Krause, L., 2010. Morphological characterization of adults of Orbigny's slider *Trachemys dorbignyi* (Duméril & Bibron 1835) (Testudines Emydidae) in southern Brazil. *Trop. Zool.* 23, 181–194.
- Bager, A., Krause, L., Freitas, T. R.O., 2012. Fidelity to nesting sites and orientation of *Trachemys dorbignyi* (Duméril & Bibron, 1835) (Testudines: Emydidae) female in Southern Brazil. *Trop. Zool.* 25, 31–38. [DOI:10.1080/03946975.2012.679393](https://doi.org/10.1080/03946975.2012.679393)
- Bleu, J., Gamelon, M., Sæther, B.E., 2016. Reproductive costs in terrestrial male vertebrates: insights from bird studies. *Proc. R. Soc. B.* 283, 20152600. [DOI: 10.1098/rspb.2015.2600](https://doi.org/10.1098/rspb.2015.2600)
- Brockelman, W.Y. 1975. Competition and fitness of offspring, and optimal clutch size. *Am. Nat.* 109, 677–699. [DOI: 10.1086/283037](https://doi.org/10.1086/283037)
- Bujes, C.S., Verrastro, L., 2008. Chelonians from the Delta of Jacuí River, RS, Brazil: habitats use and conservation. *Nat. Conserv.* 6, 157–170.
- Burnham, K.P., Anderson, D.R., 2002. *Model Selection and Multimodel Inference: A Practical Information-Theoretic Approach*. Springer-Verlag.

- Cagle, F. 1939. A system of marking turtles for future identification. *Copeia*. 1939, 170–173. DOI: [10.2307/1436818](https://doi.org/10.2307/1436818)
- Congdon, J.D., Gibbons, J.W., 1987. Morphological constraint on egg size: a challenge to optimal egg size theory? *PNAS*. 84, 4145–4147.
- Congdon, J.D., Loben Sels, R.C.V., 1991. Growth and body size in Blanding's turtles (*Emydoidea blandingi*): relationships to reproduction. *Can. J. Zool.* 69, 239-245.
- Congdon, J.D., Dunham, A.E., van Loben Sels, R.C., 1994. Demographics of common snapping turtles (*Chelydra serpentina*): Implications for conservation and management of long-lived organisms. *Am. Zool.* 34, 397–408.
- Daza, J.M., Paez, V.P., 2007. Morphometric variation and its effect on reproductive potential in female Colombian slider turtles. *Herpetologica* 63,125–134. [http://dx.doi.org/10.1655/0018-0831\(2007\)63\[125:MVAIEO\]2.0.CO;2](http://dx.doi.org/10.1655/0018-0831(2007)63[125:MVAIEO]2.0.CO;2).
- Fagundes, C.K., Bager, A., Cechin, S.T.Z., 2010. *Trachemys dorbigni* in an anthropic environment in southern Brazil: II) Reproductive ecology. *Herpetol. J.* 20, 195–199.
- Filin, I., 2015. The relation between maternal phenotype and offspring size, explained by overhead material costs of reproduction. *J. Theor. Biol.* 364, 168–178. doi:10.1016/j.jtbi.2014.09.007
- Gonçalves, F.A., Cechin, S.Z., Bager, A., 2007. Predação de ninhos de *Trachemys dorbigni* (Duméril and Bibron, 1835) (Testudines, Emydidae), no extremo sul do Brasil. *Rev. Bras. Biol.* 24, 1063–1070. <http://dx.doi.org/10.1590/S0101-81752007000400025>.
- Hahn, A.T., Rosa, C.A., Bager, A., Krause, L., 2014. Dietary variation and overlap in D'Orbigny's slider turtles *Trachemys dorbigni* (Duméril and Bibron 1835) (Testudines: Emydidae). *J. Nat. Hist.* 48, 721–728. <http://dx.doi.org/10.1080/00222933.2013.840400>

- Iraeta, P., Salvador, A., Díaz, J.A., 2013. Life-history traits of two Mediterranean lizard populations: a possible example of countergradient covariation. *Oecologia*. 172, 167–176. [doi: 10.1007/s00442-012-2492-8](https://doi.org/10.1007/s00442-012-2492-8)
- Iverson, J.B., Smith, G.R., 1993. Reproductive ecology of the painted turtles (*Chrysemys picta*) in the Nebraska Sandhills and across its range. *Copeia* 1993, 1–21. doi: [10.2307/1446291](https://doi.org/10.2307/1446291)
- Iverson, J.B., Higgins, H., Sirulnik, A., Griffiths, C., 1997. Local and geographic variation in the reproductive biology of the snapping turtle (*Chelydra serpentina*). *Herpetologica* 53, 96–117.
- Janzen, F.J., Morjan, C.L., 2002. Egg size, incubation temperature, and posthatching growth in painted turtles (*Chrysemys picta*). *J. Herpetol.* 36, 308–311.
- Lin, L.H., Mao, F., Chen, C., Ji, X., 2012. Reproductive traits of the gray ratsnake *Ptyas korros* from three geographically distinct populations. *Curr. Zool.* 58, 820–827.
- Litzgus, J.D., Mousseau, T.A., 2006. Geographic variation in reproduction in a freshwater turtle (*Clemmys guttata*). *Herpetologica* 62, 132–140. <http://dx.doi.org/10.1655/05-26.1>.
- Lovich, J.E., Madrak, S.V., Drost, C.A., Monatesti, A.J., Casper, D., Znari, M., 2012. Optimal egg size in a suboptimal environment: reproductive ecology of female Sonora mud turtles (*Kinosternon sonoriense*) in central Arizona, USA. *Amphib. Reptil.* 33, 161–170. [DOI:10.1163/156853812X634035](https://doi.org/10.1163/156853812X634035)
- Lloyd, D.G., 1987. Selection of offspring size and other size-versus-number strategies. *Am. Nat.* 129, 800–817.
- Macip-Ríos, R., Brauer-Robleda, P., Casas-Andreu, G., Arias-Cisneros, M.L., Sustaita-Rodríguez, V.H., 2012. Evidence for the morphological constraint hypothesis and optimal offspring size theory in the Mexican

- mud turtle (*Kinosternon integrum*). Zool. Sci. 29, 60–65. <http://dx.doi.org/10.2108/zsj.29.60>
- Macip-Ríos, R., Casas-Andreu, G., Sustaita-Rodríguez, V.H., 2013. Evidence of Pelvic and Nonpelvic Constraint on Egg Size in Two Species of *Kinosternon* from Mexico. Chelolian Conserv. Biol. 12, 218–226. <http://dx.doi.org/10.2744/CCB-1038.1>
- Matzenauer, R., Radin, B., and Almeida, I.R., 2011. Atlas Climático do Rio Grande do Sul. Cemet RS, Brasil.
- McGinley, M.A., 1898. The influence of a positive correlation between clutch size and offspring fitness on the optimal offspring size. Evol. Ecol. 3, 150–156. [doi:10.1007/BF02270917](https://doi.org/10.1007/BF02270917)
- Naimi, M., Znari, M., Lovich, J.E., Feddadi, Y., and Ait Baamrane, M.A., 2012. Clutch and egg allometry of the turtle *Mauremys leprosa* (Chelonia: Geoemydidae) from a polluted peri-urban river in west-central Morocco. Herpetol. J. 22, 43–49.
- Niencheski, L.F., Windom, H.L., Moore, W.S., Jahnke, R.A., 2007. Submarine groundwater discharge of nutrients to the ocean along a coastal lagoon barrier, southern Brazil. Mar. Chem. 106, 546–561. [doi:10.1016/j.marchem.2007.06.004](https://doi.org/10.1016/j.marchem.2007.06.004)
- Parker, G.A., Begon, M.B., 1986. Optimal egg size and clutch size: effects of environment and maternal phenotype. Am. Nat. 128, 573–592.
- Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., 2016. *nlme: Linear and Nonlinear Mixed Effects Models*. R package version 3, 1–126.
- R Core Team (2015). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL <https://www.R-project.org/>
- Reznick, D., Butler IV, M. J., Rodd, H., 2001. Life-history evolution in guppies. VII. The comparative ecology of high-and low-predation environments. Am. Nat. 157, 126–140. [doi: 10.1086/318627](https://doi.org/10.1086/318627)

- Roff, D.A., 1992. The evolution of life histories: theories and analysis. Chapman & Hall, New York.
- Rollinson, N., Brooks, R.J., 2008. Optimal offspring provisioning when egg size is “constrained”: A case study with the painted turtle *Chrysemys picta*. *Oikos* 117, 144–151. [10.1111/j.2007.0030-1299.16088.x](https://doi.org/10.1111/j.2007.0030-1299.16088.x).
- Rollinson, N., Rowe, L., 2015. The positive correlation between maternal size and offspring size: fitting pieces of a life-history puzzle. *Biol. Rev.* 2015, 1–15. [doi: 10.1111/brv.12214](https://doi.org/10.1111/brv.12214)
- Rowe, J.W., 1994a. Reproductive variation and the egg size-clutch size trade-off within and among populations of painted turtles (*Chrysemys picta bellii*). *Oecologia* 99, 35–44.
- Rowe, J.W., 1994 b. Egg size and shape variation within and among Nebraskan painted turtle (*Chrysemys picta bellii*) populations: relationships to clutch and maternal body size. *Copeia* 1994, 1034–1040.
- Rowe, J.W., Coval, K.A., Campbell, K.C., 2003. Reproductive characteristics of female midland painted turtles (*Chrysemys picta marginata*) from a population on Beaver Island, Michigan. *Copeia* 2003, 326–336.
- Sakai, S., Harada, Y., 2001. Why do large mothers produce large offspring? Theory and a test. *Am. Nat.* 157, 348–359. [DOI: 10.1086/319194](https://doi.org/10.1086/319194)
- Salguero-Gómez, R., Jones, O.R., Jongejans, E., Blomberg, S.P., Hodgson, D.J., Mbeau-Ache, C., Buckley, Y.M., 2016. Fast–slow continuum and reproductive strategies structure plant life-history variation worldwide. *Proc. R. Soc. B.* 113, 230–235. [DOI: 10.1086/319194](https://doi.org/10.1086/319194)
- Smith, C.C., Fretwell, S.D., 1974: The optimal balance between size and number of offspring. *Am. Nat.* 108, 499–506.

- Spencer, R.J., Janzen, F.J., 2010. Demographic consequences of adaptive growth and the ramifications for conservation of long-lived organisms. *Biol. Conserv.* 143, 1951–1959. [doi:10.1016/j.biocon.2010.04.034](https://doi.org/10.1016/j.biocon.2010.04.034)
- Stahlschmidt, Z., O’Leary, M.E., Adamo, S., 2014. Food limitation leads to risky decision making and to tradeoffs with oviposition. *Behav. Ecol.* 25, 223–227. [doi: 10.1093/beheco/art110](https://doi.org/10.1093/beheco/art110)
- Stearns, S.C., 2000. Life history evolution: successes, limitations, and prospects. *Naturwissenschaften* 87, 476–486.
- Uller, T., Olsson, M., 2005. Trade-offs between offspring size and number in the lizard *Lacerta vivipara*: a comparison between field and laboratory conditions. *J. Zool.* 265, 295–299. [10.1017/S0952836904006326](https://doi.org/10.1017/S0952836904006326)
- Valenzuela, N., 2001. Maternal effects on life history traits in the Amazonian giant river turtle *Podocnemis expansa*. *J. Herpetol.* 35, 368–378.
- Wallis, I.R., Henen, B.T., Nagy, K.A., 1999. Egg size and annual egg production by female desert tortoises (*Gopherus agassizii*): the importance of food abundance, body size, and date of egg shelling. *J. Herpetol.* 33, 394–408.
- Warne, R.W., Charnov, E.L., 2008. Reproductive allometry and the size-number trade-off for lizards. *Am. Nat.* 172, 80–98. [doi: 10.1086/589880](https://doi.org/10.1086/589880)
- Warner, D.A., Jorgensen, C.F., Janzen, F.J., 2010. Maternal and abiotic effects on egg mortality and offspring size of turtles: temporal variation in selection over seven years. *Funct. Ecol.* 24, 857–866. [doi: 10.1111/j.1365-2435.2010.01714.x](https://doi.org/10.1111/j.1365-2435.2010.01714.x)
- Wilkinson, L.R., Gibbons, J.W., 2005. Patterns of Reproductive Allocation: Clutch and Egg Size Variation in Three Freshwater Turtles. *Copeia* 2005, 868–879.

Zeng, Z.G., Zhao, J.M., Sun, B.J., 2013. Life history variation among geographically close populations of the toad-headed lizard (*Phrynocephalus przewalskii*): Exploring environmental and physiological associations. *Acta Oecol.* 51, 28–33. [doi:10.1016/j.actao.2013.05.004](https://doi.org/10.1016/j.actao.2013.05.004)

Zuur, A.F., Ieno, E.N., Smith, G.M., 2007. *Analyzing ecological data*. Springer, New York.

List of Tables

Table 1: Variation of female size and reproductive traits of D'Orbigny's slider populations in Brazil. X^2 values of Kruskal-Wallis tests.

Parameters/ Populations	Arroio Grande	Pelotas	ESEC Taim	Test
Carapace length (mm)	210.08 ± 16.52 (158.00 - 248.00) n = 245	204.70 ± 10.00 (192.00 - 229.00) n = 34	233.66 ± 13.72 (191.00 - 277.00) n = 127	$x^2=154.04$, $P<0.0001$
Egg length (mm)	39.64 ± 1.63 (35.68 - 43.97) n = 55	37.44 ± 2.25 (32.54 - 42.86) n = 89	39.05 ± 1.29 (35.88 - 42.30) n = 83	$x^2=43.09$, $P<0.0001$
Egg width (mm)	24.74 ± 1.03 (22.28 - 26.53) n = 55	20.10 ± 1.73 (16.6 - 24.3) n = 88	25.84 ± 10.88 (23.99 - 27.85) n = 83	$x^2=167.00$, $P<0.0001$
Egg volume(cm ³)	13.36 ± 1.42 (9.25 - 16.01) n = 55	8.45 ± 1.74 (5.87 - 13.79) n = 88	14.40 ± 1.22 (11.60 - 16.76) n = 83	$x^2=157.64$, $P<0.0001$
Clutch volume (cm ³)	108.84 ± 36.90 (24.33 - 179.64) n = 55	69.32 ± 24.42 (27.33 -144.69) n = 88	174.02 ± 39.43 (81.65 -292.25) n = 83	$x^2=148.13$, $P<0.0001$
Clutch size	9.23 ± 2.68 (3 - 17) n = 357	8.20 ± 2.16 (4 - 15) n = 89	11.86 ± 2.37 (6 - 19) n = 125	$x^2=110.42$, $P<0.0001$

Table 2: Linear and quadratic models fitted for three D'Orbigny's slider populations. a, b and c are parameters estimates. Indications of parameters estimates for linear models are followed by _(L) and for quadratic models by _(q). * $P < 0.05$. EW = egg width, EL = egg length and EV = egg volume.

Population	Traits	a_L	b_L	R_L^2	$AIC_{(L)}$	a_q	b_q	c_q^2	R_q^2	$AIC_{(q)}$	ΔAIC
Arroio Grande (n=47)	EW	0.018	20.841	0.074*	135.30	43.864	-0.2029	0.00053	0.079	137.40	2.10
	EL	0.022	34.941	0.037	178.07	36.059	0.0129	0.00003	0.016	180.09	2.02
	EV	0.024	8.173	0.060*	168.17	33.291	-0.2170	0.00057	0.057	170.42	2.25
ESEC Taim (n=45)	EW	0.004	24.687	0.010	120.13	9.2909	0.1356	-0.00027	0.029	122.65	2.52
	EL	0.010	36.788	0.002	138.85	46.379	-0.0714	0.00017	0.023	141.73	2.87
	EV	0.009	12.227	0.007	145.08	2.1959	0.0945	-0.00018	0.029	147.96	2.88
Pelotas (n=34)	EW	0.035	12.939	0.002	146.44	207.82	2.1550	-0.00510	0.031	148.50	2.06
	EL	0.038	30.486	0.007	147.00	116.63	-0.0788	0.00198	0.015	149.76	2.76
	EV	0.041	0.373	0.009	149.76	172.70	1.7030	-0.00398	0.010	152.63	2.87

Table 3: Response of clutch size to egg size to test for a possible trade-off for the three population of D'Orbigny's slider in Brazil.

Trait	Random	Fixed		
	<i>Intercept</i>	β	<i>SE</i>	<i>p-value</i>
Population	2.40			
Egg length		0.00036	0.144	0.99
Population	2.22			
Egg width		0.1161	0.163	0.48
Population	2.26			
Egg volume		0.0853	0.139	0.54

List of Figures

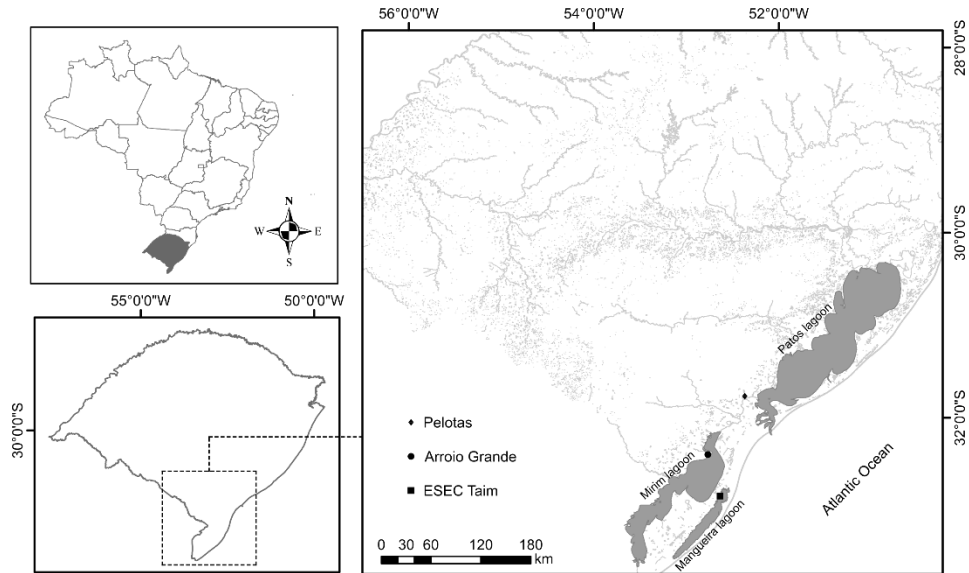


Figure 1: Location of the three populations of D'Orbigny's slider sampled in South of Brazil.

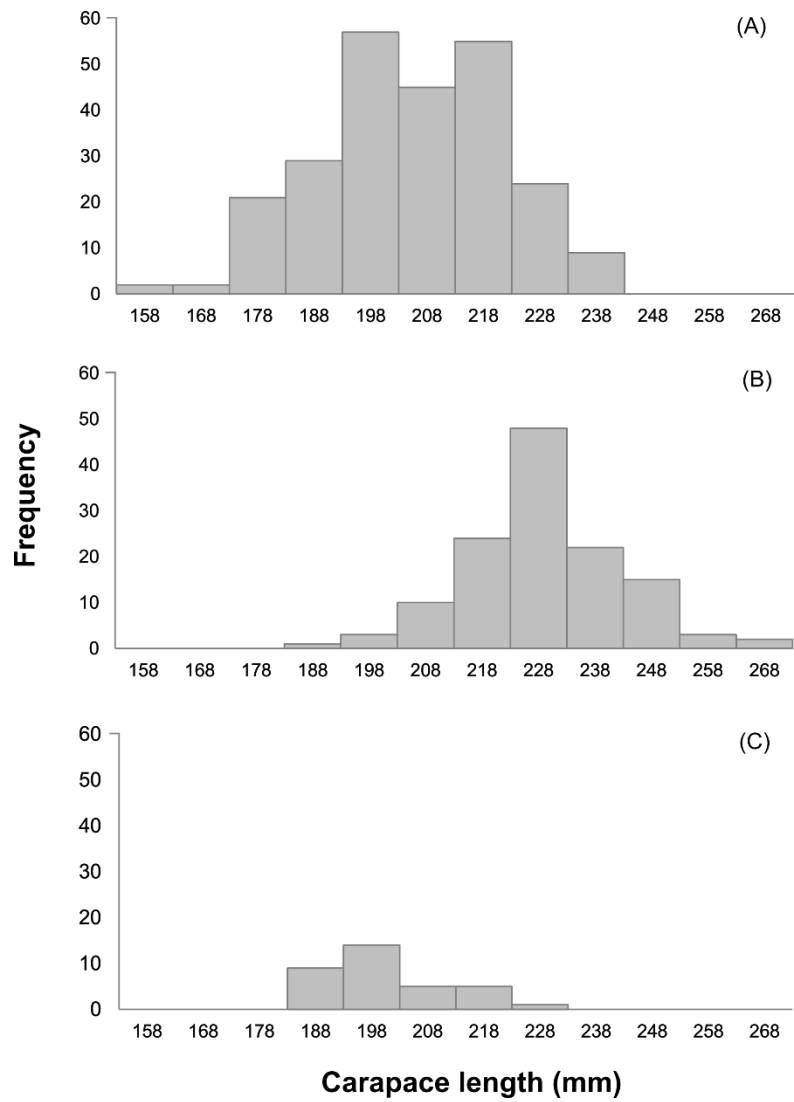


Figure 2: Carapace length frequency histograms of reproductive females. (A) Arroio Grande; (B) ESEC Taim; (C) Pelotas.

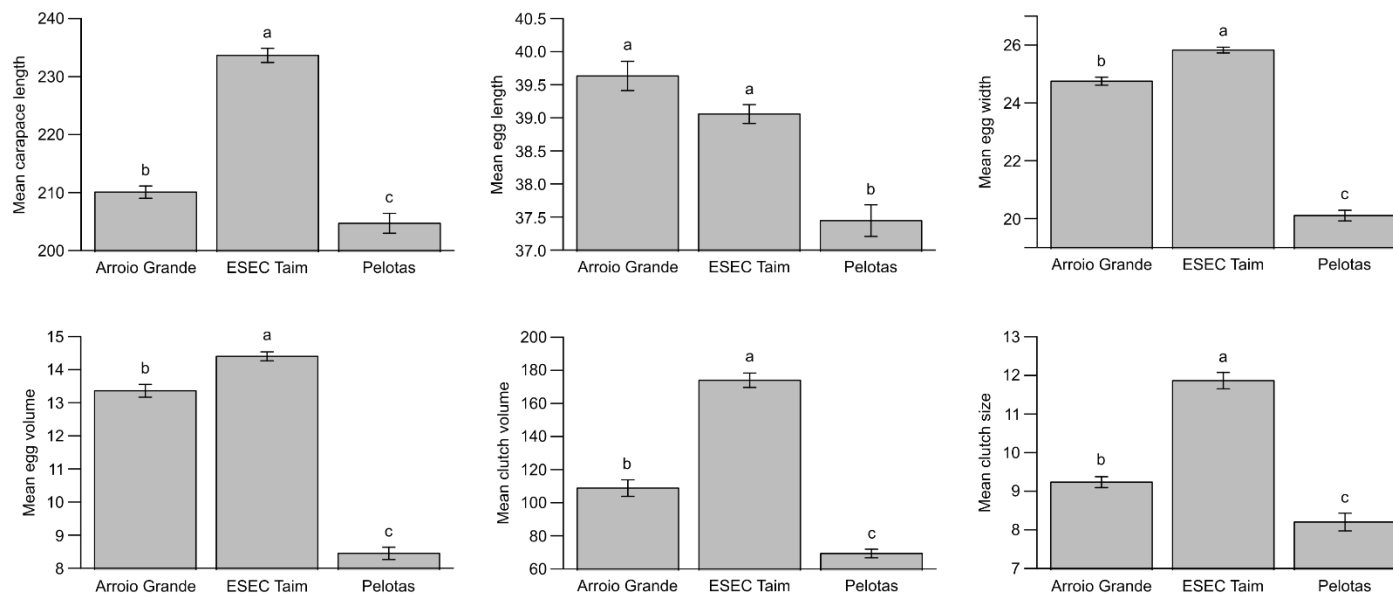


Figure 3: Maternal body size and reproductive traits of D'Orbigny's slider measured in three populations. Means with different letters were significantly different based on the Bonferroni adjusted P -value. The error bars represent 95% confidence limits.

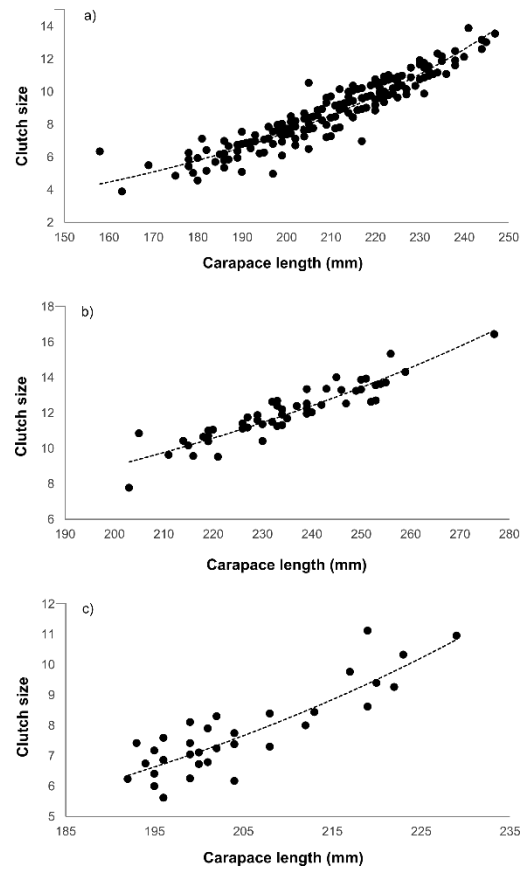


Figure 4: Linear relationship between clutch size and maternal body size at the three populations of D'Orbigny's slider. A) The linear model in Arroio Grande is described by $Y = \exp^{(0.012x - 0.508)}$. B) In ESEC Taim by $Y = \exp^{(0.007x + 0.651)}$. C) In Pelotas by $Y = \exp^{(0.0145x - 0.931)}$.

ARTIGO 2

Consequência das ameaças à viabilidade de quelônios em zonas úmidas subtropicais.

Preparado de acordo com as normas da Revista Conservation Biology.

Lucas, P.S., Miranda, M.D., Bager, A. (2016) Consequence of threats to freshwater turtles viability in subtropical wetlands.

Priscila Silva Lucas^{1,2*}, Murilo Dantas de Miranda^{3,4} & Alex Bager^{1,2}

¹Programa de Pós-Graduação em Ecologia Aplicada, Departamento de Biologia, Universidade Federal de Lavras, Campus Universitário, CP 3037, Lavras, CEP 37200 000. Brazil. Tel.: +55 35 3829 1924.

²Centro Brasileiro de Estudos em Ecologia de Estradas, Universidade Federal de Lavras, Campus Universitário, CP 3037, Lavras, CEP 37200 000. Brazil. Tel.: +55 35 3829 3129.

³Institute of Biology, Martin Luther University Halle Wittenberg, Am Kirchtor 1, 06108 Halle (Saale), Germany.

⁴German Centre for Integrative Biodiversity Research (iDiv) Halle-Jena-Leipzig, Deutscher Platz 5e, 04103 Leipzig, Germany.

*Corresponding author: Priscila da Silva Lucas: Tel.: +55 35 3829 3129,

Fax: +55 35 3829 1924; E-mail: prilucass@gmail.com

Abstract: Many threats are causing loss of biodiversity and population declines. Anthropogenic alterations such as man made structures (e.g. roads) and illegal egg poaching are serious pressures to long-lived turtles. We used data on mark-recapture, mortality on roads, nest predation and egg harvest to estimate the impact of these threats to D'Orbigny's slider turtle *Trachemys dorbigni* on the ESEC Taim. We used a stage-based deterministic matrix to infer population growth rates responses to alternative scenarios which threats were included and to indicate management options. Road mortality was estimated to account for 1 to 2% of adult lost. Nest predation ranged from 18% to 98% and egg poaching ranged 25 to 90%. The baseline projection showed that the population was declining ($\lambda = 0.989$), and road mortality has a higher potential even with low rates to contribute to a faster decline. Elasticity was higher for adult survival and the population could attain a stable population by i) increasing both survival in the early stages or ii) increasing female survival through reducing road mortality. A range of management strategies is available for turtles (nest protection, headstarting and road crossing structures) in order to decrease mortality or control egg poaching. However, we reiterate that strategic actions for this species ought to include protection in all stages to prevent the continued decline of this population.

Keywords: *Trachemys dorbigni*, stage-based model, road mortality, nest pressure, egg poaching.

Introduction

There is a growing evidence of negative impacts of different threats to biodiversity in this century (Pereira et al. 2012; Newbold et al. 2015). Potentially, among 5 - 9 millions of animal species are currently living, however the majority has not been described or will be lost before that (Costello et al. 2013). In a broader perspective, analytical approaches focused on assessments of various species show extremely high extinction rates (Chapin III et al. 2000; Pereira et al. 2010; Pimm et al. 2014), considerably higher than rates estimated for the last century (Pimm et al. 2014) or even before (Pimm et al. 1995).

Habitat loss and degradation stand out as one of the most important drivers (Pereira et al. 2010; 2012) induced mainly by the change and reduction of land and water habitats (Gibbons et al. 2000). By the end of the century, projections show extinction rates of approximately 18% due to deforestation in areas of tropical forest hotspots, even if all the remaining habitats within these hotspots are preserved (Pimm & Raven 2000). Roads and other linear infrastructures are one of the greatest landscape modifiers, especially in tropical regions (Laurance et al. 2009). Direct and indirect mechanisms (Forman & Alexander 1998) lead to biodiversity loss since the road segment scale to broader scales (Forman & Deblinger 2000; Beaudry et al. 2008; Jaeger 2015). Road mortality as one of the main effects has been documented in high rates for many taxa (Smith & Dood-Jr 2003; Bager & Fontoura 2013; and reviewed by D'Anunção et al. 2013). This is especially true for those species that make extensive overland movements and do not avoid or are attracted to roads (Aresco 2005, Fahrig

& Rytwinski 2009; Jacobson et al. 2016). Although, despite the growing number of road kills, understand how this loss affects the population viability of most species should be priority to successfully manage efforts to stabilize or recover declining populations. Indeed, only recent studies show that even low roadkill rates affecting target ages or stages is enough to cause severe declines in snakes and turtles (Row et al. 2007; Crawford et al. 2014) and birds species (Borda-de-Água et al. 2014).

Illegal wildlife trade is another driver of biodiversity loss (Fernandes-Ferreira et al. 2012; Baker et al. 2013). Unsustainable trade has been recognized as a challenge to wildlife conservation (Nijman and Shepard 2015) and still a common practice in South America (Alves et al. 2010; Baker et al. 2013) for many taxa (Bush et al. 2014; Jenkins et al. 2014). Particularly turtles, unsustainable harvest for the pet trade has been pointed out as one of the causes of population decline in the freshwater species *Erymnochelys madagascariensis* (Kuchling 1988) and on the tortoise species *Astrochelys radiata* that is projected to go extinct in 45 years (Leuteritz & Paquette 2008). Therefore, wherever individuals for pets are originated, it is wise to address the effects of removal from the wild on the viability of source populations (Bush et al. 2014).

Evaluating how closely species are to extinction is one of the most important first steps in preventing their extinction, which is a main goal of biodiversity conservation (Yeong Ryu et al. 2016). Although reptiles are among the taxa most often reported in live trade (Baker et al. 2013), the scientific community has comparatively neglected reptiles for conservation assessment. Only 35% of described species have been evaluated in the

IUCN Red List and a great percentage is classified as data deficient (Bohm et al. 2013).

One of the advantages of using mechanistic population models is that we can link specific anthropogenic threats to population declines (Heppel 1998; Crawford et al. 2014, Tuma et al. 2016). Moreover, as populations are facing many threats as stated above, addressing competing threats is crucial to rank their severity to cause population decline (Tuma et al. 2016) and to report the best management practice (Crouse et al. 1987; Heppel et al. 1996). Here, through studying D'Orbigny slider turtles *Trachemys dorbigni* we aimed 1) to determine and compare population responses from the estimated rates of road mortality, egg poaching for pet trade and nest predation in different scenarios; 2) to make conservation recommendations that will stabilize or potentially grow this population. We expected that adult road mortality would more negatively affect population growth rate even with a lower mortality rate compared to other threats.

Material and Methods

Study species

Trachemys dorbigni (D'Orbigny's sliderturtle) occurs in wetlands mainly streams, marshes and swamps in Uruguay, Argentina and in the south of Brazil (Bager et al. 2007; Fagundes et al. 2010a; Bujes et al. 2011). Its natural distribution in Brazil comprises only 0.2% of the total area of the country. The species is abundant in many places across its distribution range and shows similar demographic features like other turtle species:

usual high mortality in the first stages, low adult mortality, delayed maturation and longevity (Gonçalves et al. 2007; Fagundes et al. 2010b; Lucas & Bager 2016b). Many populations of this species are under human pressure such as intensive egg poaching for illegal pet trade, environmental pollution, agricultural activities near wetlands and collisions with vehicles (Bager & Rosado 2010; Fagundes et al. 2010a; Costa et al. 2015) at levels that can potentially cause population declines. Despite that, the species was only recently categorized as Near Threatened in a regional assessment (Vogt et al. 2015) besides not even being listed by IUCN. In Argentina, D'Orbigny's slider turtle has been initially included in a category called “commercially threatened” due to illegal capture for pet trade and the advance of the agriculture on the few known populations (Bertonatti 1994). Nowadays it is categorized as vulnerable (Prado et al. 2012). In Uruguay, the species has not been listed to any particular status.

Study population

We had been studied some populations intensively between 1995 - 2006 in the extreme south of Rio Grande do Sul state, from Pelotas to Taim Ecological Station (further refereed only as ESEC Taim), Brazil. ESEC Taim is a federal protected area located on the coastal plain and has an area of 33.818 ha. The relief is low and flat, and wetlands are conspicuous, although open fields and dunes are also present (Bager & Fontoura 2013). Within this protected area, we studied D'Orbigny's slider female turtles during 11 years using capture-mark-recapture techniques. Turtles were marked annually during the nest season (October – January) (Lucas & Bager 2016a; b) in an area of about 200 ha. This area is relatively

undisturbed (except by high nest depredation) (Gonçalves et al. 2007). However, surrounding the species suffers with agriculture that may cause impacts due to the introduction of agrochemicals in the nesting sites (Bager & Rosado 2010), nest exploitation for pet trade (A. Bager pers. obs.) and an elevated female road mortality (Bager et al. 2007; Bager & Fontoura 2013). D'Orbigny's slider turtle is one of the most road killed reptile species in this road (Bager & Fontoura 2013; Costa et al. 2015).

Estimating threats

In order to estimate road mortality for D'Orbigny's slider turtles, we used a dataset on road kills from a monitoring performed along 137 km in two Brazilian federal highways (BR-392 and BR-471). These roads connect the cities of Pelotas (31°48'19''S and 52°19' 39''W) and Santa Vitória do Palmar (32°40'35''S and 52°35'42''W). BR-471 and crosses about 15 km within the ESEC Taim area, which receives an average of 1.100 vehicles per day (Rosa & Bager 2012; Bager & Fontoura 2013; Costa et al. 2015). We used the number of D'Orbigny's slider turtle road kills within a road stretch of 14 km monitored to estimate a per capita road mortality. This rate was estimated in 0.012 (see Supporting Information for calculations).

For nest predation, we used known estimates of mortality as stated in Table 1. Other sources of egg mortality as egg infertility, embryonic mortality and developmental abnormalities that also cause egg failure (Peters et al. 1994) were not taken in account in our estimates due to the difficulty of obtaining accurate estimates. Therefore, the estimated

proportion of non-depredated nests to total nests is an approximate estimate of egg survival (Crawford et al. 2014).

Finally, egg poaching for pet trade is a widespread activity but the estimates are unknown. Illegal harvesting of D'Orbigny's slider turtle in Rio Grande do Sul may exceed 300.000 eggs/hatchlings per year (Bager & Rosado, 2010). To our knowledge, only Fagundes et al. (2007a) provided some estimate of this threat. They observed high egg harvest (~ 90%) through a rate of nests lost by local people exploration.

Population modelling: the matrix model

We constructed a female stage-classified deterministic matrix (Lefkovitch 1965; Caswell 1989; Crouse et al. 1987; Enneson & Litzgus 2008) based on published demographic data (Table 1) to estimate population growth rate (λ):

$$n(t+1) = An(t)$$

where $n(t)$ is a vector of stage-specific abundances at time t and A is the population projection matrix describing the flux of individuals from one year to the next:

$$A = \begin{pmatrix} 0 & 0 & F \\ P_{21} & P_{22} & 0 \\ 0 & P_{32} & P_{33} \end{pmatrix}$$

Our model distributes the D'Orbigny's slider turtle life cycle into three stages. The model comprises parameters such as the probability of surviving and remaining in the same stage (P_i) (Crouse et al. 1987):

$$P_i = \left(\frac{1 - \varphi_i^{d_i-1}}{1 - \varphi_i^{d_i}} \right)$$

The transition probabilities (P_{ij}) from one stage to the next between times i and $i + 1$ (G_i), if turtle survives (Crouse et al. 1987; Fordham et al. 2008):

$$G_i = \frac{\varphi_i^{d_i}(1 - \varphi_i)}{1 - \varphi_i^{d_i}}$$

where φ is annual survival and d_i is the number of years from stage i to $i + 1$.

Both probabilities plus fecundity (F) were used to estimate population growth rate (λ) from the dominant eigenvalue of the matrix A (Caswell 1989). Stage 1 comprises egg/hatchling that begins when eggs are laid in the middle of spring (October) and ends in the next October (lasts one year). Stage 2 is the juvenile stage that lasts 3 years (ages 2 to 4) and stage 3 comprises the adults combined into age 6 and greater. No maximum age was included in the model, so P_{33} corresponded to annual adult survival rate. We calculated fecundity by multiplying mean clutch size, mean clutch frequency and annual first stage survival (Crawford et al. 2014). This product was latter multiplied by the proportion of female offspring (assumed to be 0.5), since we are dealing with a female matrix model (Enneson & Litzgus 2008; Crawford et al. 2014).

Because in this population there is a high nest predation, we included a mean mortality rate based on other populations of D'Orbigny's slider to exclude this potential effect and infer how this increased pressure could affect other populations (A. Bager, unpubl. data). For the vector of stage-specific abundances at year t , we used population size estimates ($N = 1588$ individuals) for ESEC Taim established in Lucas and Bager (2016b) projected by stage using stable stage distribution (the right eigenvector of the transition matrix A) (Mitro 2003).

Model scenarios, elasticity and sensitivity

From the baseline matrix, including parameters specified in Table 1, we altered those to account for the anthropogenic threats egg poaching for pet trade, road mortality and nest predation pressure. First, we implemented individual threats at time to explore the consequences in population growth rate (λ) from changing vital rates of the baseline matrix. For road mortality, we simulated the effect of decreasing the annual survival of adults based on the estimated per capita road mortality rate. As we do not have estimates for more than one year, we also used and road mortality rate of 0.02 to infer the potential of this threat to occur in years with higher mortality on roads. We simulated the effect of mortality due to nest predation pressure on the population by decreasing the annual survival of eggs using known estimates (18% and 98%) (Fagundes et al. 2007a; Gonçalves et al. 2007) and using intermediate levels of 50% and 75%. For egg poaching, we simulated this effect by removing 90% of the eggs laid every year from the population size estimate in this stage and looking at that effect on the entire population. We also set scenarios of simulating 75%, 50% and 25% levels of egg removal. Second, we implemented all threats in concert to explore the consequences in the population. In this case, we set a “best scenario” and a “worst scenario”. In the best scenario, we used the lowest values of each threat and in the worst scenario, we used the highest values of each threat. The population was considered extirpated at 10 or less female individuals.

We tested the potential to stabilize or grow the D'Orbigny's slider population through of reducing nest and adult female mortality and headstarting juvenile (i.e. captive hatching and rearing of turtles through an

early part of their life cycle) (Heppel et al. 1996). Our analysis were performed using “popbio” package (Stubben & Milligan 2007) in R (R Core Development Team, 2015) to carry out eigen analysis to estimate asymptotic λ , reproductive value (v) and stable stage distribution (w). Sensitivity and elasticity were set for all matrix combination generated. Sensitivities are the direct contributions of each population matrix element to determining λ and elasticities correspond to proportional changes in λ induced by proportional changes in the matrix elements (de Kroon et al. 1986; Crouse et al. 1987; Stevens 2009).

Results

The baseline projection matrix A for D’Orbygni slider turtle population was:

$$A = \begin{pmatrix} 0 & 0 & 3.10 \\ 0.42 & 0.276 & 0 \\ 0 & 0.005 & 0.98 \end{pmatrix}$$

and produced a $\lambda = 0.989$. From the matrix A we also estimated the reproductive value (v):

$$v = (1 \quad 2.35 \quad 336.0)$$

These estimates indicate that in average juvenile will contribute 2.35 times more to future generation as compared to the first stage, and in average adult stage will contribute 336 times more to future generations as compared to the first stage. The stable stage distribution (w) indicate that the population should be composed mostly by individuals from the egg/hatchling stage with a lower proportion of adult individuals (Figure 1).

Sensitivity was highest for the matrix element that included the transition between juvenile to adulthood (1.805) followed by annual adult

survival (0.978). Elasticity for annual adult survival (0.969) was higher than juvenile survival to adulthood (0.009). Proportional changes in female adult survival resulted in greater changes in λ compared to other vital rates. The ranks of these values for sensitivity and elasticity did not change when performing alternative scenarios.

Changing baseline matrix parameters by reducing stage-specific survival rates due to threats road mortality and nest predation pressure resulted in lower population growth rates for all scenarios (Table 2). Among scenarios in which only one threat was simulated at a time, observed λ for nest predation and road mortality showed that even road mortality with low estimated rates (1 to 2%), has the potential to decrease the population more rapidly if compared to higher level of nest predation (Table 2). For the egg poaching, the model showed a sharp decline at levels of 90% and 75% on the population size (Figure 2). The decline was also evident at lower levels, but the decay was not as pronounced and the overall decline was smoother and slower.

When acting in concert, all the threats produced accentuated declines (Figure 3), notably higher in the worst scenario. By using the lowest values (best scenario) of each threat, D'Orbygni slider turtle population would take 170 years to be extirpated. In the worst scenario, these losses would be anticipated to 72 years.

Increasing the survival rates in the first or second stage separately, through nest protection or headstarting juveniles, is not enough to produce a stable population ($\lambda = 1$). However, increasing only female adult survival in 44% through reducing road kill mortality a stable population is achieved. A stable population is also obtained by combining a management approach

that reduce nest depredation, egg poaching and headstarting juveniles. The most effective combination between the survival rates in the first and second stage would be 76.2% and 57.9%, respectively, if no mortality on the adult stage occur. In order to grow the population ($\lambda > 1$), we should increase survival at higher rates than above specified.

Discussion

Our most noticeable result is that D'Orbigny's slider turtle population is declining at a rate of about 1% per year ($\lambda = .989$). By estimating rates of road mortality, egg poaching and nest predation in this species, our model provides how these threats affect population growth rate and allow us to compare the magnitude of their effects. Our simulation indicate that as other long-lived turtles, this species is more sensitive to changes in adult mortality than changes in early stages (eggs, hatchlings and juveniles) (Congdon et al. 1993; Heppell et al. 1996; Enneson & Litzgus 2008; Crawford et al. 2014). Our simulations also showed that D'Orbigny's slider turtle population would decrease more rapidly with 2% of adult mortality than with 98% of nest mortality or 90% of egg removal.

Road kill can be considered one of the main sources of female mortality for turtle and other reptile species. It was hypothesized to severely affect populations, but only in recent years, this impact was quantified using population modelling approaches (Row et al. 2007; Crawford et al. 2014). For instance, diamondback terrapins (*Malaclemys terrapim*) has a moderate to high per capita road mortality (0.044-0.164) and Crawford et al. (2014) showed that about 3% of adult mortality is sufficient to aggravate

short-term population persistence. In fact, many early studies concerning long-lived turtles has demonstrated through sensitivity analysis a higher fragility of the late stages (Crouse et al. 1987; Heppel 1998; Enneson & Litzgus, 2008). In this study, we demonstrated that even a low per capita mortality rate (~1%) for D'Orbigny's slider living in areas surrounded by roads, is sufficient to decrease the population growth rate to a level that would harm their short-term persistence. As the study area is in a region with low road density, it is likely that the species suffers with higher road mortality through its range of distribution mainly in developed areas (i.e. higher road density). Therefore, many local populations might be at great risk of extirpation.

The results of our simulations also indicated that egg poaching at overwhelming intensities (90%) cause population to decline rapidly (Figure 2). Comparing to the baseline matrix curve, 90% of egg poaching anticipated in more than 350 years the time to the population be considered extirpated. The population responds in a different way when threats occur in early or late stages. When there is an increased loss of adults, the population has a more abrupt initial decline because there is a high direct effect on losing adult females that contribute to population growth in the next generations. When removal of eggs occurs, even if the total decline is large, there is a delay in the population response that is related to the time a turtle takes to develop from egg to reach sexual maturity and start reproducing (Tomillo et al. 2008). In the literature, there is no accurate records of when D'Orbigny's slider egg poaching has begun in southern Brazil. The collection of eggs and hatchlings to supply the black market has been documented since the late 80s (Molina & Rocha 1987; Lema &

Ferreira 1990), but it is likely that this practice is even older. From 1995 to 2003, many zoos received more than 1200 individuals from people who bought these animals in *pet shops* or markets (Voght et al. 2015). The high rates of D'Orbigny's slider egg poaching in south of Brazil was observed near urban centers (Fagundes et al. 2007a). Roads in this context can act causing indirect effects such as promoting increased hunting and poaching of individuals or in this case, eggs by local people thus facilitating illegal activities (Laurance et al. 2009; 2014). Many populations of D'Orbigny's slider live surrounding these man made structures and consequently, this threat has a potential to be widespread through its range of distribution and a real driver of extinction.

Another important source of mortality on the early stages and simulated in our study is nest predation. Rates of depredated nests range from usually 15% (Moll & Ledgler 1971; Fagundes et al. 2007a) and can attain extremely high values (over 90%) in many species (Congdon et al. 1983; Aresco 2004; Gonçalves et al. 2007). For instance, at rates of 18% nest predation, D'Orbigny's slider population has a very low short-term chance of extirpation (Figure 2). At this rate, the population would take more than 300 years to disappear. However, with increased rates, this interval would be attained in 201 years. The known estimates of nest predation for D'Orbigny's slider population is within the range of values specified above. Usually, low mortality rates occurred in populations living in areas closer to urban centers with only one predator responsible for the whole mortality (*Salvator merianae*). At another extreme, high rates were found in undisturbed areas where there is high density of nests and diversity of predators (Gonçalves et al. 2007). Researchers and managers need to

identify what are the predators and what is attracting them to turtle nests in order to design strategies reduce nest predation. This can be very important, particularly in areas where populations are declining due to little recruitment or low survival rates in other stages (Hopkins et al. 1981).

The accuracy of matrix population models to estimate the population growth rate and consequently to evaluate management strategies is highly correlated with the demographic parameters estimated. In turtles, although many life history traits are known, there are knowledge gaps in some parts of their life cycle, for example, hatchling and juvenile stage (Congdon et al. 1994; Pike et al. 2008; Enneson & Litzgus 2008). Fagundes et al. (2007b) provide the only known estimate of juvenile D'Orbigny's slider survival in an area of urban marsh and we are aware of our lack to provide variation in the demographic rates. Lower survival rates in this particular life cycle could be due to problems related to field searching or actually, it might reflect the reality (Congdon et al. 1993). Species with delayed sexual maturity and low fecundity demand high juvenile survivorship to maintain a stable population (Congdon et al. 1993). Females of D'Orbigny's slider can attain sexual maturity earlier (at 5 year old), and has a higher fecundity if compared to other freshwater species (Heppel 1998; Enneson & Litzgus 2008). Therefore, the observed juvenile rate could reflect the current recruitment in this population (Congdon et al. 1993). If this rate were higher than the specified value, this would reduce the elasticity of the adult stage and potentially would increase the relative importance of other life stages (Hart 2005; Crawford et al. 2014). However, this alteration would not change which stage conservation strategies should be focused. The elasticity still would aim the adult stage as a priority,

mainly due to its high reproductive value. Sources of juvenile mortality such as bycatch, even not mentioned in our simulations is possible to affect D'Orbigny's slider survival. Many local people depend on the freshwater ecosystem formed by the large continental lakes found in these regions for their livelihoods. In the last years, the number of people carrying out this activity without legal permission has increased (Piedras et al. 2012). Thus, the use of fishing nets could be an additional factor contributing to juvenile mortality through its drowning (Crouse et al. 1987; Larocque et al. 2012).

A range of management strategies is available for turtles. Early studies focused mainly on the nest protection due to the facility to monitoring losses on nesting sites (Crouse et al. 1987). Despite this facility, this method would be difficult to implement because nests are extremely hard to find in the field and managers should have high nest-related skills to quantitatively assess its success. As other stages were being identified as a priority for conservation, other strategies have emerged, for example, headstarting programs (Heppel et al. 1996). Headstarting turtles through their most vulnerable stages (eggs, hatchlings and juveniles) and release them into the wild, are costly and effective only when early stages are the most vulnerable (Heppel et al. 1996). Our results indicate that this action could be an option. However, this would be effective only by increasing the survival of the first stage in 72.9% (through nest protection or poaching control) and rearing juveniles to 5 year (until maturity) with a high success rate. In practice, it seems challenging to protect only early stages and have to account that other problems be irrelevant (e.g. egg failure, nutritional deficiency from confined hatchlings maintenance).

Adults are the main stage which management strategies should be focused as pointed out by our elasticity analysis in all scenarios. Road kill is the main source of D'Orbigny's slider adult female mortality in south of Brazil (Bager et al. 2007). The most widespread strategies to reduce mortality of reptiles and highly effective include fences and wildlife crossing structures (Dood et al. 2004; Aresco 2005; Andrews et al. 2015). As the species has a wide mortality within its range of distribution (Hengemühle & Cademartori 2008; Bager & Fontoura 2013), it is challenging to think that these management actions would be employed as the most preferable. Turtles in special have higher mortality risk linked to specific annual periods (i.e. *hot-moments*) (Beaudry et al. 2010; Costa et al. 2015). This occurs during the reproductive season (October-January) when they usually move longer distances between permanent and seasonal wetlands to find suitable nesting areas. Target strategies to reduce mortality during this season, for example, could include periodic road closures, exclusionary fencing and volunteers to patrol roadside areas. These strategies could be designed in many high-risk roads within species distribution (Beaudry et al. 2008). Road closures are applied for many snake and turtle species in United States (Shepard et al. 2008) and could be an option only if alternate route for drivers are available (Andrews et al. 2015).

As we demonstrated, road mortality, nest predation and egg poaching pressures will affect turtle survival and consequently, their persistence. Strategic actions for this species ought to include protection in adult stage to guarantee that failing into accounting to any level of the threat do not harm the efficacy of the whole protection plan.

Acknowledgements

Field support, both logistical and financial, was provided by Brazilian Agency CNPq for A. Bager. PSL was supported by a PhD grant from Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (Capes) in Brazil. MDM was supported by a PhD grant from Ciência sem Fronteira Program. Suggestions from Ana Bárbara Gonçalves were very helpful.

Literature cited

- Alves RR da N, Nogueira EEG, Araujo HFP, Brooks SE. 2010. Bird-keeping in the Caatinga, NE Brazil. *Human Ecology* **38**:147–156.
- Andrews KM, Langen TA, Struijk RPJH. 2015. Reptiles: Overlooked but often at risk from roads. Pages 271-280 in van der Ree R, Smith DJ, Grilo C, editors. *Handbook of Road Ecology*. Wiley Blackwell.
- Aresco MJ. 2004. Reproductive ecology of *Pseudemys floridana* and *Trachemys scripta* (Testudines: Emydidae) in northwestern Florida. *Journal of Herpetology* **38**: 249-256.
- Aresco MJ. 2005. The effect of sex-specific terrestrial movements and roads on the sex ratio of freshwater turtles. *Biological Conservation* **123**:37–44.
- Bager A, Fontoura V. 2013. Evaluation of the effectiveness of a wildlife roadkill mitigation system in wetland habitat. *Ecological Engineering* **53**:31–38.
- Bager A, Freitas TRO De, Krause L. 2007. Nesting ecology of a population of *Trachemys dorbignyi* (Emydidade) in Southern Brazil. *Herpetologica* **63**:56–65.
- Bager A, Rosado JLO. 2010. Estimation of Core Terrestrial Habitats for Freshwater Turtles in Southern Brazil Based on Nesting Areas.

- Journal of Herpetology **44**:658–662.
- Baker SE, Cain R, Kesteren van F, Zommers ZA, D`Cruze N, MacDonald DW. 2008. Rough Trade: Animal Welfare in the Global Wildlife Trade. *BioScience* **63**:1–12.
- Beaudry F, deMaynadier PG, Hunter ML. 2008. Identifying road mortality threat at multiple spatial scales for semi-aquatic turtles. *Biological Conservation* **141**:2550–2563.
- Beaudry F, DeMaynadier PG, Hunter Jr. ML. 2010. Identifying hot moments in road-mortality risk for freshwater turtles. *Journal of Wildlife Management* **74**:152–159.
- Bertonati C. 1994. Lista propuesta de anfibios y reptiles amenazados de extinción. Asociación Herpetologica Argentina. Cuadernos de Herpetología **8**:164-171
- Bohm M et al. 2013. The conservation status of the world's reptiles. *Biological Conservation* **157**:372–385.
- Borda-de-Água L, Grilo C, Pereira HM. 2014. Modeling the impact of road mortality on barn owl (*Tyto alba*) populations using age-structured models. *Ecological Modelling* **276**:29–37.
- Bujes CS, Molina FB, Verrastro L. 2011. Population Characteristics of *Trachemys dorbigni* (Testudines , Emydidae) from Delta Do Jacuí State Park , Rio Grande Do Sul , Southern Brazil. *South American Journal of Herpetology*, **6**:27–34.
- Burnham KP, Anderson DR. 2002. Model selection and multimodel inference: a practical information-theoretic approach.
- Bush ER, Baker SE, Macdonald DW. 2014. Global trade in exotic pets 2006-2012. *Conservation Biology* **28**:663–676.
- Caswell H. 1989. *Matrix Population Models: Construction, Analysis, and Interpretation*. Sinauer Associates, Sunderland, MA.

- Chapin FS, Zavaleta ES, Eviner VT, Naylor RL, Vitousek PM, Reynolds HL, Hooper DU, Lavorel S, Sala OE, Hobbie SE, Mack MC, Diaz S. 2000. Consequences of changing biodiversity. *Nature* **405**:234–42.
- Congdon JD, Tinkle DW, Breitenbach GL, van Loben Sels RC. 1983. Nesting ecology and hatching success in the turtle *Emydoidea blandingii*. *Herpetologica* **1983**: 417-429.
- Congdon JD, Dunhan AE, van loben Sels, RC. 1993. Delayed sexual maturity and demographics of Blandingii's turtle (*Emydoidea blandingii*): implications for conservation and management of long-lived organisms. *Conservation Biology* **7**: 826-833.
- Congdon JD, Dunhan AE, van loben Sels RC. 1994. Demographics of the common snapping turtles (*Chelydra serpentina*): implications for conservation and management of long-lived organisms. *The American Zoologist* **34**: 397-408.
- Costa AS, Ascensão F, Bager A. 2015. Mixed sampling protocols improve the cost-effectiveness of roadkill surveys. *Biodiversity and Conservation* **24**:2953–2965.
- Costello MJ, May RM, Stork NE. 2013. Can We Name Earth 's Species. *Science* **339**:413–416.
- Cooch E, White G. 2001. Using MARK - a gentle introduction. Cornell University, Ithaca.
- Crawford BA, Maerz JC, Nibbelink NP, Buhlmann KA, Norton TM. 2014. Estimating the consequences of multiple threats and management strategies for semi-aquatic turtles. *Journal of Applied Ecology* **51**:359–366.
- Crouse DT, Crowder LB, Caswell H, Oct N, Crouse T. 1987. A Stage-Based Population Model for Loggerhead Sea Turtles and Implications for Conservation. *Ecology* **68**:1412–1423.
- D'Anuniação PER, Lucas PS, Silva VX, Bager A. 2013. Road ecology and Neotropical amphibians: Contributions for future studies. *Acta*

- Herpetologica **8**:129–140.
- De Kroon H, a. P, J. VG, H. C. 1986. Elasticity: the relative contribution of demographic parameters to population growth rate. *Ecology* **67**:1427–1431.
- Dodd CK, Barichivich WJ, Smith LL. 2004. Effectiveness of a barrier wall and culverts in reducing wildlife mortality on a heavily traveled highway in Florida. *Biological Conservation* **118**:619–631.
- Dunham E. 2011. Demographics of Common Snapping Turtles (*Chelydra Serpentina*): Implications for Conservation and Management of Long-Lived Organisms Author (s): Justin D . Congdon , Arthur E . Dunham , R . C . van Loben Sels Published by : Oxford University Press cont. Population (English Edition) **34**: 397–408.
- Enneson JJ, Litzgus JD. 2008. Using long-term data and a stage-classified matrix to assess conservation strategies for an endangered turtle (*Clemmys guttata*). *Biological Conservation* **141**:1560–1568.
- Fagundes CK, Bager A, Terezinha S, Cechin Z. 2010a. *Trachemys dorbigni* in an anthropic environment in southern Brazil: I) Sexual size dimorphism and population estimates. *Herpetological Journal* **20**: 185-193.
- Fagundes CK, Bager A, Terezinha S, Cechin Z. 2010b. *Trachemys dorbigni* in an anthropic environment in southern Brazil: II) Reproductive Ecology. *Herpetological Journal* **20**: 195-199.
- Fahrig L, Rytwinski T. 2009. Effects of roads on animal abundance: an empirical review and synthesis. *Ecology and Society* **14**: 21–41.
- Fernandes-Ferreira H, Mendonça SV, Albano C, Ferreira FS, Alves RRN. 2012. Hunting, use and conservation of birds in Northeast Brazil. *Biodiversity and Conservation* **21**: 221–244.
- Fordham DA, Georges A, Brook BW. 2008. Indigenous harvest, exotic pig predation and local persistence of a long-lived vertebrate: Managing

- a tropical freshwater turtle for sustainability and conservation. *Journal of Applied Ecology* **45**: 52–62.
- Forman RTT, Alexander LE. 1998. Roads and Their Major Ecological Effects. *Annual Review of Ecology and Systematics* **29**: 207–231.
- Forman RTT, Deblinger RD. 2000. The Ecological Road-Effect Zone of a Massachusetts (USA) Suburban Highway. *Conservation Biology* **14**: 36-46.
- Gibbon JW et al. 2000. The Global Decline of Reptiles, Déjà Vu Amphibians. *BioScience* **50**: 653–666.
- Gonçalves F a., Cechin SZ, Bager A. 2007. Predação de ninhos de *Trachemys dorbigni* (Duméril & Bibron) (Testudines, Emydidae) no extremo sul do Brasil. *Revista Brasileira de Zoologia* **24**:1063–1070.
- Hart KM. 2005. Population biology of diamondback terrapins (*Malaclemys terrapin*): defining and reducing threats across their geographic range. PhD thesis, Duke University, Durham, North Carolina, USA.
- Hengemühle A, Cademartori CV. 2008. Levantamento de mortes de vertebrados silvestres devido a atropelamento em um trecho da Estrada do Mar (RS-389). *Biodiversidade Pampeana* **6**:4–10.
- Hepell SS, Crowder LB, Crouse DT. 1996. Models to Evaluate Headstarting as a Management Tool for Long-Lived Turtles. *Ecological Applications* **6**:556–565.
- Hepell SS. 1998. Application of Life-history theory and population model analysis to turtle conservation. *Copeia* **1998**: 367-375.
- Hopkins SR, Murphy Jr TM, Stansell KB, Wilkinson PM. 1978. Biotic and abiotic factors affecting nest mortality in the Atlantic loggerhead turtle. *Proceeding of the Annual Conference of the Southeastern Association of Fish and Wildlife Agencies* **32**: 213-223.
- Jacobson SL, Bliss-ketchum LL., Rivera CE De, Smith WP, 2016. A

- behavior-based framework for assessing barrier effects to wildlife from vehicle traffic volume. *Ecosphere* **7**: 1–15.
- Jaeger JAG. 2015. Improving Environmental Impact Assessment and Road Planning at the Landscape Scale. Pages 32-40 in van der Ree R, Smith DJ, Grilo C, editors. *Handbook of Road Ecology*. Wiley Blackwell.
- Jenkins RKB et al. 2014. Extinction risks and the conservation of Madagascar's reptiles. *PLoS ONE* **9**:e100173.
- Kuchling G. 1988. Population structure, reproductive potential and increasing exploitation of the freshwater turtle *Erymnochelys madagascariensis*. *Biological Conservation* **43**:107–113.
- Laurance WF, Goosem M, Laurance SGW. 2009. Impacts of roads and linear clearings on tropical forests. *Trends in Ecology and Evolution* **24**:659–669.
- Laurance WF, Clements GR, Sloan S, O'Connell CS, Mueller ND, Goosem M, Venter O, Edwards, DP, Phalan B, Balmford A, Van Der Ree R, Arrea IB. 2014. A global strategy for road building. *Nature* **513**: 229–232.
- Lefkovich L. 1965. The study of population growth in organisms grouped by stages. *Biometrics*, 21, 1–18.
- Lema T, Ferreira MTS. 1990. Contribuição ao conhecimento dos Testudines do Rio Grande do Sul (Brasil) – Lista sistemática comentada (Reptilia). *Acta Biologica Leopoldensia* **12**:125-164.
- Leuteritz T, Rioux Paquette S. (Madagascar Tortoise and Freshwater Turtle Red List Workshop). 2008. *Astrochelys radiata*. *The IUCN Red List of Threatened Species* 2008:e.T9014A12950491.<http://dx.doi.org/10.2305/IUCN.UK.2008.RLTS.T9014A12950491.en>
- Lucas PS, Bager A. 2016 a. Contrasting reproductive strategies of D'Orbigny's slider turtle at the same latitude range in subtropical

wetlands. Manuscript submitted for publication.

Lucas OS, Bager A. 2016 b. Demography of *Trachemys dorbigni* in a protected area in South of Brazil. Unpublished manuscript.

Mitro MG. 2003. Demography and viability analyses of a diamondback terrapin population. *Canadian Journal of Zoology* **81**: 716-726.

Molina FDB, Rocha MB. 1987. Tartaruga japonesa: biologia e manejo. *Aquacultura* **3**: 27-28.

Moll EO, Legler L. 1971. The life history of a neotropical slider turtle *Pseudemys scripta* (Schoepff), in Panama. *Bulletin of the Los Angeles County Museum of Natural History* **11**: 72-79.

Newbold T, Hudson LN, Hill SLL, Contu S, Lysenko I, Senior RA, Börger L, Bennett DJ, Choimes A, Collen B, Day J, De Palma A, Díaz S, Echeverria-Londoño S, Edgar MJ, Feldman A, Garon M, Harrison MLK, Alhousseini T, Ingram DJ, Itescu Y, Kattge J, Kemp V, Kirkpatrick L, Kleyer M, Correia DLP, Martin CD, Meiri S, Novosolov M, Pan Y, Phillips HRP, Purves DW, Robinson A, Simpson J, Tuck SL, Weiher E, White HJ, Ewers RM, Mace GM, Scharlemann JPW, Purvis A. 2015. Global effects of land use on local terrestrial biodiversity. *Nature* **520**: 45–50.

Nijman V, Shepherd CR. 2015. Analysis of a decade of trade of tortoises and freshwater turtles in Bangkok, Thailand. *Biodiversity and Conservation* **24**:309–318.

Pereira HM et al. 2010. Scenarios for global biodiversity in the 21st century. *Science* **330**:1496–1501.

Pereira HM, Navarro LM, Martins IS. 2012. Global biodiversity change: the bad, the good, and the unknown. *Annual Review of Environment and Resources* **37**:25–50.

Piedras SRN, Santos JD, Fernandes JM, Tavares R a., Souza DM, Pouey JLOF. 2012. Caracterização da atividade pesqueira na Lagoa Mirim,

- Rio Grande do Sul - Brasil. *Revista Brasileira de Agrociência* **18**:107–116.
- Pike DA, Pizzatto L, Pike BA, Shine R. 2008. Estimating survival rates of uncatchable animals: The myth of high juvenile mortality in reptiles. *Ecology* **89**:607–611.
- Pimm SL, Russell G J, Gittleman JL, Brooks TM. 1995. The Future of Biodiversity. *Science* **269**:347–350.
- Pimm SL, Jenkins CN, Abell R, Brooks TM, Gittleman JL, Joppa LN, Raven PH, Roberts CM, Sexton JO. 2014. The biodiversity of species and their rates of extinction, distribution, and protection. *Science* **344**:1246752.
- Pimm SL, Raven P. 2000. Biodiversity. Extinction by numbers. *Nature* **403**:843–845.
- Prado WS, Albareda DA, Cabrera MR, Etchepare E, Giraudo AR, Prosdocimi L, Richard E. 2012. Categorización del estado de conservación de las tortugas de la República Argentina **26**:375–387.
- R Core Team. 2015. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL <https://www.R-project.org/>
- Rosa CA da, Bager A. 2012. Seasonality and habitat types affect roadkill of neotropical birds. *Journal of Environmental Management* **97**:1–5.
- Row JR, Blouin-Demers G, Weatherhead PJ. 2007. Demographic effects of road mortality in black ratsnakes (*Elaphe obsoleta*). *Biological Conservation* **137**:117–124.
- Ryu HY, Shoemaker KT, Kneip E, Pidgeon AM, Heglund PJ, Bateman BL, Thogmartin WE, Akçakaya HR. 2016. Developing population models with data from marked individuals. *Biological Conservation* **197**:190–199.
- Shepard DB, Kuhns AR, Dreslik MJ, Phillips CA. 2008. Roads as barriers

- to animal movement in fragmented landscapes. *Animal Conservation* **11**:288–296.
- Silveira ML. 2014. Variação morfológica e populacional de *Trachemys dorbigni* (Testudines, Emydidae) no extremo sul do Brasil. Master thesis, Universidade Federal de Lavras.
- Smith LL, Dodd Jr, CK. 2003. Wildlife mortality on highway US 441 across Paynes Prairie, Alachua County, Florida. *Florida Scientist* **66**: 128–140.
- Stevens MH. 2009. *A Primer of Ecology with R*. Springer Science & Business Media.
- Stubben C, Milligan B. 2007. Estimating and Analyzing Demographic Models. *Journal Of Statistical Software* **22**:1–23.
- Tomillo PS, Saba VS, Piedra R, Paladino F V., Spotila JR. 2008. Effects of illegal harvest of eggs on the population decline of leatherback turtles in Las Baulas Marine National Park, Costa Rica. *Conservation Biology* **22**:1216–1224.
- Tuma MW, Millington C, Schumaker N, Burnett P. 2016. Modeling Agassiz's desert tortoise population response to anthropogenic stressors. *Journal of Wildlife Management* **80**:414–429.
- Vogt RC, Fagundes CK, Bataus YSL, Balestra RAM, Batista FRW, Uhlig VM, Silveira AL, Bager A, Batistella AM, Souza FL, Drummond GM, Reis IJ, Bernhard R, Mendonça SHT, Luz VLF. 2015. Avaliação do Risco de Extinção de *Trachemys dorbigni* (Duméril & Bibron, 1835) no Brasil. Processo de avaliação do risco de extinção da fauna brasileira. ICMBio.

List of Tables

Table 1: Parameters used in the baseline deterministic matrix of D'Orbigny's slider turtle at the ESEC Taim, Brazil.

Parameter	Value	Sources
Nest survival	0.02 – 0.82	Gonçalves et al. 2007, Fagundes et al. 2007
Juvenile survival	0.276	Fagundes et al 2007, Supporting Information
Adult survival	0.98	Lucas and Bager 2016b
Female age at first breeding	5	Lucas and Bager 2016b
Mean clutch size	10.3	Lucas and Bager 2016a
Mean clutch frequency	1.44	Bager et al. 2007

Table 2: D'Orbygni slider turtle population growth rate (λ) response to simulations based on the threats road mortality and nest predation pressure. Scenario represents the percentage of the threat levels.

Threat	Scenario	λ
Nest predation	98	0.980
	75	0.982
	50	0.984
	18	0.987
Road mortality	1	0.979
	2	0.969

List of Figures

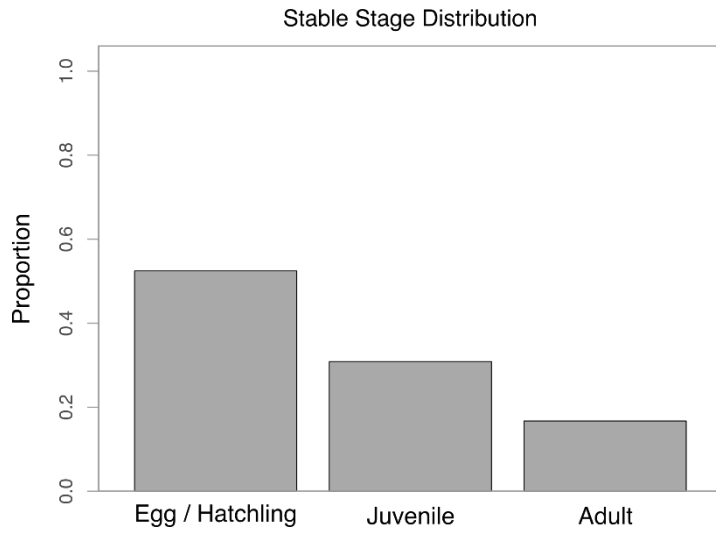


Figure 1: Proportion of D'Orbygni slider turtle individuals in each stage in a stable distribution.

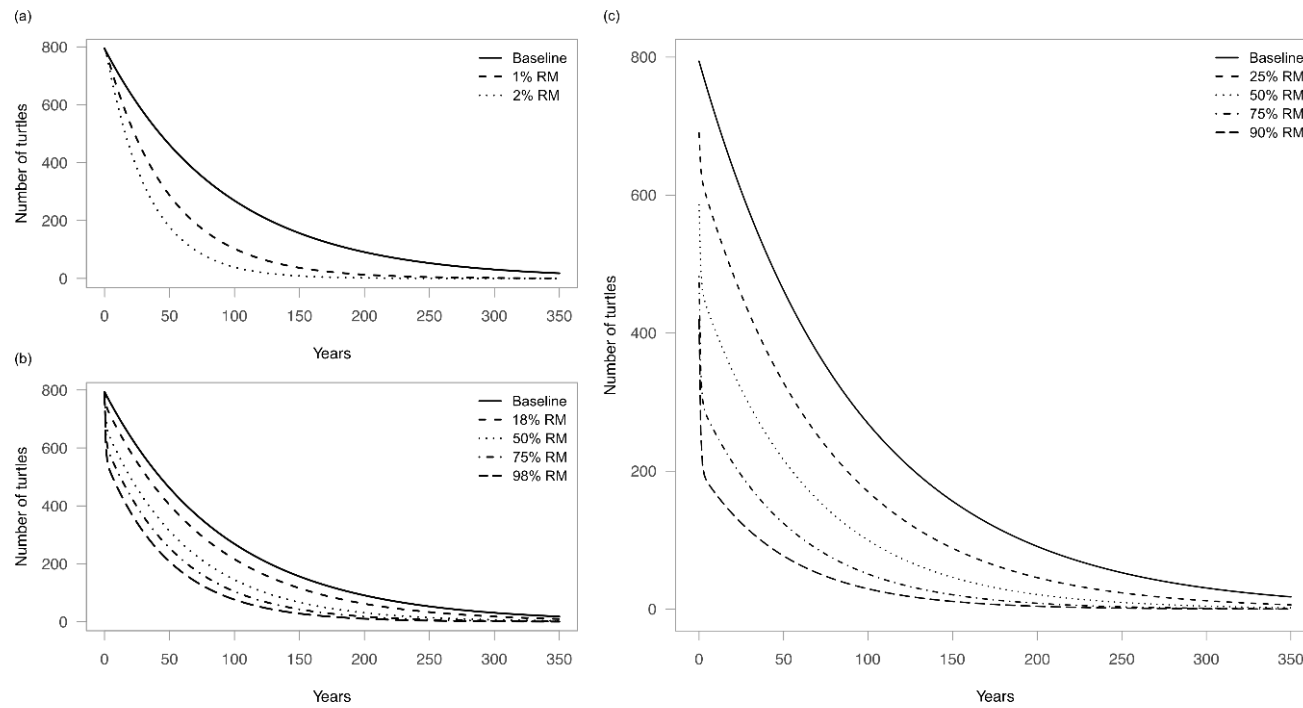


Figure 2: The effect of different threats and levels of a) road mortality, b) nest predation and c) egg poaching on the population size of D'Orbygni slider turtle in Brazil. Baseline is the deterministic matrix projection.

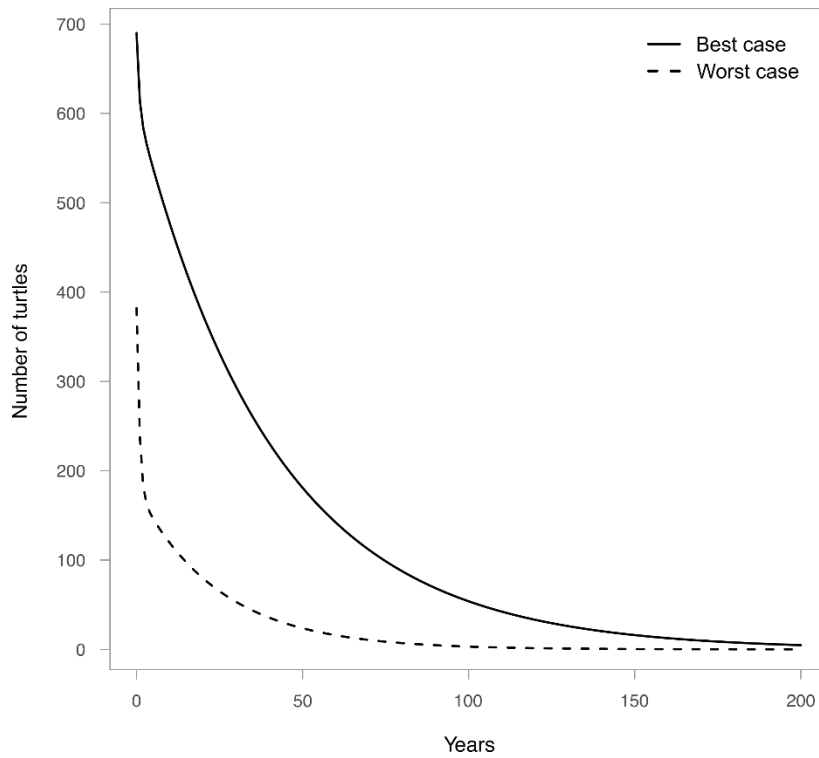


Figure 3: Best and worst case scenarios including lowest and highest values of all threats and their effects on the time to D'Orbygni slider population extirpation.

Supporting Information

Estimating per capita road mortality rate

Annual road mortality rates of D'Orbigny's slider turtle vary between 0.496 - 0.615 ind./km/yr (Costa et al. 2015). In order to estimate a per capita road mortality rate we considered the individuals road killed in a 14 km road stretch (total length monitored near ESEC Taim) in 2005 based on the database provided by A. Bager. To estimate per capita mortality rate we needed data on the road killed animals and population size estimates. We found 22 female individuals road killed, and we used data on population size estimate provided by Lucas and Bager (2016b). D'Orbigny's slider turtle population size was estimated in 1588 individuals in two square kilometers (or 794 individuals/km²) in ESEC Taim with a sex ratio of 1:1 (Silveira 2013). As we are dealing with a female matrix and road mortality affect most adult females in this species, we considered here an adult female population size consisted of 133 individuals/km². This number was estimated based on the stable stage distribution (dominant eigenvector) of the deterministic matrix population, i.e. the distribution of individuals among the stages (see figure 1).

As we have turtle mortality on roads measured in a length unit (kilometers), we should convert this to a mortality rate per capita per area. Based on D'Orbigny's slider turtle female nest movements Bager et al. (2012) estimated a mean displacement of 500 m in a year. Considering this distance, in both roadsides, we established that in an area of 14 km² would occur 1862 adult females (i.e. 133 ind./km² × 14 km²). After that, per capita road mortality rate was computed as:

$$RM = N_{killed}/N_{size}$$

where, N_{killed} is the number of females killed on roads and N_{size} is the number of estimated adult females in the total area (14 km²).

Estimating juvenile annual survival rate

To our knowledge, only Fagundes et al. (2007b) provided juvenile survival rate. However, their estimates were set as weekly survival rate, based on a sampling done every week from January to December in 2006. We used the same database (provided by A. Bager) to perform analysis based on capture and recapture data of D'Orbigny's slider turtle in the program MARK (Cooch & White 2013). We grouped encounter histories (1 as recaptured individuals and 0 as not recaptured, see Cook & White 2013) in a monthly-unit effort, instead of using the weekly-unit effort as Fagundes et al. (2007b).

We used Cormack-Jolly Seber models to estimate apparent annual survival (ϕ) and recapture probabilities (p). Four models were fitted to the data: a fully time-dependent model ($\phi t p t$) and reduced models [$(\phi t p (.); \phi (.) p t; \phi (.) p (.)$], where t means time dependent and $(.)$ means constant survival or recapture through time. Models were compared by AIC (Burnham & White 2002) and then the parameter estimates were generated. The estimate of the best model for the monthly-survival probability was 0.901 (ranging from 0.830 to 0.963). However, we needed the annual survival, so Cook and White (2013) derived a simple equation to extrapolate to the expected annual survival as e^n , where e is the monthly-survival estimate and n is the number of months included in the encounter history. Thus, we get the annual survival as 0.901^{12} .

ARTIGO 3

Densidade global de rodovias compromete a persistência de tartarugas.

Preparado de acordo com as normas da Revista Diversity and Distributions.

Lucas P.S., Bager A., Pereira HM. (2016) Global road density jeopardize turtle persistence.

Priscila da Silva Lucas^{1,2,*}, Alex Bager^{1,2} & Henrique M. Pereira^{3,4}

¹Programa de Pós-Graduação em Ecologia Aplicada, Departamento de Biologia, Universidade Federal de Lavras, Campus Universitário, CEP 3037, Lavras, CEP 37200 000 Brasil. Tel.: +55 35 3829 1928

²Centro Brasileiro de Estudos em Ecologia de Estradas, Universidade Federal de Lavras, Campus Universitário, CP 3037, Lavras, CEP 37200 000 Brasil. Tel.: +55 35 3829 3129

³German Centre for Integrative Biodiversity Research (iDiv) Halle-Jena-Leipzig, Deutscher Platz 5e, 04103 Leipzig, Germany

⁴Institute of Biology, Martin Luther University Halle-Wittenberg, Am Kirchtor 1, 06108 Halle (Saale), Germany

*Corresponding author: Priscila da Silva Lucas: prilucass@gmail.com

Abstract: Habitat loss and fragmentation imposes negative effects on global biodiversity. Roads are an important source of impact of habitat fragmentation and this is especially an alarming issue to turtles because of their particular life history traits. We integrated a multispecies approach in a global scale to evaluate turtle species response to road density based on life history, demographic, species distribution, road mortality data and road infrastructure information. This assessment is based on a demographic model that takes into account population growth rates in suitable and human-modified habitats and dispersal distance. This approach give us estimates of critical values of road density and patch size in which species are expected to go extinct, allow us to rank more vulnerable species and map within their distribution, where they are most impacted. We identified 16 species to be affected by roads. These species belong to the families Emydidae, Kinosternidae, Geoemydidae, Chelidae and Tryonichidae. Most of the species are categorized as Vulnerable, Endangered and Critically Endangered regarding IUCN and only 18.7% of the species in the 5th percentile have roads related as one of the major threats. The species *Hardella thurji*, *Heosemys anandalli* with distribution in Asia and *Pseudemys alabamensis* occurring in North America pointed out by our model are expected to be in danger in 80 % of their distribution range. The highest number of species affected by roads per grid cell is observed in Asia (up to four species) and North America (up to three species). Further studies ought to extend our approach to other turtle species, because more than half of the described species are not even listed in the IUCN. Many of these species may already be in their thresholds of road threat. In addition,

there is the need to promote reassessments of the major threats to various turtle species worldwide.

Keywords: global assessment, long-lived animals, road effect, planning, conservation

INTRODUCTION

In the current scenario habitat loss and fragmentation impose expressive negative effects on the biota (Fahrig 2003, Pereira et al. 2010) and consequently land-use change are one of the main drivers of global biodiversity change (Pereira et al. 2012, Newbold et al. 2015). Roads and other linear clearings are an important source of habitat fragmentation (Goosem 2007, Laurence et al. 2001, 2009, 2014). Road density is linked to this process through the road network expansion that cause an increased loss of natural areas (Lee et al. 2004, Goosem 2007) and moreover, reduce suitable patch sizes for species persistence (Jaeger & Holderegger 2005, Jaeger et al. 2005, Jaeger 2015).

A great amount of species, including turtle are declining on a global scale (Gibbons et al 2000, Irwin & Irwin 2005, Todd et al 2010), mostly induced by terrestrial and wetland habitat reduction and alteration (Gibbons et al 2000). The precise mechanisms of these declines still remain unknown but are presumably related to a general degradation of habit quality from anthropogenic land conversion (Todd et al 2010), via indirect effects on long-term survival and reproduction. Roads can affect wildlife populations by different local-scale mechanisms (Forman & Alexander 1998),

especially turtles with particular life history traits it is unlikely that some populations can recover even with low adult mortality rates (Congdon et al 1993, Crawford et al 2014a). For instance, there is some evidence that adults, mainly females are more susceptible to road mortality (Aresco 2005a, Gibbs & Steen 2005) because females often move over land in search of suitable nesting sites, thus possibly being exposed to roads (Aresco 2005a, Bager et al. 2007). Biased populations could be an indirect indicative that roads can affect local population persistence (Aresco 2005a; Dorland et al 2014). In particular, sex ratio skewness in low-density populations can reduce population growth rates by demographic stochasticity (Stephens et al. 1999).

Concerning road density, few studies that were carried on a single or few-species and local to regional scales level (Mech et al. 1988, Gibbs & Shiver, 2002, Beaudry et al. 2008) can give us some perceptions about the problems. The increased land-use intensity by this feature was associated with decreased genetic diversity (Reid & Peery 2014), changes in population structure (Steen & Gibbs, 2004, Reid & Perry 2014) and distribution of female body sizes (i.e. females with large carapace length in landscapes where road density is high) (Hamer et al. 2016). However, its direct effects on population persistence it is poorly understood yet (Jaeger 2015, Harmer et al. 2016). In a broader scale, understand how the density and configuration of the road network affect the functional relationships within and between ecosystems (Heilman et al. 2002) and population persistence (Ceia-Hasse et al. 2016) are priority to better support road planning and transportation strategies (Forman and Deblinger 1998, van der Grift 2005, van der Ree et al. 2011, Jaeger, 2015). Further, there is the

need to integrate different approaches, for example, models based on life history and habitat parameters (Pereira et al 2004, Pereira & Daily 2006) to better address the problem.

In this study, we integrated a multispecies approach in a global scale to evaluate species response to road density within their distribution and to rank the vulnerability of that ones who are most jeopardized. This assessment is based on a demographic model (Skellam 1951, Borda-da-Água et al 2012) that takes into account population growth rates in suitable and human-modified habitats as well dispersal distance measures. We intended to determine, i) the maximum road density above which turtle populations are not expected to survive, ii) the minimal patch size surrounded by roads below which populations are not expected to survive, iii) to rank the turtle species most vulnerable to these threats and iv) to specify where within their distribution species are most affected and constructing new roads should be avoided.

MATERIAL AND METHODS

Turtle data

Many studies have demonstrated that turtle taxonomy is very confuse (Fujita et al. 2004, Bickham et al. 2007, Guillon et al. 2012). Accordingly, there is always the need for revision due to an inconsistent consensus in the scientific community (Bickham et al. 2007, Ihlow et al. 2012). As this is still discussed, we herein decided to follow the last checklist of taxonomy adopted by the Turtle Taxonomy Working Group that comprises 335 species (Van Dijk et al. 2014). In this assessment, we only included turtle species that met the following criteria: i) to be listed in

the IUCN red list, ii) to be associated with terrestrial systems and, iii) to present available map about its range distribution. After these steps, we included 156 turtle species listed in the IUCN distributed throughout the world (Figure 1).

Determining critical values of road density and patch size

Maximum road density (RD_{mx}) and minimal patch size PA_{mm} in which turtle populations are not expected to survive were estimated based on the equations derived by Borda-de-Água et al. (2012) that took in account the model proposed by Skellam (1951) (see Supporting Information; SI).

$$RD_{mx} = r_1 / (r_1 + |r_0|)$$

and

$$PA_{mm} = \pi^2(\sigma^2/r_1)$$

where r_1 is the population growth rate in a suitable habitat, r_0 is the population growth rate in a unsuitable habitat and σ^2 is dispersal variance.

Life history and demographic data

Before computing the critical values, we first calculated two parameters of the demographic model (r_1 and r_0) for each freshwater turtle species. We estimated the population growth rate, r_1 , in a suitable habitat using a modified version of the Euler equation as in Pereira & Daily (2006). We used this equation because it makes some simpler assumptions 1) constant mortality rate, μ , between age-classes; 2) fecundity equals the number of female offspring (50% of the clutch size), b , at regular birth

pulses intervals after the breeding age, β ; and 3) birth pulse intervals are spaced by the mean interval between clutches, Δ , as follow.

$$b \times \int_0^{\infty} \sum_{y=0}^{\infty} \delta(x - y\Delta - \beta) e^{-(r+m)x} dx = 1$$

where $\delta(x)$ is the birth pulse function, which has a value of $1/T$ for x between 0 and T and 0 elsewhere. Mortality rate (μ) was computed as the inverse of mean life span. We are aware about the limitations of the assumptions of constant mortality. In turtles, it is usual to observe high mortality during the first stages (Tinkle et al 1981; Congdon 1994) but low adult mortality (Pike et al 2008). As they are long-lived animals, reliable life table studies from long-term monitoring of the population are scarce (Congdon 1994; Congdon et al. 1993) and we do not have enough data to estimate survival in all age classes, so we tried to identify the “best scenario” for survival (Pereira et al. 2004).

To calculate fecundity, clutch size was assumed to be the number of eggs per year. We calculated this by multiplying the number of eggs per clutch by the number of clutches per year because many turtle species can divide their reproductive output reproducing more than once during the reproductive season (Iverson 1992). While noting a high potential number of eggs laid per year, its outbreak is not entirely successful. It is common to observe high nest predation rates, for example, between 80 % and 98% for some species (Ernst and Lovich, 2009; Flitz & Mullin 2006, Jones 2006, Gonçalves et al. 2007). We estimated a mean rate of predation based on previous literature (Table A1; SI), and we used this rate as a reduction in fecundity for each species in order to calculate population growth rates.

Other features as egg infertility, embryonic mortality and developmental abnormalities that also cause egg failure (Peters et al 1994) were not assumed due to the difficulty of obtaining accurate estimates. Birth pulse interval was assumed to be of one-year interval because great amount of female turtles within populations can reproduce in consecutive years (Bager et al 2007, Litzig et al 2008).

We estimated the population growth rate, r_0 , on roads as Ceia-Hasse et al. (2016 in press) from studies published on turtle road mortality (Table A2; SI):

$$r_0 = N_k/N$$

where N_k is the number of individuals *road killed* per $\text{km}^2 * \text{yr}^{-1}$ and N is the population density measured in individuals/ km^2 . For species with missing data, we used r_0 median values.

We used life history data (Table A3; SI) and body size data to compute the model parameters for each species. Missing data ranged from 5% for body size to 93% for dispersal. For body size, age breeding and clutch frequency, where data was lacking, we computed the mean of the genus (in most cases) or the family. For clutch size and mortality rates, allometric relationships (Table A5; SI) family-based were established, and then used to estimate the life history parameters for other species. In these cases, for non-significant relationships mean of the genus or family was also used. For dispersal variance, we applied an allometric relationship with body size using all data grouped.

In order to determine dispersal variance, we converted mean dispersal distances d based in 11 turtle and tortoise species data (Table A3,

Kazmaier et al. 2002, Roe et al. 2009) assuming a Gaussian dispersal (Pereira et al 2004) using:

$$\sigma^2 = d^2(2/\pi)$$

measured in square kilometers per life span. This give us a lifetime dispersal in units of km²/generation. To convert this to annual dispersal (km²/year), we divided lifetime dispersal by mean life span, $1/\mu$.

Determining observed road density and patch size

We used turtle distribution maps available from the IUCN and road network maps from the Open Street Map (OSM). We intersected the maps for each species to compute the observed road density (RD_{ob}) and patch size (PA_{ob}) inside their ranges. We included road categories from OSM as highways, roads and unpaved roads. We calculated the observed road density (RD_{ob}) and the observed patch size (PA_{ob}) that are the patches surrounded by roads inside each species range distribution.

Ranking and mapping species

We computed the 5th and 25th percentiles as criteria to include the species most affected by road density and patch size based on the following index: RD_{mx}/RD_{ob} that is the ratio between the maximum road densities to the observed road density; $P(PA_{ob}>PA_{mn})$ that is the proportion of patches delimited by roads observed within the species range with area (PA_{ob}) larger than the minimal patch size (PA_{mn}). We also indicated for the above species, where within their range RD_{ob} is higher than RD_{mx} and/or $PA_{ob}>PA_{mn}$ using grid cell of 100 x 100km in ArcGis v.10.1. We used equal-area world Mollweide projection given that different road density

measures should represent similar areas independent of their latitudinal position (Safi et al. 2011).

RESULTS

From the 156 turtle species assessed, we observed sixteen species to be most affected by roads ranked by the 5th percentile (Table 1, Figure 2, Figure 3). These species belong to the families Emydidae (five species, corresponding to 12% of the species analyzed in this family), Kinosternidae (one species, 5%), Geoemydidae (six species, 14%), Chelidae (one species, 4%) and Tryonichidae (three species, 33%).

Most of the species we identified for road effect are categorized as Vulnerable, Endangered and Critically Endangered by IUCN. Only 18.7% of the species in the 5th percentile have roads related as one of the major threats (Figure 2). Of the species categorized as Vulnerable and Endangered, *Hardella thurji*, *Heosemys anandalli* and *Pseudemys alabamensis* pointed out by our model are expected to be in danger in 80 % of their distribution range (Table 1). The above mentioned species have distributions throughout Asia and United States, and is within these IUCN status mainly due to over exploitation to commercial issues and habitat loss. Threats related to roads had never been reported before.

Within the 25th percentile coefficient, we identified more 55 species that we expected to be affected by roads. They belong to the families Emydidae (17 species), Geoemydidae (13 species), Chelidae (seven species), Kinosternidae (five species), Tryonichidae (five species), Podocnemidae (five species) and Chelydridae (two species) (Table A4). They are distributed overall in all continents, with most of species in

representative regions such as North and South America and Asia. Of these species, only a low percentage (23.6%) are stated for road threat by IUCN, besides the species already identified at the 5th percentile (Table 1, Figure 2).

Species most affected identified by the 5th percentile and mapped have distributions in Asia (56.3% of the species), North America (37.5%) and Oceania (6.2%), (Figure 4). Species in Asia, for example, *Rafetus euphraticus*, *Hardella thurjii*, *Dogania subplana* and *Amyda cartilaginea* show a strong effect of roads throughout its range. The highest number of species affected by roads per grid cell is observed in Asia (up to four species) and North America (up to three species) (Figure 4).

DISCUSSION

Through directly using life history data, demographic estimates, road mortality, infrastructure information and turtle species distribution in a spatially explicit model, our study provided an approach to predict species that are expected to be affected by roads, ranking species vulnerability and show where these effects are observed. The results of our study show support to the current scenario that roads in general is one of the major threats to turtle species persistence (Gibbs and Shriver 2002; Beaudry et al. 2008; Crawford et al. 2014a; Tuma et al. 2016).

North America has a very remarkable turtle biodiversity, followed by Latin America and Asia (Mittermeier et al. 2015). In many of these places, besides road impact, turtles are facing other threats in overwhelming numbers such as harvest for food, pet trade and traditional medicine, disease and bycatch (Gibbons et al. 2000; Rhodin et al. 2011).

The great turtle biodiversity and high number of priority areas for conservation (Mittermeier et al. 2015) in many of these regions overlap with high road density areas, for instance, in North America. United States is one of the countries with highest road density (Forman and Alexander 1998, Forman 2000) and is the country where we found the highest number of species affected by roads (see Figure 3). Furthermore, we identified many species expected to be affected in the 25 percentile (Table A4; SI) distributed in regions where the road number and extent are projected to expand severely. These places are mainly developing nations such as China, India and Brazil (mainly areas in Amazon) (Laurance et al. 2009; Dulac 2013, Barber et al. 2014). Therefore, other species assessed and not anticipated in the 5th percentile rank of vulnerability might be compromised in a short-term due to road expansion. This is particularly alarming, because a huge turtle biodiversity is found in these areas.

Our estimates are conservative but not without uncertainties. For example, the *RD_{mx}* equation proposals an easy rule of thumb to estimate the maximum road density based on population growth rate (r_1) and mortality on the roads (Borda-de-Água et al. 2012), but it assumes that the population in the suitable habitat has an exponential growth, that may not be the case in many populations (Pereira & Daily 2006). In addition, we also assumed that turtles always die when find a road, but this cannot be always true, i.e. drivers can avoid turtles (Gibbs and Shriver 2002) (but also can roadkill them intentionally, see Ashley et al. 2007). Only a low proportion of individuals can cross safely (about 5%; see Aresco 2004), and roads play an important role acting as a barrier to turtle movement in general (Beaudry et al. 2008). This is mainly due to its pauser behavior

when crossing roads. Turtles usually stop when face a danger, in this case, vehicles on roads that ends with high mortality rates (Smith and Dood-Jr 2003; Aresco 2005b; Andrews et al. 2015; Jacobson et al. 2016). In some cases, road density alone seems not to be the best predictor of population declines (Beaudry et al. 2008). Other features such as road traffic volume and the landscape location of the road network in connection to suitable habitat (Gibbs and Shriver, 2002; Beaudry et al. 2008) can act in concert with road density increasing chances of population extinctions (van Langevelde and Jaarsma 2009). However, little is known at present about these subjects to model them together in a global scale.

Furthermore, higher road density facilitate more human access, hunting and poaching, introduction and establishment of exotic species (Forman and Alexander 1998; Laurance et al 2009), reduces size of suitable patches (Jaeger 2015) and in a landscape perspective, increases annual mortality rates (Gibbs and Shriver 2002). For instance, regions with road density higher than $1\text{km}/\text{km}^2$ and traffic volume higher than 100 vehicles/ lane/day is sufficient to contribute excessively to the annual adult mortality rates of turtles reducing viability and increasing the probability of population extinctions (Gibbs and Shriver, 2002).

By indicating where species are most affected within their ranges, our results can assist many fields of research, identify areas for conservation strategies, where implementing mitigation measures and support decision-makers, for example, during the strategic planning of roads. Many road mitigation measures are currently being applied to turtle conservation (Aresco 2004; 2005b; Dood-Jr et al. 2004), and to successfully implement these measures for multiple taxa the biology and

behavior of the target species should be taken in account (Baxter-Gilbert et al. 2015). For example, crossing structures and fencing constructed prioritizing non-herptile species are rarely effective (Bager and Fontoura, 2013; Baxter-Gilbert et al. 2015). In spite of being one of the most effective measures for reducing mortality and restoring connectivity (Aresco 2004; Andrews et al. 2015) they need to be carefully planned for this taxa (Woltz et al. 2008; Andrews et al. 2008; Clevenger and Huijser, 2011; D’Anuniação et al. 2013). Furthermore, turtles in most cases show road mortality peaks at specific periods (i.e. *hot-moments*) usually related to the beginning of the nesting season (Beaudry et al. 2010; Cureton II and Deaton, 2012; Crawford et al. 2014b; Costa et al. 2015). In addition, temporary mitigation measures such as traffic management, warning road signage and public participation if well planned can be employed with some success in the conservation of turtles (Beaudry et al. 2008; 2010; Andrews et al. 2015).

In summary, our approach demonstrated what species are more prone to be affected by roads and where within their range, they are most affected. Further studies ought to extend our approach to other turtle and herpetile species, because more than half of the recognized species are not even listed in the IUCN. Many of these species may already be in their thresholds of road threat. In addition, novel approaches that include other sources of information, for example, traffic intensity and/or network configuration could be developed in order to assess species response to other road threats. In addition, this study is good start to promote reassessments about the major threats to various turtle species worldwide.

ACKNOWLEDGMENTS

PSL was supported by a PhD grant from Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) in Brazil during the PDSE Doutorado Sanduíche program of internship (Process Number: BEX 4214/14-5). We thank to German Centre for Integrative Biodiversity Research – iDiv staff and associates. To Ramon Gomes de Carvalho for helping with the figures. To Murilo Dantas de Miranda for improvements on the first draft of this manuscript.

REFERENCES

- Andrews, K., Gibbons, W., Jochimsen, D., 2008. Ecological effects of roads on amphibians and reptiles: a literature review. *Herpetol. Conserv.* 3, 121-143.
- Andrews, K.M., Langen, T.A., Struijk, R.P.J.H., 2015. Reptiles: Overlooked but often at risk from roads, in: Van Der Ree, R., Smith, D.J., Grilo, C. (Eds.), *Handbook of Road Ecology*. Wiley Blackwell, pp. 271-280.
- Aresco MJ. 2004. Highway mortality of turtles and other herpetofauna at Lake Jackson, Florida, USA, and the efficacy of a temporary fence/culvert system to reduce roadkills. IN: *Proceedings of the 2003 International Conference on Ecology and Transportation*, Eds. Irwin CL, Garrett P, McDermott KP. Center for transportation and the Environment, North Carolina State University, Raleigh, NC: pp. 433-449.
- Aresco, M.J., 2005a. The effect of sex-specific terrestrial movements and roads on the sex ratio of freshwater turtles. *Biol. Conserv.* 123, 37–44. doi:10.1016/j.biocon.2004.10.006

- Aresco, M.J., 2005b. Mitigation measures to reduce highway mortality of turtles and other herpetofauna at a north Florida lake. *J. Wildl. Manage.* 69, 549–560. doi:10.2193/0022-541X(2005)069[{}0549:MMTRHM]2.0.CO;2
- Ashley, E.P., Kosloski, A., Petrie, S.A., 2007. Incidence of Intentional Vehicle–Reptile Collisions. *Hum. Dimens. Wildl.* 12, 137–143. doi:10.1080/10871200701322423
- Bager, A., Fontoura, V., 2013. Evaluation of the effectiveness of a wildlife roadkill mitigation system in wetland habitat. *Ecol. Eng.* 53, 31–38. doi:10.1016/j.ecoleng.2013.01.006
- Bager, A., Freitas, T.R.O. De, Krause, L., 2007. Nesting ecology of a population of *Trachemys dorbignyi* (Emydidade) in Southern Brazil. *Herpetologica* 63, 56–65. doi:10.1655/0018-0831(2007)63[56:NEOAPO]2.0.CO;2
- Barber, C.P., Cochrane, M.A., Souza, C.M., Laurance, W.F., 2014. Roads, deforestation, and the mitigating effect of protected areas in the Amazon. *Biol. Conserv.* 177, 203–209. doi:10.1016/j.biocon.2014.07.004
- Baxter-Gilbert, J.H., Riley, J.L., Lesbarreres, D., Litzgus, J.D., 2015. Mitigating reptile road mortality: Fence failures compromise ecopassage effectiveness. *PLoS One* 10, 1–15. doi:10.1371/journal.pone.0120537
- Beaudry, F., DeMaynadier, P.G., Hunter Jr., M.L., 2010. Identifying hot moments in road-mortality risk for freshwater turtles. *J. Wildl. Manage.* 74, 152–159. doi:10.2193/2008-370
- Beaudry, F., deMaynadier, P.G., Hunter, M.L., 2008. Identifying road mortality threat at multiple spatial scales for semi-aquatic turtles. *Biol. Conserv.* 141, 2550–2563. doi:10.1016/j.biocon.2008.07.016
- Bickham, J.W., Iverson, J.B., Parham, J.F., Philippen, H.-D., Rhodin,

- A.G.J., Shaffer, H.B., Spinks, P.Q., Dijk, P.P. Van, 2007. Turtle Taxonomy: Methodology, Recommendations, and Guidelines. *Chelonian Res. Monogr.* 4, 73–84.
- Borda-de-Água, L., Navarro, L., Gavinhos, C., Pereira, H.M., 2011. Spatio-temporal impacts of roads on the persistence of populations: analytic and numerical approaches. *Landscape Ecol.* 26, 253–265.
- Ceia-Hasse, A., Borda-de-Água, L., Grilo, C., Pereira, H.M. 2016. Global impact of roads on carnivores. Manuscript submitted for publication.
- Clevenger, A.P., Huijser, M.P., 2011. Wildlife Crossing Structure Handbook: Design and Evaluation in North America 211. doi:FHWA-CFL/TD-11-003
- Congdon, J.D., Dunhan, A.E., van loben Sels, R.C., 1993. Delayed sexual maturity and demographics of Blandingii's turtle (*Emydoidea blandingii*): implications for conservation and management of long-lived organisms. *Cons. Biol.* 7, 826-833.
- Congdon, J.D., Dunhan, A.E., van loben Sels, R.C., 1994. Demographics of the common snapping turtles (*Chelydra serpentina*): implications for conservation and management of long-lived organisms. *Amer. Zool.* 34, 397-408.
- Costa, A.S., Ascensão, F., Bager, A., 2015. Mixed sampling protocols improve the cost-effectiveness of roadkill surveys. *Biodivers. Conserv.* 24, 2953–2965. doi:10.1007/s10531-015-0988-3
- Crawford, B.A., Maerz, J.C., Nibbelink, N.P., Buhlmann, K.A., Norton, T.M., 2014a. Estimating the consequences of multiple threats and management strategies for semi-aquatic turtles. *J. Appl. Ecol.* 51, 359–366. doi:10.1111/1365-2664.12194
- Crawford, B.A., Maerz, J.C., Nibbelink, N.P., Buhlmann, K.A., Norton, T.M., Albeke, S.E., 2014b. Hot spots and hot moments of diamondback terrapin road-crossing activity. *J. Appl. Ecol.* 51, 367–

375. doi:10.1111/1365-2664.12195

- Cureton II, J.C., Deaton, R., 2012. Hot moments and hot spots: Identifying factors explaining temporal and spatial variation in turtle road mortality. *J. Wildl. Manage.* 76, 1047–1052. doi:10.1002/jwmg.320
- D’Anuniação, P.E.R., Lucas, P.S., Silva, V.X., Bager, A., 2013. Road ecology and Neotropical amphibians: Contributions for future studies. *Acta Herpetol.* 8, 129–140. doi:10.13128/Acta_Herpetol-12331
- Dodd Jr, C.K., Barichivich, W.J., Smith, L.L., 2004. Effectiveness of a barrier wall and culverts in reducing wildlife mortality on a heavily traveled highway in Florida. *Biol. Conserv.* 118, 619–631. doi:10.1016/j.biocon.2003.10.011
- Dorland, A., Rytwinski, T., Fahrig, L., 2014. Do roads reduce painted turtle (*Chrysemys picta*) populations? *PLoS One* 9. doi:10.1371/journal.pone.0098414
- Dulac, J., 2013. Global Land Transport Infrastructure Requirements: Estimating road and railway infrastructure capacity and costs to 2050 54.
- Ernst, C.H., Lovich, J. E., 2009. *Turtles of the united states and Canada.* JHU Press.
- Fahrig, L., 2003. Effects of Habitat Fragmentation on Biodiversity. *Annu. Rev. Ecol. Environ. Syst.* 34, 487–515. doi:10.1146/132419
- Flitz, B.A., Mullin, S.J., 2006. Nest-Site Selection in the Eastern Box Turtle, *Terrapene carolina carolina*, in Illinois. *Chelonian Conserv. Biol.* 5, 309–312. doi:10.2744/1071-8443(2006)5[309:NSITEB]2.0.CO;2
- Forman, R.T.T. 2000. Estimate of the Area Affected Ecologically by the Road System in the United States. *Conserv. Biol.* 14, 31–35.

- Forman, R.T.T., Alexander, L.E., 1998. Roads and Their Major Ecological Effects. *Annu. Rev. Ecol. Syst.* 29, 207–231. doi:10.1146/annurev.ecolsys.29.1.207
- Forman, R.T.T., Deblinger, R.D., 1998. The ecological road-effect zone for transportation planning, and a Massachusetts highway example, in: G. L. Evink, P. Garrett, D. Zeigler, J. Berry, (Eds.). Proceedings of the international conference on wildlife ecology and transportation. Publication FL-ER-69-98. Florida Department of Transportation, Tallahassee, pp. 78-96.
- Fujita, M.K., Engstrom, T.N., Starkey, D.E., Shaffer, H.B., 2004. Turtle phylogeny: Insights from a novel nuclear intron. *Mol. Phylogenet. Evol.* 31, 1031–1040. doi:10.1016/j.ympev.2003.09.016
- Gibbon, J.W., Scott, D.E., Ryan, T.J., Buhlmann, K. a., Tuberville, T.D., Metts, B.S., Greene, J.L., Mills, T., Leiden, Y., Poppy, S., Winne, C.T., 2000. The Global Decline of Reptiles, Déjà Vu Amphibians. *Bioscience* 50, 653–666. doi:10.1641/0006-3568(2000)050[0653:TGDORD]2.0.CO;2
- Gibbs, J.P., Shriver, W.G., 2002. Estimating the effects of road mortality on turtle populations. *Conserv. Biol.* 16, 1647–1652. doi:10.1046/j.1523-1739.2002.01215.x
- Gibbs, J.P., Steen, D.A., 2005. Trends in sex ratios of turtles in the United States: Implications of road mortality. *Conserv. Biol.* 19, 552–556. doi:10.1111/j.1523-1739.2005.000155.x
- Gonçalves, F. a., Cechin, S.Z., Bager, A., 2007. Predação de ninhos de *Trachemys dorbigni* (Duméril & Bibron) (Testudines, Emydidae) no extremo sul do Brasil. *Rev. Bras. Zool.* 24, 1063–1070. doi:10.1590/S0101-81752007000400025
- Goosem, M., 2007. Fragmentation impacts caused by roads through rainforests. *Curr. Sci.* 93, 1587–1595.

- Guillon, J.M., Guéry, L., Hulin, V., Girondot, M., 2012. A large phylogeny of turtles (Testudines) using molecular data. *Contrib. to Zool.* 81, 147–158.
- Hamer, A.J., Harrison, L.J., Stokeld, D., 2016. Road density and wetland context alter population structure of a freshwater turtle. *Austral Ecol.* 41, 53–64. doi:10.1111/aec.12298
- Heilman, G.E., Strittholt, J.R., Slosser, N.C., Dellasala, D.A., 2002. Forest Fragmentation of the Conterminous United States: Assessing Forest Intactness through Road Density and Spatial Characteristics: Forest fragmentation can be measured and monitored in a powerful new way by combining remote sensing, geographic informat. *Bioscience* 52, 411–422. doi:10.1641/0006-3568(2002)052[0411:ffotcu]2.0.co;2
- Ihlow, F., Dambach, J., Engler, J.O., Flecks, M., Hartmann, T., Nekum, S., Rajaei, H., Rödder, D., 2012. On the brink of extinction? How climate change may affect global chelonian species richness and distribution. *Glob. Chang. Biol.* 18, 1520–1530. doi:10.1111/j.1365-2486.2011.02623.x
- Irwin, L., Irwin, K., 2006. Global threats affecting the status of reptile populations, in: Gardner, S.C., Oberdörster, E., (Eds.), *Toxicology of reptiles*. Taylor and Francis, pp. 9-34.
- Iverson, J.B., 1992. A Revised Checklist with Distribution Maps of the Turtles of the World 363.
- Jacobson, S.L., Bliss-ketchum, L.L., Rivera, C.E. De, Smith, W.P., 2016. A behavior- based framework for assessing barrier effects to wildlife from vehicle traffic volume. *Ecosphere* 7, 1–15. doi:10.1002/ecs2.1345
- Jaeger, J.A.G., 2015. Improving Environmental Impact Assessment and Road Planning at the Landscape Scale, in: Van Der Ree, R., Smith, D.J., Grilo, C. (Eds.), *Handbook of Road Ecology*. Wiley Blackwell,

pp. 32-42. doi:10.1002/9781118568170.ch5

- Jaeger, J.A.G., Holderegger, R., 2005. Thresholds of landscape fragmentation (in German: Schwellenwerte der Landschaftszerschneidung). *GAIA*. 14, 113-118.
- Jaeger, J.A.G., Bowman, J., Brennan, J., Fahrig, L., Bert, D., Bouchard, J., Charbonneau, N., Frank, K., Gruber, B., Von Toschanowitz, K.T., 2005. Predicting when animal populations are at risk from roads: An interactive model of road avoidance behavior. *Ecol. Modell.* 185, 329–348.
- Jones, R.L., 2006. Reproduction and nesting of the endangered ringed map turtle, *Graptemy oculifera*, in Mississippi. *Chelonian Cons. Biol.* 5, 195-209.
- Kazmaier, R.T., Hellgreen, E.C., Ruthven, D. C., 2002. Home range and dispersal of texas tortoise *Gopherus berlandieri*, in a managed Thornscrub ecosystem. *Chelonian Cons. Biol.* 4, 488-496.
- Laurance, W. F., Cochrane, M. A., Bergen, S., Fearnside, P. M., Delamônica, P., Barber, C., D'Angelo, S., Fernandes, T., 2001. The future of the Brazilian Amazon. *Science*. 291, 438-439.
- Laurance, W.F., Clements, G.R., Sloan, S., O'Connell, C.S., Mueller, N.D., Goosem, M., Venter, O., Edwards, D.P., Phalan, B., Balmford, A., Van Der Ree, R., Arrea, I.B., 2014. A global strategy for road building. *Nature* 513, 229–232. doi:10.1038/nature13717
- Laurance, W.F., Goosem, M., Laurance, S.G.W., 2009. Impacts of roads and linear clearings on tropical forests. *Trends Ecol. Evol.* 24, 659–669. doi:10.1016/j.tree.2009.06.009
- Lee, P.F., Ding, T.S., Hsu, F.H., Geng, S., 2004. Breeding bird species richness in Taiwan: distribution on gradients of elevation, primary productivity and urbanization. *J. Biogeogr.* 31, 307–314. doi:10.1046/j.0305-0270.2003.00988.x

- Litzgus, J.D., Bolton, F., Schulte-Hostedde, A.I., 2008. Reproductive Output Depends on Body Condition in Spotted Turtles (*Clemmys Guttata*). *Copeia* 2008, 86–92. doi:10.1643/CH-07-093
- Mech, L. D., Fritts, S. H., Radde, G. L., Paul, W. J., 1988. Wolf distribution and road density in Minnesota. *Wild. Soc. Bull.* 16, 85-87.
- Mittermeier, R.A., van Dijk, P.P., Rhodin, A.G.J., Nash, S.D., 2015. Turtle Hotspots: An Analysis of the Occurrence of Tortoises and Freshwater Turtles in Biodiversity Hotspots, High-Biodiversity Wilderness Areas, and Turtle Priority Areas. *Chelonian Conserv. Biol.* 14, 2–10. doi:10.2744/ccab-14-01-2-10.1
- Newbold, T., Hudson, L.N., Hill, S.L.L., Contu, S., Lysenko, I., Senior, R.A., Börger, L., Bennett, D.J., Choimes, A., Collen, B., Day, J., De Palma, A., Díaz, S., Echeverria-Londoño, S., Edgar, M.J., Feldman, A., Garon, M., Harrison, M.L.K., Alhousseini, T., Ingram, D.J., Itescu, Y., Kattge, J., Kemp, V., Kirkpatrick, L., Kleyer, M., Correia, D.L.P., Martin, C.D., Meiri, S., Novosolov, M., Pan, Y., Phillips, H.R.P., Purves, D.W., Robinson, A., Simpson, J., Tuck, S.L., Weiher, E., White, H.J., Ewers, R.M., Mace, G.M., Scharlemann, J.P.W., Purvis, A., 2015. Global effects of land use on local terrestrial biodiversity. *Nature* 520, 45–50. doi:10.1038/nature14324
- Pereira, H. M., Daily, G. C., Roughgarden, J., 2004. A framework for assessing the relative vulnerability of species to land-use change. *Ecological Applications.* 14, 730-742.
- Pereira, H.M., Daily, G.C., 2006. Modeling biodiversity dynamics in countryside landscapes. *Ecology* 87, 1877–1885. doi:10.1890/0012-9658(2006)87[1877:MBDICL]2.0.CO;2..
- Pereira, H.M., Navarro, L.M., Martins, I.S., 2012. Global biodiversity change: the bad, the good, and the unknown. *Annu. Rev. Environ. Resour.* 37, 25–50. doi:10.1146/annurev-environ-042911-093511
- Peters, A.; Verhoeven, K.J.F., Strijbosch, H., 1994. Hatching and emergence in the turkish mediterranean loggerhead turtle, *Caretta*

- caretta- natural causes for egg and hatchling failure. *Herpetologica*. 1994, 369-373.
- Pike, D.A., Pizzatto, L., Pike, B.A., Shine, R., 2008. Estimating survival rates of uncatchable animals: The myth of high juvenile mortality in reptiles. *Ecology* 89, 607–611. doi:10.1890/06-2162.1
- Reid, B.N., Peery, M.Z., 2014. Land use patterns skew sex ratios, decrease genetic diversity and trump the effects of recent climate change in an endangered turtle. *Divers. Distrib.* 20, 1425–1437. doi:10.1111/ddi.12243
- Rhodin, A.G.J., Walde, A.D., Horne, B.D., Van Dijk, P.P., Blanck, T., and Hudson, R., 2011. *Turtles in Trouble: The World's 25+ Most Endangered Tortoises and Freshwater Turtles*. Public 54. doi:10.1071/EC12189
- Roe, J.H., Brinton, A.C., and Georges, A. 2009. Temporal and spatial variation in landscape connectivity for a freshwater turtle in a temporally dynamic wetland system. *Ecol. Applic.* 19, 1288-1299.
- Safi, K., Cianciaruso, M.V., Loyola, R.D., Brito, D., Armour-Marshall, K., Diniz-Filho, J.A.F., 2011. Understanding global patterns of mammalian functional and phylogenetic diversity. *Philos. Trans. R. Soc. London* 366, 2536–2544. doi:10.1098/rstb.2011.0024
- Skellam, J.G., 1951. Random Dispersal in Theoretical Populations. *Biometrika*. doi:10.1093/biomet/38.1-2.196
- Smith, L.L., Dodd Jr., C.K., 2003. Wildlife mortality on highway US 441 across Paynes Prairie, Alachua County, Florida. *Florida Scientist* 66, 128–140.
- Steen, D.A., Gibbs, J.P., 2004. Effects of roads on the structure of freshwater turtle populations. *Conserv. Biol.* 18, 1143–1148. doi:10.1111/j.1523-1739.2004.00240.x

- Stephens, P. A., Sutherland, W. J., Freckleton, R. P., 1999. What is the Allee effect?. *Oikos*. 87, 185-190.
- Todd, B. D., Willson, J. D., Gibbons, J. W., 2010. The global status of reptiles and causes of their decline, in: Sparling, D. W., Linder, G., Bishop, C. A., Krest, S. (Eds.), *Ecotoxicology of amphibians and reptiles*. CRC Press, pp. 47-67.
- Tinkle, D.W., Congdon, J.D., Rosen, P.C., 1981. Nesting frequency and success: implications for the demography of painted turtles. *Ecology*. 62, 1426-1432.
- Tuma, M.W., Millington, C., Schumaker, N., Burnett, P., 2016. Modeling Agassiz's desert tortoise population response to anthropogenic stressors. *J. Wildl. Manage.* 80, 414–429. doi:10.1002/jwmg.1044
- Van Der Grift, E.A., 2005. Defragmentation in the Netherlands: A success story? *Gaia* 14, 144–147.
- van der Ree, R., Jaeger, J.A.G., van der Grift, E.A., Clevenger, A.P., 2011. Effects of roads and traffic on wildlife populations and landscape function: Road ecology is moving toward larger scales. *Ecol. Soc.* 16. doi:48
- van Dijk, P.P., Iverson, J.B., Rhodin, A.G.J., Shaffer, H.B., Bour, R., 2014. Turtles of the World, 7th Edition: Annotated Checklist of Taxonomy, Synonymy, Distribution with Maps, and Conservation Status. *Conserv. Biol. Freshw. Turtles Tortoises* 5, 243–328. doi:10.3854/crm.5.000.checklist.v7.2014
- van Langevelde, F., Jaarsma, C.F., 2009. Modeling the effect of traffic calming on local animal population persistence. *Ecol. Soc.* 14. doi:39
- Woltz, H.W., Gibbs, J.P., Ducey, P.K., 2008. Road crossing structures for amphibians and reptiles: Informing design through behavioral analysis. *Biol. Conserv.* 141, 2745–2750. doi:10.1016/j.biocon.2008.08.010

List of Tables

Table 1: Turtle species most affected by minimal patch size. $P [PA_{ob} > PA_{mn}]$ are the lowest values inside the 5th percentile. IUCN Red List status are CR: Critically Endangered, EN: Endangered, LC: Least Concern and VU: Vulnerable.

Species	Common name	Family	P[PAobs > PAcrit]	%range affected	IUCN status
<i>Hardella thurjii</i>	Crowned River Turtle	Geoe.	0.03	80	VU
<i>Batagur borneoensis</i>	Painted Terrapin	Geoe.	0.06	42	CR
<i>Batagur baska</i>	Common Batagur	Geoe.	0.07	58	CR
<i>Rafetus euphraticus</i>	Euphrates Softshell Turtle	Tryo.	0.07	75	EN
<i>Dogania subplana</i>	Malayan Soft-shelled Turtle	Tryo.	0.12	50	LC
<i>Pseudemys alabamensis</i>	Alabama Red-bellied Cooter	Emyd.	0.13	80	EN
<i>Heosemys annandalii</i>	Yellow-headed Temple Turtle	Geoe.	0.14	80	EN
<i>Amyda cartilaginea</i>	Asiatic Softshell Turtle	Tryo.	0.18	63	VU

List of Figures

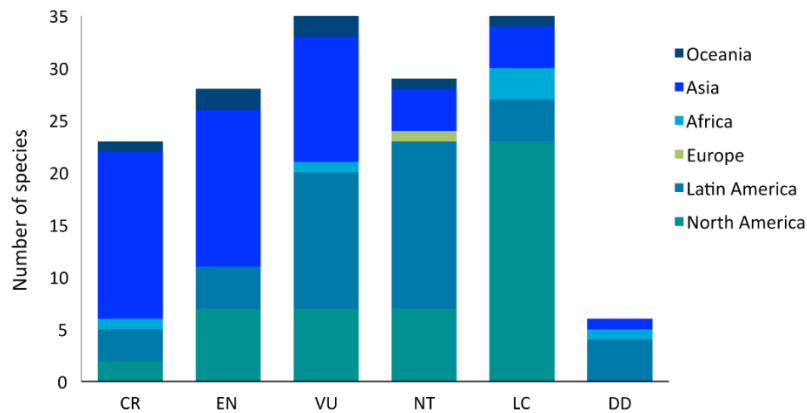


Figure 1: Number of species assessed classified by IUCN status and their distribution within regions worldwide.

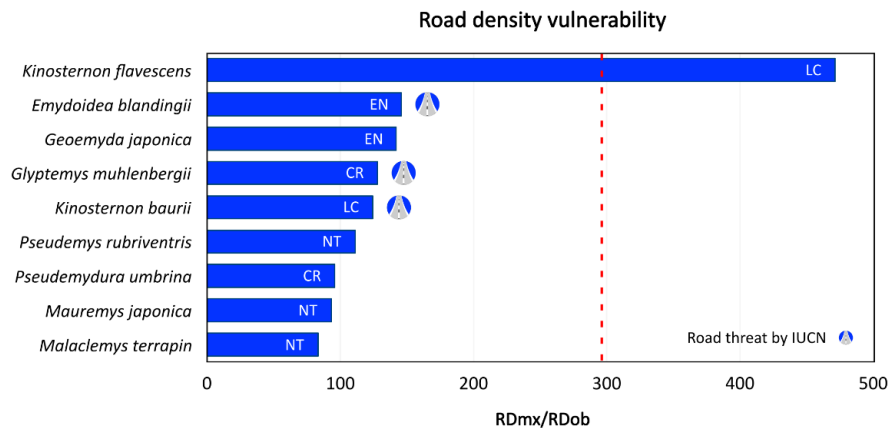


Figure 2: Turtle species most affected by road density. Values of RD_{mx}/RD_{ob} comparing species within the 5th percentile with a species not affected (*Kinosternon flavescens*). Dashed red line include the boundary of road density values for all the species within the 25th percentile. LC = least concern, EN = endangered, CR = critically endangered, NT = near threatened status in IUCN.

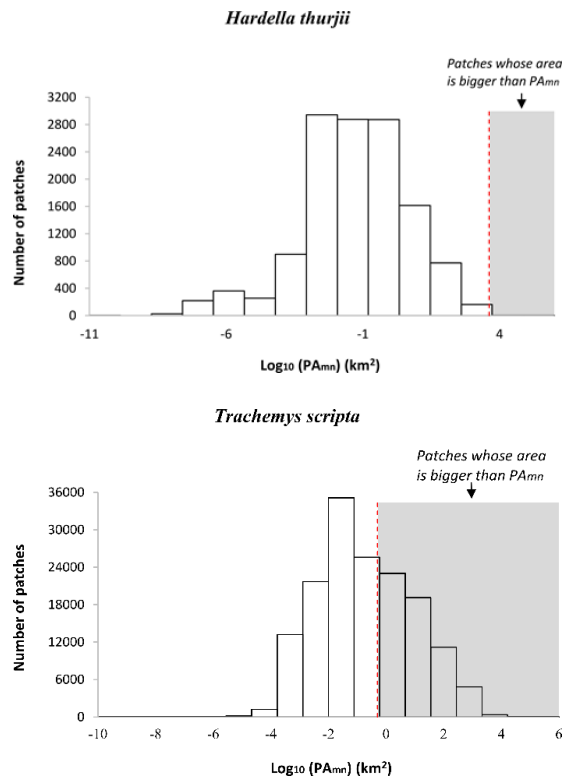


Figure 3: Turtle species most affected by patch size within the 5th percentile. *Hardella thurjii* is among the 5% species with lower $P[PA_{ob} > PA_{mn}]$. In contrast, *Trachemys scripta* has high $P[PA_{ob} > PA_{mn}]$. Fewer patches larger than PA_{min} (areas shaded in grey) reveal a high vulnerability for *H. thurjii*.

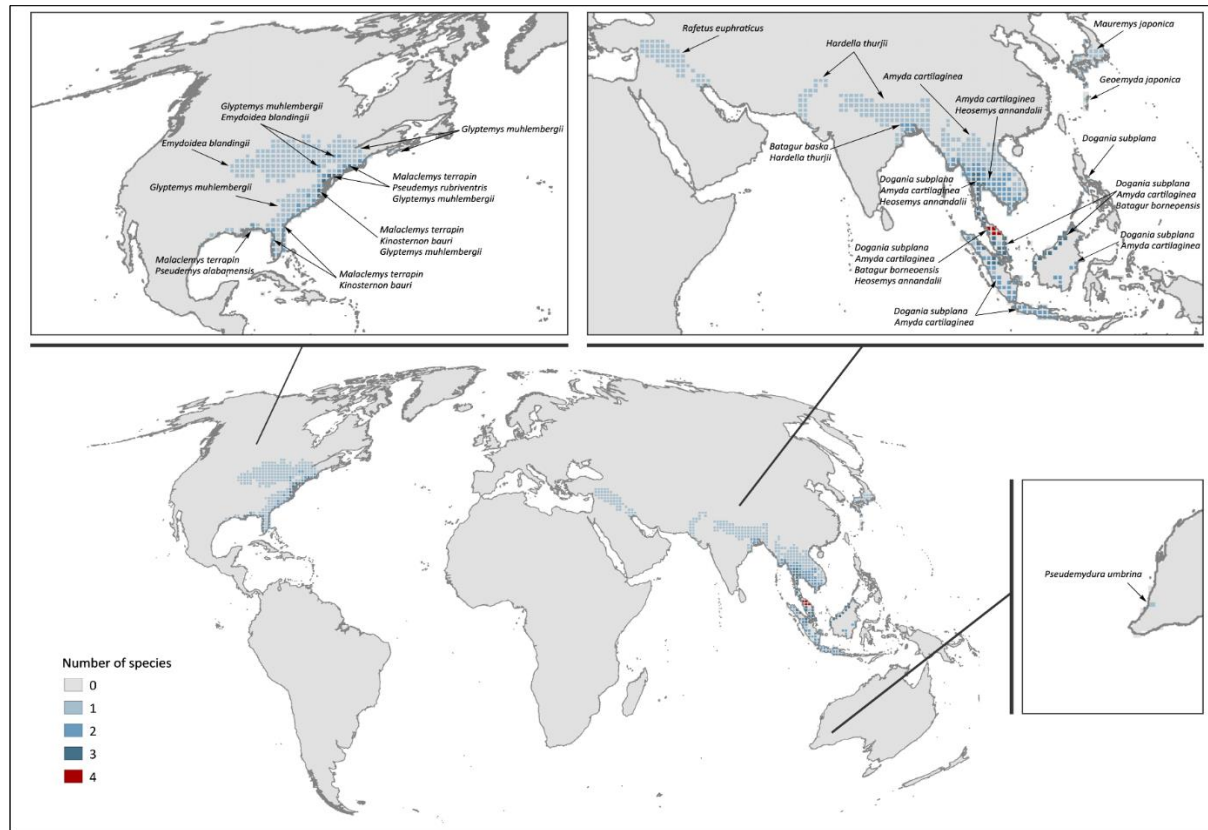


Figure 4: Number of turtle species affected within 100 x 100 km grid cell where RD_{ob} is higher than RD_{mx} and/or $PA_{ob} > PA_{mm}$.

Appendix A. Supporting Information

Skellam Model

This model describes population dynamic in a landscape with two types of habitat: patches of suitable habitat which population growth rate is positive (r_1), roads as unsuitable habitat in which population growth rate is negative (r_0) and the dispersal variance modeled by individual dispersal distances (σ^2) (Pereira et al. 2004, Pereira & Daily 2006; Borda-de-Água et al 2012). The dynamics are given by:

$$\frac{dN(x, y, t)}{dt} = \begin{cases} \frac{S^2}{2} \nabla^2 N(x, y, t) + r_1 N(x, y, t) \left(1 - \frac{N(x, y, t)}{K}\right) & \text{if } (x, y) \notin \text{road} \\ \frac{S^2}{2} \nabla^2 N(x, y, t) + r_0 N(x, y, t) & \text{if } (x, y) \in \text{road} \end{cases}$$

Where $N(x, y, t)$ is the population density at location (x, y) at time t , K is the carrying capacity, and ∇^2 is $(d^2/dx^2 + d^2/dy^2)$. The first term on the right hand side of the equations (top and bottom branches) describes the changes in population density in space and time on the basis of the dispersal distance, which is assumed to follow a Gaussian distribution. The second term describes the logistic growth outside roads (top branch), and the population decay (since r_0 is assumed to be negative) on roads (bottom branch) (Borda-de-Água et al. 2012).

Table A1: Estimates of nest mortality rates (NMR) based on available literature.

Species	NMR (%)	Reference
<i>Chelydra serpentina</i>	77	Congdon et al. 1994
<i>Chrysemys picta</i>	30	Spencer and Jensen, 2010
<i>Clemmys guttata</i>	32	Litzgus and Mousseau, 2006
<i>Emydoidea blandingii</i>	74	Congdon et al. 1993
<i>Emys orbicularis</i>	75	Zuffi, 2000
<i>Glyptemys insculpta</i>	31	Walde et al. 2007
<i>Graptemys ernsti</i>	90	Ernst and Lovich 2009
<i>Graptemys flavimaculata</i>	89	Horne et al. 2003
<i>Graptemys oculifera</i>	86	Jones, 2006
<i>Kinosternon subrubrum</i>	84	Burke et al. 1998
<i>Kinosternon flavescens</i>	79	Iverson, 1991
<i>Malaclemys terrapin</i>	46-69	Burger, 1977; Butler et al. 2004
<i>Podocnemis expansa</i>	5.31	Salera-Jr. et al. 2009
<i>Podocnemis unifilis</i>	37	Pignati et al. 2013
<i>Stenotherus odoratus</i>	78	Ernst and Lovich 2009
<i>Terrapene carolina</i>	87.5	Flitz and Mullin, 2006
<i>Trachemys scripta</i>	84-98	Burke et al. 1998; Aresco, 2004

Literature Cited

- Aresco, M. J., 2004. Reproductive ecology of *Pseudemys floridana* and *Trachemys scripta* (Testudines: Emydidae) in northwestern Florida. *J. Herpet.* 38, 249-256.
- Burger, J., 1977. Determinants of hatching success in diamondback terrapin, *Malaclemys terrapin*. *Am. Midland Nat.* 1977, 444-464.
- Burke, V. J., Rathbun, S. L., Bodie, J. R., Gibbons, J. W., 1998. Effect of density on predation rate for turtle nests in a complex landscape. *Oikos*, 3-11.
- Butler, J. A., Broadhurst, C., Green, M., Mullin, Z., 2004. Nesting, nest predation and hatchling emergence of the Carolina diamondback terrapin, *Malaclemys terrapin centrata*, in northeastern Florida. *Am. Midland Nat.* 152, 145-155.
- Congdon, J.D., Dunhan, A.E., van loben Sels, R.C., 1993. Delayed sexual maturity and demographics of Blandingii's turtle (*Emydoidea blandingii*): implications for conservation and management of long-lived organisms. *Cons. Biol.* 7, 826-833.
- Congdon, J.D., Dunhan, A.E., van loben Sels, R.C., 1994. Demographics of the common snapping turtles (*Chelydra*

- serpentina): implications for conservation and management of long-lived organisms. *Amer. Zool.* 34, 397-408.
- Ernst, C.H., Lovich, J. E., 2009. *Turtles of the united states and Canada.* JHU Press.
- Flitz, B.A., Mullin, S.J., 2006. Nest-Site Selection in the Eastern Box Turtle, *Terrapene carolina carolina*, in Illinois. *Chelonian Conserv. Biol.* 5, 309–312. doi:10.2744/1071-8443(2006)5[309:NSITEB]2.0.CO;2
- Horne, B. D., Brauman, R. J., Moore, M. J., Seigel, R. A., 2003. Reproductive and nesting ecology of the yellow-blotched map turtle, *Graptemys flavimaculata*: implications for conservation and management. *Copeia*, 2003, 729-738.
- Iverson, J. B., 1991. Life history and demography of the yellow mud turtle, *Kinosternon flavescens*. *Herpetologica*, 373-395.
- Jones, R.L., 2006. Reproduction and nesting of the endangered ringed map turtle, *Graptemy oculifera*, in Mississippi. *Chelonian Cons. Biol.* 5, 195-209.
- Litzgus, J. D., & Mousseau, T. A., 2006. Geographic variation in reproduction in a freshwater turtle (*Clemmys guttata*). *Herpetologica*, 62, 132-140.
- Pignati, M. T., Fernandes, L. F., Miorando, P. S., Ferreira, P. D., Pezzuti, J. C. B., 2013. Nesting site and hatching success of *Podocnemis unifilis* (Testudines: Podocnemididae) in a floodplain area in lower Amazon River, Para, Brazil. *S. Am. J. Herpet.* 8, 175-185.
- Salera-Jr., G., Malvasio, A., Portelinha, T. C. G., 2009. Evaluation of predation in *Podocnemis expansa* and *Podocnemis unifilis* (Testudines, Podocnemididae) in the Javaés River, Tocantins. *Acta Amazonica.* 39, 207-213.
- Spencer, R. J., & Janzen, F. J., 2010. Demographic consequences of adaptive growth and the ramifications for conservation of long-lived organisms. *Biol. Con.* 143, 1951-1959.
- Walde, A.D., Bider, J.R., Masse, D., Saumure, R.A., Titman, R.D., 2007. Nesting ecology and hatching success of the wood turtle, *Glyptemys insculpta*, in Quebec. *Herpet. Cons. Biol.* 2, 49-60.
- Zuffi, M. A. L., 2000. Conservation biology of the European pond turtle *Emys orbicularis* (L.) in Italy. *Stapfia.* 69, 219-228.

Table A2: References used to estimate population growth rate on unsuitable habitat (r_0) based on road kill mortality data.

Road kill average rate (RAR) as individual/km/year.

Species	RAR	Reference
<i>Apalone ferox</i>	3.33	Smith and Dood Jr. 2003; Aresco 2005
<i>Apalone spinifera</i>	0.01	McCallum 2011
<i>Acanthochelys spixii</i>	0.04	Coelho et al. 2008
<i>Clemmys guttata</i>	0.63	Ashley and Robinson 1996; MacKinnon et al. 2005
<i>Chelydra serpentina</i>	3.07	Ashley and Robinson 1996; Smith and Dood Jr 2003, Aresco 2005; MacKinnon et al 2005; Glista et al 2008; Glista & Devault 2008; Shepard et al. 2008; McCallum 2011
<i>Chrysemys picta</i>	9.48	Ashley and Robinson 1996; MacKinnon et al. 2005; Glista et al. 2008; Glista and Devault 2008; Shepard et al. 2008
<i>Emydoidea blandingii</i>	2.28	Ashley and Robinson 1996; MacKinnon et al. 2005
<i>Emys orbicularis</i>	0.12	Tok et al. 2011; Kambourova-Ivanova et al. 2012
<i>Graptemys geographica</i>	0.62	Ashley and Robinson 1996; MacKinnon et al. 2005; Glista et al. 2008
<i>Kinosternon bauri</i>	12.34	Smith and Dood Jr 2003; Dood Jr et al. 2004
<i>Kinosternon subrubrum</i>	5.92	Smith and Dood Jr 2003; Aresco 2005
<i>Malaclemys terrapin</i>	20.79	Wood and Herlands 1997; Szerlag and McRobert 2006; Crawford et al. 2014b;
<i>Pseudemys concinna</i>	0.07	Cureton II and Deaton 2012
<i>Pseudemys nelsoni</i>	0.62	Smith and Dood Jr 2003; Dood Jr et al. 2004
<i>Sternotherus odoratus</i>	5.22	Smith and Dood Jr 2003; Aresco 2005; MacKinnon et al. 2005; Shepard et al. 2008
<i>Terrapene carolina</i>	0.25	Smith and Dood Jr 2003; Aresco 2005; Glista et al. 2007; Shepard et al. 2008; McCallum 2011
<i>Terrapene ornata</i>	0.007	Shepard et al. 2008
<i>Trachemys scripta</i>	11.9	Smith and Dood Jr 2003; Aresco 2005; Glista et al. 2008; Shepard et al. 2008

Literature cited

- Aresco, M.J., 2005b. Mitigation measures to reduce highway mortality of turtles and other herpetofauna at a north Florida lake. *J. Wildl. Manage.* 69, 549–560. doi:10.2193/0022-541X(2005)069[{}0549:MMTRHM]2.0.CO;2
- Ashley, E. P., Robinson, J. T., 1996. Road mortality of amphibians, reptiles and other wildlife on the Long Point Causeway, Lake Erie, Ontario. *Can. Field Nat.* 110, 403–412.
- Coelho, I. P., Kindel, A., Coelho, A. V. P., 2008. Roadkills of vertebrate species on two highways through the Atlantic Forest Biosphere Reserve, southern Brazil. *Eur. J. Wild. Res.* 54, 689–699.
- Crawford, B.A., Maerz, J.C., Nibbelink, N.P., Buhlmann, K.A., Norton, T.M., Albeke, S.E., 2014b. Hot spots and hot moments of diamondback terrapin road-crossing activity. *J. Appl. Ecol.* 51, 367–375. doi:10.1111/1365-2664.12195
- Cureton II, J.C., Deaton, R., 2012. Hot moments and hot spots: Identifying factors explaining temporal and spatial variation in turtle road mortality. *J. Wildl. Manage.* 76, 1047–1052. doi:10.1002/jwmg.320
- Dodd Jr, C.K., Barichivich, W.J., Smith, L.L., 2004. Effectiveness of a barrier wall and culverts in reducing wildlife mortality on a heavily traveled highway in Florida. *Biol. Conserv.* 118, 619–631. doi:10.1016/j.biocon.2003.10.011
- Glista, D. J., DeVault, T. L., DeWoody, J. A., 2008. Vertebrate road mortality predominantly impacts amphibians. *Herpet. Cons. Biol.* 3, 77–87.
- Glista, D. J., DeVault, T. L., 2008. Road mortality of terrestrial vertebrates in Indiana. In: *Proceedings of the Indiana Academy of Science.* 117, 55–62.
- Kambourova-Ivanova, N., Koshev, Y., Popgeorgiev, G., Ragyov, D., Pavlova, M., Mollov, I., Nedialkov, N., 2012. Effect of traffic on mortality of amphibians, reptiles, birds and mammals on two types of roads between Pazardzhik and Plovdiv Region (Bulgaria)—preliminary results. *Acta Zool. Bulgarica.* 64, 57–67.
- McCallum, M. L. 2011. Road mortality of turtles and bullfrogs during

- a major flood. *Herpet. Notes.* 4, 183-186.
- MacKinnon, C. A., Moore, L. A., Brooks, R. J., 2005. Why did the reptile cross the road? Landscape factors associated with road mortality of snakes and turtles in the South Eastern Georgian Bay area, in: *Proceedings of the Parks Research Forum of Ontario (PRFO) and Carolinian Canada Coalition (CCC) Annual General Meeting*, pp. 153-166.
- Shepard, D. B., Dreslik, M. J., Jellen, B. C., Phillips, C. A., 2008. Reptile road mortality around an oasis in the Illinois corn desert with emphasis on the endangered eastern massasauga. *Copeia.* 2008, 350-359.
- Smith, L.L., Dodd Jr., C.K., 2003. Wildlife mortality on highway US 441 across Paynes Prairie, Alachua County, Florida. *Florida Scientist* 66, 128-140.
- Szerlag, S., McRobert, S. P., 2006. Road occurrence and mortality of the northern diamondback terrapin. *Applied Herpet.* 3, 27-37.
- Tok, C. V., Ayaz, D., Cicek, K., 2011. Road mortality of amphibians and reptiles in the Anatolian part of Turkey. *Turk. J. Zool.* 35, 851-857.
- Wood, R. C., Herlands, R., 1997. Turtles and tires: the impact of roadkills on northern diamondback terrapin, *Malaclemys terrapin* terrapin, populations on the Cape May Peninsula, southern New Jersey, USA. In: *Proceedings: Conservation, Restoration, and Management of Tortoises and Turtles-An International Conference.* 1, 46-53.

Table A3: Life history data for turtle species. We indicate carapace length (CL) (mm), age at first breeding (yr), clutch size, clutch frequency, EPY = eggs per year, Disp = mean dispersal (km), LF = life span (yr) and IUCN Red List Status. Families are truncated to the first five letters and are: Carettochelyidae, Chelidae, Chelydridae, Emydidae, Geoemydidae, Kinosternidae, Pelomedusidae, Platysternidae, Podocnemidae, Staurotypidae and Tryonichidae.

Species	Family	CL	Breed.			EPY	Disp.	LF	IUCN
			Age	CS	CF				
<i>Acanthochelys macrocephala</i>	Cheli.	162.0		6		6			NT
<i>Acanthochelys pallidipectoris</i>	Cheli.	144.0		5	1	5			VU
<i>Acanthochelys radiolata</i>	Cheli.	174.0		3.3		3.3			NT
<i>Acanthochelys spixii</i>	Cheli.	139.8		4.4		4.4			NT
<i>Actinemys marmorata</i>	Emydi.	129.0		3	1	3	1	27.3	VU
<i>Amyda cartilaginea</i>	Tryon.	700.0	3	20		37.6			VU
<i>Apalone ferox</i>	Tryon.	339.3		16.14		16.14		26.2	LC
<i>Apalone mutica</i>	Tryon.	211.0	9	10.4	1	10.4	4	20	LC
<i>Apalone spinifera</i>	Tryon.	372.5	8	31.5		31.5		25.5	LC
<i>Batagur baska</i>	Geoem.	488.0		26		50.96		21.7	CR
<i>Batagur borneoensis</i>	Geoem.	466.0		11.4		22.34			CR
<i>Batagur dhongoka</i>	Geoem.	420.0		23		45.08			EN
<i>Batagur kachuga</i>	Geoem.	560.0		17		33.32			CR
<i>Batagur trivittata</i>	Geoem.	483.5				27.09			EN
<i>Carettochelys insculpta</i>	Caret.	457.0	14	15	2	30		38.4	VU
<i>Chelodina mccordi</i>	Cheli.	241.0		9.9		16.43			CR
<i>Chelodina novaeguineae</i>	Cheli.	193.0		10		16.6			LC
<i>Chelodina oblonga</i>	Cheli.	213.5	6.2	12	1	12			NL
<i>Chelodina parkeri</i>	Cheli.	220.0		8	2	16			VU
<i>Chelodina pritchardi</i>	Cheli.	214.1				11.83			EN
<i>Chelodina reimanni</i>	Cheli.	203.0		15	2	30			NT
<i>Chelydra rossignonii</i>	Chely.	370.0		30		30			VU
<i>Chelydra serpentina</i>	Chely.	284.0	10	28.5	1	28.5		47	LC
<i>Chrysemys picta</i>	Emydi.	182.0	6	13.9	2	27.8		61	LC
<i>Claudius angustatus</i>	Staur.	104.0	4	4	2	8		16.1	NT
<i>Clemmys guttata</i>	Emydi.	103.7	7	3.58	2	7.16	0.60	30	EN
<i>Cuora amboinensis</i>	Geoem.	175.0	5	1	3	3		38.2	VU
<i>Cuora aurocapitata</i>	Geoem.	172.7				7.28			CR
<i>Cuora flavomarginata</i>	Geoem.	152.0	6	2	2	4		19.1	EN
<i>Cuora galbinifrons</i>	Geoem.	180.0	10	3	1	3			CR
<i>Cuora mccordi</i>	Geoem.	137.1		1.5	3	4.5			CR
<i>Cuora mouhotii</i>	Geoem.	160.0		2.4		5.18			EN

<i>Cuora pani</i>	Geoem.	172.7				7.28			CR
<i>Cuora trifasciata</i>	Geoem.	230.0		5	2	10		31.8	CR
<i>Cuora yunnanensis</i>	Geoem.	175.0		4	2	8			CR
<i>Cuora zhoui</i>	Geoem.	172.7				7.28			CR
<i>Cyclenys dentata</i>	Geoem.	200.0		3		5.88		14.7	NT
<i>Dogania subplana</i>	Tryon.			7		13.16			LC
<i>Elseya branderhorsti</i>	Cheli.					10.68			VU
<i>Elseya novaeguineae</i>	Cheli.					10.68			LC
<i>Elusor macrurus</i>	Cheli.	317.0	15	14.7	1	14.7			EN
<i>Emydoidea blandingii</i>	Emydi.	209.0	14	12.5	2	25		77	EN
<i>Emydura subglobosa</i>	Cheli.	210.0		10.25		13.12			LC
<i>Emys orbicularis</i>	Emydi.	109.0		5	2	10		19.8	NT
<i>Erymnochelys madagascariensis</i>	Podoc.	345.0	18	13	3	39			CR
<i>Geoemyda japonica</i>	Geoem.	155.0		4	1	4			NT
<i>Geoemyda spengleri</i>	Geoem.	101.0		1	3	3			DD
<i>Glyptemys insculpta</i>	Emydi.	198.3	14	11	1	11		60	CR
<i>Glyptemys muhlenbergii</i>	Emydi.	91.8	7	4	1	4		40	CR
<i>Graptemys barbouri</i>	Emydi.	220.0	14	8.5	3	25.5		31.7	VU
<i>Graptemys caglei</i>	Emydi.	213.0		6	2	12		14.5	EN
<i>Graptemys ernsti</i>	Emydi.	212.0	14	7.2	4	28.8			NT
<i>Graptemys flavimaculata</i>	Emydi.	206.0	10	4.7	2	9.4			VU
<i>Graptemys geographica</i>	Emydi.	226.0	12	10.1	2	20.2		3.31	LC
<i>Graptemys gibbonsi</i>	Emydi.	295.0	15	7.5	2	15			EN
<i>Graptemys nigrinoda</i>	Emydi.	155.0	8	5.5	3	16.5		20.3	LC
<i>Graptemys oculifera</i>	Emydi.	148.0	7	3	2	6		20.3	VU
<i>Graptemys ouachitensis</i>	Emydi.	205.0	6	10.5	2	21		15	LC
<i>Graptemys pearlensis</i>	Emydi.	247.0				27.04			EN
<i>Graptemys pseudogeographica</i>	Emydi.	225.0	8	14.1	4	56.4		0.72	35.4
<i>Graptemys pulchra</i>	Emydi.	247.0	14	7.2	6	43.2		15.7	NT
<i>Graptemys versa</i>	Emydi.	150.3	7	5.6	4	22.4			LC
<i>Hardella thurjii</i>	Geoem.	610.0		16	1	16		18	VU
<i>Heosemys annandalii</i>	Geoem.	450.0		6		9		9.3	EN
<i>Heosemys depressa</i>	Geoem.	250.0		2	1	2			CR
<i>Heosemys grandis</i>	Geoem.	295.0		5	2	10		29.6	VU
<i>Heosemys spinosa</i>	Geoem.	186.0		1		1.5		22.9	EN
<i>Hydromedusa maximiliani</i>	Cheli.	142.7	14	2	1	2			VU
<i>Kinostemon acutum</i>	Kinos.	120.0	5	2	4	8			NT
<i>Kinostemon alamosae</i>	Kinos.	105.0	5	4	2	8			DD
<i>Kinostemon angustipons</i>	Kinos.	112.0		1.5		4.61			VU

<i>Kinostemon arizonense</i>	Kinos.	136.0	6	4.7	3	14.1		25	LC
<i>Kinostemon baurii</i>	Kinos.	91.6	6	2.55	6	15.3		49.6	LC
<i>Kinostemon chimalhuaca</i>	Kinos.	107.0	7	4	2	8			LC
<i>Kinostemon creaseri</i>	Kinos.	116.0	10	1	4	4			LC
<i>Kinostemon durni</i>	Kinos.	150.0		2	2	4			VU
<i>Kinostemon durangoense</i>	Kinos.	131.1				9.35			DD
<i>Kinostemon flavescens</i>	Kinos.	102.0	11	5.68	2	11.36		40	LC
<i>Kinostemon herrerae</i>	Kinos.	143.1		2	2	4		19.5	NT
<i>Kinostemon hirtipes</i>	Kinos.	107.4	5	3	4	12			LC
<i>Kinostemon integrum</i>	Kinos.	149.3		5	3	15		18.5	LC
<i>Kinostemon oxaccae</i>	Kinos.	115.0	7			9.35			DD
<i>Kinostemon sonoriense</i>	Kinos.	124.0	7	4.31	2	8.62	0.10	36.5	NT
<i>Kinostemon subrubrum</i>	Kinos.	96.3	4	3.25	4	13		40	LC
<i>Leucocephalon yuwonoi</i>	Geoem.	240.0	7	1	3	3			CR
<i>Lissemys punctata</i>	Tryon.	205.0		5.83	2	11.66		17.8	LC
<i>Lissemys scutata</i>	Tryon.	205.0				13.50			DD
<i>Macrochelys temminckii</i>	Chely.	400.0	15	24.5	1	24.5		70.3	VU
<i>Malaclemys terrapin</i>	Emydi.	150.0	6	6.7	2	13.4	0.30	50	NT
<i>Malayemys subtrijuga</i>	Geoem.	148.0		4	1	4		14.2	VU
<i>Mauremys annamensis</i>	Geoem.	285.0	7	2	2	4		23.9	CR
<i>Mauremys japonica</i>	Geoem.	150.0		6	3	18		16.2	NT
<i>Mauremys mutica</i>	Geoem.	130.0		4	2	8		22.1	EN
<i>Mauremys nigricans</i>	Geoem.	257.0		5.4	4	21.6		18	EN
<i>Mauremys reevesii</i>	Geoem.	195.0	9	6.7	3	20.1		24.2	EN
<i>Mauremys sinensis</i>	Geoem.	159.0	5	12.6	1	12.6		22.8	EN
<i>Melanochelys tricarinata</i>	Geoem.	150.0	8	3	1	3			VU
<i>Melanochelys trijuga</i>	Geoem.	175.0		3	2	6		40	NT
<i>Mesoclemmys dahli</i>	Cheli.	191.0		4		5.12		27.4	CR
<i>Mesoclemmys hogeii</i>	Cheli.	269.0		7		8.96			EN
<i>Mesoclemmys vanderhaegei</i>	Cheli.	150.0		6.4		8.19			NT
<i>Mesoclemmys zuliae</i>	Cheli.	263.0		7		8.96			VU
<i>Morenia ocellata</i>	Geoem.	230.0			1	10.50			VU
<i>Morenia petersi</i>	Geoem.	180.0		6	1	6			VU
<i>Notochelys platynota</i>	Geoem.	203.0				8.95		18.8	VU
<i>Peltocephalus dumerilianus</i>	Podoc.	324.0		10.7		14.98			VU
<i>Pelusios broadleyi</i>	Pelom.	216.5				15.16			VU
<i>Pelusios castanoides</i>	Pelom.	220.0	3	25		25			LC
<i>Pelusios rhodesianus</i>	Pelom.	230.0		12.5		12.5			LC
<i>Pelusios subniger</i>	Pelom.	195.0	5	8		8		47	LC

<i>Pelusios upembae</i>	Pelom.	221.0				15.16		DD
<i>Platysternon megacephalum</i>	Platy.	180.0	8	3	1	3	27	EN
<i>Podocnemis erythrocephala</i>	Podoc.	251.0		8.7	1	8.7	18	VU
<i>Podocnemis expansa</i>	Podoc.	660.0	7	91.5	1	91.5	25.3	LR/CD
<i>Podocnemis lewyana</i>	Podoc.	475.0		16.5		16.5		EN
<i>Podocnemis sextuberculata</i>	Podoc.	436.6	4	13	1	13		VU
<i>Podocnemis unifilis</i>	Podoc.	360.2	5	27.3	1	27.3		VU
<i>Pseudemydura umbrina</i>	Cheli.	125.0	11	4.5	1	4.5		CR
<i>Pseudemys alabamensis</i>	Emydi.	375.0		10.8	2	21.6		EN
<i>Pseudemys concinna</i>	Emydi.	400.0	13	17	4	68	44	LC
<i>Pseudemys gorzugi</i>	Emydi.	372.0		9		28.44		NT
<i>Pseudemys nelsoni</i>	Emydi.	298.0	7	14.3	6	85.8	26.1	LC
<i>Pseudemys peninsularis</i>	Emydi.	350.0	5	15	2	30	25	LC
<i>Pseudemys rubriventris</i>	Emydi.	304.0	11	17	2	34		NT
<i>Pseudemys texana</i>	Emydi.	300.0		8.5	3	25.5		LC
<i>Rafetus euphraticus</i>	Tryon.	630.0		32	1	32		EN
<i>Rafetus swinhoei</i>	Tryon.	630.0				59.25		CR
<i>Rheodytes leukops</i>	Cheli.	249.0		18.8		24.06		VU
<i>Rhinemys rufipes</i>	Cheli.	220.0	6	4		5.12		NT
<i>Rhinoclemmys annulata</i>	Geoem.	226.0		2		6		NT
<i>Rhinoclemmys areolata</i>	Geoem.	166.6		1	3	3	19.3	NT
<i>Rhinoclemmys funerea</i>	Geoem.	273.0		3.2		9.6		NT
<i>Rhinoclemmys nasuta</i>	Geoem.	218.0	14	1		3		NT
<i>Rhinoclemmys rubida</i>	Geoem.	220.9				9.97	11.8	NT
<i>Siebenrockiella crassicollis</i>	Geoem.	186.0		1.3	3	3.9	16.7	VU
<i>Siebenrockiella leytensis</i>	Geoem.	190.6		2	1	2		CR
<i>Staurotypus salvinii</i>	Staur.	180.0		5.36	6	32.16	26.3	NT
<i>Staurotypus triporcatus</i>	Staur.	285.0		10.61		63.66	33.5	NT
<i>Sternotherus carinatus</i>	Kinos.	116.5	4	4	3	12	29.3	LC
<i>Sternotherus depressus</i>	Kinos.	95.0	6	3	2	6	40	CR
<i>Sternotherus minor</i>	Kinos.	105.1	6	3.3	3	9.9	23.9	LC
<i>Sternotherus odoratus</i>	Kinos.	85.9	5	3.5	3	10.5	54.8	LC
<i>Terrapene carolina</i>	Emydi.	134.0	7	3.4	2	6.8	0.59 138	VU
<i>Terrapene coahuila</i>	Emydi.	101.6		2.3	3	6.9	18.8	EN
<i>Terrapene nelsoni</i>	Emydi.	134.0		2.7		5.4	24.9	DD
<i>Terrapene omata</i>	Emydi.	130.0	11	4.7	1	4.7	28.4	NT
<i>Trachemys adiutrix</i>	Emydi.	160.0		6		10.5		EN
<i>Trachemys decorata</i>	Emydi.	261.0		12		21		VU
<i>Trachemys gaigeae</i>	Emydi.	200.0		19	1	19		VU

<i>Trachemys ornata</i>	Emydi.	215.3	4	20		35			VU
<i>Trachemys scripta</i>	Emydi.	240.0	5	8.4	2.5	21	1.10	41.3	LC
<i>Trachemys stejnegeri</i>	Emydi.	215.3				21.74			NT
<i>Trachemys taylori</i>	Emydi.	215.3				21.74			EN
<i>Trachemys terrapen</i>	Emydi.	215.3				21.74			VU
<i>Trachemys yaquia</i>	Emydi.	215.3				21.74			VU
<i>Vijayachelys silvatica</i>	Geoem.	121.0		2	2	4			EN

Table A4: Parameters estimated for each species of turtle. Population growth rate in suitable habitats (r_1 : #/yr), population growth rate on roads (r_0 : #/yr, median = -0.02), maximum road density (RD_{mx}), the ratio of maximum to observed road density (RD_{mx}/RD_{ob}), dispersal variance (σ^2 ; km²/yr), minimal patch size (PA_{mm}), proportion of patches whose area is bigger than PA_{mm} ($P[PA_{ob}>PA_{mm}]$), P_{25} are the species within the 25th percentile for RD_{mx}/RD_{ob} and/or $P[PA_{ob}>PA_{mm}]$. We indicated the species status in the IUCN and those that are identified as threatened by roads.

Species	r_1	r_0	RD_{mx}	RD_{mx}/RD_{ob}	σ^2	PA_{mm}	$PA_{ob}>PA_{mm}$	P_{25}	IUCN	IUCN Road threat
<i>Acanthochelys macrocephala</i>	0.15		0.89	1421.55	0.20	13.78	24.04		NT	
<i>Acanthochelys pallidipectoris</i>	0.14		0.88	425.83	0.11	8.20	74.77		EN	
<i>Acanthochelys radiolata</i>	0.11		0.85	495.70	0.30	26.37	2.72		NT	
<i>Acanthochelys spixii</i>	0.13	0.001	0.96	574.38	0.09	7.47	6.55		NT	
<i>Actinemys marmorata</i>	0.11		0.85	222.04	0.02	2.05	8.66	+	VU	
<i>Amyda cartilaginea</i>	0.79		0.98	801.65	488.98	6084.66	0.17		VU	
<i>Apalone ferox</i>	0.25	-0.08	0.76	186.07	11.18	438.35	0.79	+	LC	+
<i>Apalone mutica</i>	0.19		0.91	282.93	0.50	26.21	7.73	+	LC	
<i>Apalone spinifera</i>	0.33	0.001	0.99	323.50	18.49	557.38	1.57	+	LC	+
<i>Batagur baska</i>	0.38		0.95	791.95	86.11	2230.40	0.07		CR	
<i>Batagur borneoensis</i>	0.30		0.94	708.28	68.07	2260.40	0.06		CR	
<i>Batagur dhongoka</i>	0.37		0.95	537.72	40.07	1073.94	1.76		EN	
<i>Batagur kachuga</i>	0.34		0.95	511.57	173.70	5082.77	0.22	+	CR	
<i>Batagur trivittata</i>	0.32		0.94	1578.09	82.14	2562.04	0.22	+	EN	
<i>Carettochelys insculpta</i>	0.21		0.91	5020.36	34.82	1653.22	0.72	+	VU	
<i>Chelodina mccordi</i>	0.34		0.95	199.43	1.38	40.01	6.25	+	CR	
<i>Chelodina novaeguineae</i>	0.34		0.95	56162.01	0.44	12.85	100		LC	
<i>Chelodina oblonga</i>	0.30		0.94	294.71	0.74	24.24	5.97	+	NT	
<i>Chelodina parkeri</i>	0.34		0.95	133775.72	0.86	25.37	100		VU	
<i>Chelodina pritchardi</i>	0.30		0.94	3829.47	0.75	24.63	50		EN	
<i>Chelodina reimanni</i>	0.21		0.91	12528.62	0.57	27.40	100		NT	

<i>Chelydra rossignonii</i>	0.29		0.94	699.17	8.06	277.28	0.86	+	VU	
<i>Chelydra serpentina</i>	0.24	-0.18	0.57	172.71	2.51	102.24	5.28	+	LC	+
<i>Chrysemys picta</i>	0.51		0.97	304.00	0.20	3.87	31.87		LC	+
<i>Claudius angustatus</i>	0.32		0.94	795.25	0.04	1.35	17.24		NT	
<i>Clemmys guttata</i>	0.28		0.95	164.02	0.007	0.26	40.30	+	EN	+
<i>Cuora amboinensis</i>	0.19		0.91	892.98	0.26	13.90	7.19		VU	
<i>Cuora aurocapitata</i>	0.22		0.92	475.58	0.34	15.59	5.93		CR	
<i>Cuora flavomarginata</i>	0.17		0.90	302.06	0.25	15.18	3.13		EN	
<i>Cuora galbinifrons</i>	0.11		0.84	916.96	0.42	39.68	0.86	+	CR	
<i>Cuora mccordi</i>	0.17		0.90	1316.01	0.10	6.01	23.07		CR	
<i>Cuora mouhotii</i>	0.19		0.91	1193.10	0.23	12.32	2.58		EN	
<i>Cuora pani</i>	0.22		0.92	1285.91	0.34	15.55	4.85		CR	
<i>Cuora trifasciata</i>	0.25		0.93	511.67	1.26	49.23	1.75	+	CR	
<i>Cuora yunnanensis</i>	0.23		0.92	354.00	0.36	15.96	2.81		CR	
<i>Cuora zhoui</i>	0.22		0.92	813.08	0.34	15.55	0.77	+	CR	
<i>Cyclemys dentata</i>	0.15		0.88	1224.67	1.34	89.40	1.84		NT	
<i>Dogania subplana</i>	0.28		0.94	1107.25	124.57	4343.51	0.12		LC	
<i>Eelseya branderhorstii</i>	0.19		0.91	149656.0	0.64	33.87	100		VU	
<i>Eelseya novaeguineae</i>	0.19		0.91	14867.72	0.64	33.87	17.14		LC	
<i>Elusor macrurus</i>	0.15		0.89	301.77	6.43	413.75	1.46	+	EN	
<i>Emydoidea blandingii</i>	0.19	-0.11	0.63	145.81	0.32	16.57	12.92		EN	+
<i>Emydura subglobosa</i>	0.20		0.91	14740.74	0.78	38.33	4.19		LC	
<i>Emys orbicularis</i>	0.15	0.001	0.99	220.70	0.04	2.91	17.08	+	NT	
<i>Erymnochelys madagascariensis</i>	0.16		0.89	7824.89	15.16	965.49	1.49	+	CR	
<i>Geoemyda japonica</i>	0.13		0.87	141.78	0.29	21.71	9.25		EN	
<i>Geoemyda spengleri</i>	0.11		0.85	660.24	0.03	2.93	6.11		EN	
<i>Glyptemys insculpta</i>	0.20		0.91	198.51	0.31	15.87	13.38	+	EN	+
<i>Glyptemys muhlenbergii</i>	0.18		0.90	127.80	0.01	0.52	40.65		CR	+
<i>Graptemys barbouri</i>	0.19		0.91	281.35	1.01	53.78	14.34	+	VU	
<i>Graptemys caglei</i>	0.16		0.89	246.88	1.88	118.37	7.45	+	EN	
<i>Graptemys ernsti</i>	0.11		0.85	304.03	1.36	122.84	5.82		NT	
<i>Graptemys flavimaculata</i>	0.09		0.82	359.52	1.17	134.83	8.96		VU	
<i>Graptemys geographica</i>	0.18	-0.06	0.77	166.56	0.35	19.08	8.19	+	LC	+
<i>Graptemys gibbonsi</i>	0.13		0.87	355.49	7.33	541.70	4.29		EN	
<i>Graptemys nigrinoda</i>	0.26		0.93	410.41	0.26	10.27	17.24		LC	
<i>Graptemys oculifera</i>	0.10		0.84	294.52	0.21	20.01	14.85	+	VU	
<i>Graptemys ouachitensis</i>	0.34		0.95	273.16	1.49	43.56	11.84	+	LC	
<i>Graptemys pearlensis</i>	0.23		0.92	302.66	2.96	124.90	7.93		EN	
<i>Graptemys pseudogeographica</i>	0.40		0.95	286.04	0.01	0.23	37.93	+	LC	

<i>Graptemys pulchra</i>	0.19	0.91	346.87	3.69	192.26	3.88	NT		
<i>Graptemys versa</i>	0.32	0.94	390.43	0.23	7.31	14.87	LC		
<i>Hardella thurjii</i>	0.25	0.93	607.44	323.84	12543.54	0.03	VU		
<i>Heosemys annandalii</i>	0.14	0.88	421.94	132.90	9053.34	0.14	EN		
<i>Heosemys depressa</i>	0.07	0.79	2840.96	3.80	504.52	12.5	CR		
<i>Heosemys grandis</i>	0.23	0.92	474.83	4.85	204.31	3.04	VU		
<i>Heosemys spinosa</i>	0.07	0.79	862.74	0.59	81.17	1.90	EN		
<i>Hydromedusa maximiliani</i>	0.07	0.77	267.52	0.10	16.28	2.89	+	VU	
<i>Kinostemon acutum</i>	0.30	0.94	803.70	0.05	1.74	13.30	NT		
<i>Kinostemon alamosae</i>	0.30	0.94	1128.10	0.02	0.68	15.70	DD		
<i>Kinostemon angustipons</i>	0.19	0.91	731.81	0.03	1.71	32.78	VU		
<i>Kinostemon arizonense</i>	0.32	0.94	1751.85	0.11	3.43	3.58	LC	+	
<i>Kinostemon baurii</i>	0.35	-0.32	0.52	124.18	0.01	0.21	40.46	LC	+
<i>Kinostemon chimalhuaca</i>	0.23	0.92	1172.13	0.02	1.01	15.55	LC		
<i>Kinostemon creaseri</i>	0.13	0.87	553.92	0.04	3.21	13.10	LC		
<i>Kinostemon dumni</i>	0.15	0.89	6879.58	0.23	15.22	5.55	VU		
<i>Kinostemon durangoense</i>	0.25	0.93	1143.04	0.09	3.78	5.99	DD		
<i>Kinostemon flavescens</i>	0.15	0.89	471.32	0.01	1.02	25.62	LC		
<i>Kinostemon herrerae</i>	0.15	0.89	659.84	0.18	11.75	5.16	NT	+	
<i>Kinostemon hirtipes</i>	0.36	0.95	591.41	0.02	0.67	11.44	LC		
<i>Kinostemon integrum</i>	0.29	0.94	680.21	0.24	8.33	5.50	LC		
<i>Kinostemon oaxacae</i>	0.25	0.93	943.44	0.03	1.56	16.35	DD	+	
<i>Kinostemon sonoriense</i>	0.24	0.93	746.81	0.001	0.007	73.50	NT		
<i>Kinostemon subrubrum</i>	0.44	0.95	242.88	0.01	0.26	41.71	+	LC	+
<i>Leucocephalon yuwonoi</i>	0.12	0.86	7516.10	2.71	221.61	0.28	+	CR	
<i>Lissemys punctata</i>	0.25	0.93	479.43	1.26	48.88	8.56	LC		
<i>Lissemys scutata</i>	0.27	0.93	1360.37	1.26	46.03	3.99	DD		
<i>Macrochelys temminckii</i>	0.17	0.89	288.19	9.64	574.73	2.10	+	VU	
<i>Malacllemys terrapin</i>	0.33	-0.29	0.54	83.21	0.001	0.03	44.07	NT	
<i>Malayemys subtrijuga</i>	0.12	0.86	413.40	0.30	25.51	15.64	VU		
<i>Mauremys annamensis</i>	0.16	0.89	1026.22	5.03	315.09	1.00	+	CR	
<i>Mauremys japonica</i>	0.28	0.94	93.34	0.28	9.82	6.18	NT		
<i>Mauremys mutica</i>	0.22	0.92	360.53	0.09	4.50	7.79	EN		
<i>Mauremys nigricans</i>	0.31	0.94	472.74	3.94	126.02	0.98	+	EN	
<i>Mauremys reevesii</i>	0.25	0.93	294.78	0.71	27.88	3.65	+	EN	
<i>Mauremys sinensis</i>	0.35	0.95	303.88	0.26	7.62	5.49	EN		
<i>Melanochelys tricarinata</i>	0.19	0.91	503.48	0.11	6.01	30.28	VU		
<i>Melanochelys trijuga</i>	0.27	0.93	451.36	0.25	9.19	14.23	NT		
<i>Mesoclemmys dahli</i>	0.13	0.87	630.51	0.57	41.9	5.44	CR		

<i>Mesoclemmys hogei</i>	0.17	0.90	323.97	3.27	188.65	1.38	+	CR		
<i>Mesoclemmys vanderhaegei</i>	0.17	0.89	728.42	0.16	9.95	12.41		NT		
<i>Mesoclemmys zuliae</i>	0.17	0.90	981.62	2.91	168.15	0.92	+	VU		
<i>Morenia ocellata</i>	0.21	0.92	1062.03	2.18	100.19	1.10	+	VU		
<i>Morenia petersi</i>	0.12	0.86	441.20	0.62	52.94	5.47		VU		
<i>Notochelys platynota</i>	0.15	0.89	1144.39	1.13	74.09	1.97		VU		
<i>Peltocephalus dumerilianus</i>	0.19	0.91	12277.83	11.01	582.71	0.29	+	VU		
<i>Pelusios broadleyi</i>	0.38	0.95	9965.04	0.63	16.25	100		VU		
<i>Pelusios castanoides</i>	0.60	0.97	1900.38	0.68	11.23	12.43		LC		
<i>Pelusios rhodesianus</i>	0.35	0.95	1217.90	0.85	24.26	8.84		LC		
<i>Pelusios subniger</i>	0.24	0.92	1566.89	0.37	15.36	14.81		LC		
<i>Pelusios upembae</i>	0.38	0.95	1630.30	0.70	18.05	14.89		DD		
<i>Platysternon megacephalum</i>	0.08	0.81	527.48	0.42	51.58	2.63		EN		
<i>Podocnemis erythrocephala</i>	0.20	0.91	17538.05	3.50	171.70	0.24		VU		
<i>Podocnemis expansa</i>	0.55	0.97	8197.37	344.28	6175.03	0.18	+	LC		
<i>Podocnemis lewyana</i>	0.29	0.94	1086.62	77.41	2611.83	0.21	+	CR		
<i>Podocnemis sextuberculata</i>	0.33	0.94	27678.73	50.34	1511.14	0.20	+	VU		
<i>Podocnemis unifilis</i>	0.27	0.93	6581.94	18.89	678.81	1.00	+	VU		
<i>Pseudemydura umbrina</i>	0.09	0.82	95.89	0.05	6.09	3.84		CR		
<i>Pseudemys alabamensis</i>	0.22	0.92	220.80	16.43	732.92	0.13		EN		
<i>Pseudemys concinna</i>	0.24	0.001	0.98	277.52	15.41	632.56	1.36	+	LC	+
<i>Pseudemys gorzugi</i>	0.24	0.93	796.65	15.77	636.38	0.96	+	NT		
<i>Pseudemys nelsoni</i>	0.43	-0.07	0.86	170.50	5.79	133.83	1.13	+	LC	+
<i>Pseudemys peninsularis</i>	0.41	0.95	178.25	13.73	333.12	0.46	+	LC	+	
<i>Pseudemys rubriventris</i>	0.22	0.92	111.37	5.63	255.98	0.29		NT		
<i>Pseudemys texana</i>	0.24	0.92	329.26	5.26	221.04	6.15		LC		
<i>Rafetus euphraticus</i>	0.32	0.94	868.92	285.77	8730.74	0.07		EN		
<i>Rafetus swinhoei</i>	0.40	0.95	314.37	285.77	7097.38	1.76		CR		
<i>Rheodytes leukops</i>	0.20	0.91	1478.24	1.87	90.49	8.07		VU		
<i>Rhinemys rufipes</i>	0.15	0.89	26873.76	0.99	64.21	0.91	+	NT		
<i>Rhinoclemmys annulata</i>	0.05	0.71	1805.75	2.52	520.53	1.63	+	NT		
<i>Rhinoclemmys areolata</i>	0.03	0.64	484.56	0.40	114.27	2.54		NT	+	
<i>Rhinoclemmys funerea</i>	0.07	0.78	2212.33	6.60	927.23	2.45		NT		
<i>Rhinoclemmys nasuta</i>	0.02	0.47	957.94	2.09	1206.15	0.28	+	NT		
<i>Rhinoclemmys rubida</i>	0.05	0.74	742.35	2.78	505.73	1.78		NT		
<i>Siebenrockiella crassicollis</i>	0.08	0.80	626.49	0.81	102.89	2.77		VU		
<i>Siebenrockiella leytensis</i>	0.04	0.68	1014.54	0.83	200.95	28.5		CR		
<i>Staurotypus salvinii</i>	0.51	0.96	685.00	0.43	8.54	4.73		NT		
<i>Staurotypus triporcatus</i>	0.66	0.97	817.78	3.59	53.88	2.21		NT		

<i>Sternotherus carinatus</i>	0.33	0.94	272.62	0.04	1.28	24.3	+	LC		
<i>Sternotherus depressus</i>	0.18	0.90	264.39	0.01	0.62	26.6	+	CR		
<i>Sternotherus minor</i>	0.21	0.92	270.06	0.03	1.45	32.9	+	LC	+	
<i>Sternotherus odoratus</i>	0.21	-0.01	0.97	220.32	0.004	0.22	47.5	+	LC	+
<i>Terrapene carolina</i>	0.10	-0.01	0.93	219.29	0.001	0.15	42.8	+	VU	+
<i>Terrapene coahuila</i>	0.11	0.85	1209.82	0.03	2.93	100		EN	+	
<i>Terrapene nelsoni</i>	0.11	0.85	2177.74	0.10	9.34	8.62		DD		
<i>Terrapene ornata</i>	0.09	0.009	0.99	450.01	0.07	8.61	15.6		NT	+
<i>Trachemys adiutrix</i>	0.29	0.94	931.51	0.15	5.20	2.24		EN		
<i>Trachemys decorata</i>	0.40	0.95	229.76	1.86	45.88	2.71	+	VU		
<i>Trachemys gaigeae</i>	0.38	0.95	544.71	0.47	12.32	3.02		VU		
<i>Trachemys ornata</i>	0.54	0.97	859.45	0.69	12.74	2.57		VU		
<i>Trachemys scripta</i>	0.37	-0.03	0.92	300.30	0.01	0.49	34.1		LC	+
<i>Trachemys stejnegeri</i>	0.41	0.95	230.53	0.69	16.92	6.08	+	NT		
<i>Trachemys taylori</i>	0.41	0.95	3372.83	0.69	16.92	5.88		EN	+	
<i>Trachemys terrapen</i>	0.41	0.95	383.37	0.69	19.92	3.54		VU		
<i>Trachemys yaquia</i>	0.41	0.95	1210.39	0.69	16.92	3.2		VU		
<i>Vijayachelys silvatica</i>	0.09	0.82	185.85	0.08	9.40	21.2	+	EN		

Table A5: Allometric relationship between the following variables with Carapace Length (mm). N = sample size.

Variable	Family	Estimate	R2	N
Mortality rate (μ)	Kinosternidae	0.00001 ^{1.69}	0.76	12
Eggs per year (b)	Chelidae	0.0007 ^{1.77}	0.37	19
Eggs per year (b)	Emydidade	0.003 ^{1.58}	0.58	36
Eggs per year (b)	Geoemydidae	0.007 ^{1.27}	0.31	43
Eggs per year (b)	Tryonichidae	0.010 ^{1.31}	0.50	17
Dispersal distance (σ^2)	-	3e-12 ^{5.09}	0.38	11